ANNUAL REPORT OF THE

BEAN IMPROVEMENT COOPERATIVE

BIC

A VOLUNTARY AND INFORMAL ORGANIZATION
TO EFFECT THE EXCHANGE OF INFORMATION AND MATERIALS

VOLUME 61
2018

THE LXI

Report of The

BEAN IMPROVEMENT COOPERATIVE

No. 61 April 2018

[ISSN 0084-7747]

Please address correspondence about BIC membership and BIC annual reports to:

Dr. Phillip N. Miklas
USDA-ARS
24106 No. Bunn Road
Prosser, WA 99350-9687
Phone: 509-786-9258
FAX: 509-786-9277
phil.miklas@ars.usda.gov

http://www.bic.uprm.edu
SITE REGULARLY UPDATED

Note: It will be assumed that interested individuals may freely cite (including author credit) any report or note in this BIC report, unless the author indicates to the contrary. As a professional courtesy, individuals citing BIC notes should notify the authors of his or her intentions. The BIC Coordinating Committee approved this statement on November 5, 1975.

TABLE OF CONTENTS

RESEARCH PAPERS FOR 2018

ORAL PRESENTATIONS (2017 BIC MEETING):

POSTER PRESENTATIONS (2017 BIC MEETING):

2018 RESEARCH ARTICLES:

GENETIC STOCKS AND RELEASE NOTES

Cover: Market in Chimaltenango, Guatemala (photo courtesy of Howard Schwartz)

THE 61st ANNUAL REPORT OF THE BEAN IMPROVEMENT COOPERATIVE

The Bean Improvement Cooperative (BIC) celebrated the twenty-ninth Biennial Meeting in East Lansing, Michigan, at the Kellogg Center housed on the beautiful campus of Michigan State University, which was a fantastic venue. This BIC meeting had 164 registrants including participants from Africa, Asia, Canada, Europe, Mexico, and South America which contributed to a diverse and electric atmosphere. The meeting began with the Frazier-Zaumeyer Distinguished Lectureship 'New Plant Phenotyping and Analytics Platforms for Improved Grain Legumes' which was presented by Dr. David Kramer, Hannah Distinguished Professor in Photosynthesis and Bioenergetics at Michigan State University. This lectureship kicked off the symposium '*Abiotic Stresses*' which highlighted innovations and advances for characterizing abiotic stress response and proposed strategies for breeding abiotic stress tolerant beans. The meeting ended Tuesday evening with the session 'International Diversity' with topics ranging from 'Bean Improvements in the East African Corridor' to 'Spatial and Temporal Scales of Range Expansion in wild *P. vulgaris*'. Overall there were 40 oral and 68 poster presentations.

The meeting received generous support from numerous donors – Platinum: College of Agricultural and Natural Resources – Michigan State University, Michigan Bean Commission; Silver: Monsanto, ProVita, Inc., Syngenta; Copper: Alberta Pulse Growers Association, American Pulse Association, Michigan Crop Improvement Association, Treasure Valley Seeds, USA Dry Pea & Lentil Council; Bronze: Crites Seed, Inc., and PureLine Seeds, Inc. On behalf of the BIC, I wish to acknowledge the substantial role of the organizing committee, Jim Kelly, Karen Cichy, Marty Chilvers, Greg Varner and staff, and would like to thank them, the sponsors and all the participants for making this meeting 'way cool'.

At the Awards Banquet, the Frazier-Zaumeyer Lecturer was recognized, and Technical Merit Awards were presented to Rian Lee (NDSU) and Evan Wright (MSU), Distinguished Achievement Awards were presented to Dr. Deidré Fourie (South Africa) and Dr. Clare Mukankusi (Uganda), and Meritorious Service Awards to Dr. Maria Celeste Gonçalves-Vidigal (Brazil), Mr. Greg Varner (Michigan Dry Bean Advisory Board), and Dr. Irv Widders (MSU). Four Student Travel Grants were awarded, two international to Luseko Chilange (Tanzania) and Michele Nay (Colombia), and two domestic to Isaac Fisher (Delaware State) and Cecilia Monclova (NDSU). Katelynn Walter (NDSU) received the Graduate Student Award for best poster presentation 'Pre-germination Flooding Tolerance of Middle American Dry Bean Genotypes' and Jennifer Wilker (Guelph) for best oral presentation 'Symbiotic Nitrogen Fixation in the MesoAmerican Genepool of Common Bean'. Congratulations to all the Awardees!

The BIC Coordinating Committee directed the President to i) establish a two-year membership renewal process whereby membership now costs $40 and is good for two years, ii) begin search for a new BIC President to hand the gavel to at the 2019 BIC Biennial Meeting, and iii) continue to increase review of technical reports for quality. Committee changes included Dr. Jim Kelly as new Chair of the BIC Genetics Committee replacing Dr. Kirstin Bett.

The next BIC Biennial Meeting is planned for Fargo, North Dakota in October/November 2019. The local organizing committee consists of Phil McClean, Juan Osorno, and Julie Pasche. As the 2019 BIC meeting approaches, details will be posted on the BIC Web page http://www.bic.uprm.edu. Please note the new web address.

Wishing you a successful year …….

Dr. Phillip Miklas, BIC President

BIC COMMITTEE MEMBERSHIP - 1957 to 2018

Coordinating Committee (approximate year of appointment):

1957 Dean, Enzie, **Frazier* (BIC Coordinator/President)**, McCabe, Zaumeyer
1960 Anderson, Atkin, Dean, Enzie, **Frazier**, McCabe, Zaumeyer
1962 Anderson, Atkin, Dean, **Frazier**, Pierce, Polzak, Zaumeyer
1968 Anderson, **Coyne**, Dean, Jorgensen, Polzak, Zaumeyer
1971 Briggs, **Coyne**, Dean, Jorgensen, Polzak, Zaumeyer
1972 Burke, **Coyne**, Dean, Jorgensen, Kiely, Polzak, Zaumeyer
1974 Ballantyne, Bravo, Burke, **Coyne**, Dickson, Emery, Evans, Kiely, Saettler, Zaumeyer
1977 Ballantyne, Bliss, Coyne, **Dickson**, Emery, Evans, Graham, Meiners, Morris, Saettler, Zaumeyer
1978 Atkin, Ballantyne, Bliss, Coyne, **Dickson**, Graham, Meiners, Morris, Saettler, Sprague
1979 Atkin, Bliss, **Dickson**, Graham, Hagedorn, Meiners, Morris, Sprague, Wallace
1980 Atkin, Bliss, **Dickson**, Hagedorn, Morris, Sprague, Steadman, Temple, Wallace
1982 Atkin, Coyne, **Dickson**, Hagedorn, Sprague, Steadman, Temple, Wallace, Wyatt
1983 Coyne, **Dickson**, Hagedorn, Saettler, Silbernagel, Steadman, Temple, Wallace, Wyatt
1985 Coyne, **Dickson**, Mok, Saettler, Silbernagel, Steadman, Temple, Wallace, Wyatt
1986 Coyne, **Dickson**, Mok, Saettler, Schoonhoven, Schwartz, Silbernagel, Steadman, Wallace
1988 Brick, Dickson, Emery, Magnuson, Roos, **Schwartz**, Singh, Steadman, Uebersax
1992 Dickson, Emery, Grafton, Magnuson, **Schwartz**, Singh, Stavely, Steadman, Uebersax
1994 Antonius, Dickson, Grafton, Magnuson, Park, **Schwartz**, Singh, Stavely, Uebersax
1996 Antonius, Grafton, Park, **Schwartz**, Singh, Stavely, Myers, Kotch, Miklas, Riley
1998 Antonius, Park, Schwartz (ex officio), Singh, Myers, Kotch, Miklas, Riley, Beaver, Vandenberg, **Kelly**
2000 Antonius, Beaver, **Kelly**, Kotch, Miklas, Myers, Park, Riley, Schwartz, Singh, Vandenberg
2001 ntonius, Beaver, **Kelly**, Kotch, Miklas, Myers, Park, Riley, de Ron, Schwartz, Vandenberg
2003 Beaver, **Kelly**, Kmiecik, Kurowski, Miklas, Myers, Park, Riley, de Ron, Schwartz, Vandenberg
2007 Beaver, **Kelly**, Kmiecik, Miklas, Myers, Park, Riley, de Ron, Schwartz , Shellenberger, Vandenberg
2008 Beaver, **Kelly**, Kmiecik, Miklas, Myers, Pauls, Riley, de Ron, Schwartz , Shellenberger, Vandenberg
2010 Beaver, Kelly, Kmiecik, **Miklas**, Myers, Pauls, Riley, de Ron, Schwartz , Shellenberger, Vandenberg
2011 Bett, Kelly, Kmiecik, **Miklas**, Myers, Osorno, Pastor-Corrales, Pauls, Riley, de Ron, Wahlquist
2016 Bett, Cichy, Kelly, Kmiecik, **Miklas**, Myers, Osorno, Pauls, Souza, Trapp, Wahlquist

Awards Committee:

1971 Baggett, Briggs, Burke, Dean, Wallace
1973 Burke, Dean, Mauth, Zaumeyer
1975 Ballantyne, Frazier, Mauth
1977 Ballantyne, Curme, Frazier, Schuster
1979 Ballantyne, Schuster, Silbernagel, Temple
1981 Abawi, Bliss, Monis, Silbernagel
1983 Adams, Bliss, Burke, Dean, Morris
1985 Emery, Hagedorn, Sandsted, Schwartz
1987 Emery, Hagedorn, Sandsted
1989 **Coyne**, Silbernagel, Wallace
1995 **Coyne**, Dickson, Stavely
1997 **Coyne**, Schwartz, Stavely
2001 **Hosfield**, Magnuson, Schwartz
2004 Hosfield, **Schwartz**, Singh
2008 Hosfield, **Schwartz**, Singh
2012 Noffsinger, **Schwartz**, Singh
2014 **Beaver**, Noffsinger, Urrea
2016 **Beaver**, Myers, Urrea

Genetics Committee

2004 **Bassett** (Chair), Beaver, Blair, Gepts, McClean, Miklas, Welsh (ex officio)
2005 **Beaver** (Acting Chair), Blair, Gepts, McClean, Miklas, Porch, Welsh (ex officio)
2007 Beaver, Blair, Gepts, McClean, Miklas, **Porch** (Chair), Welsh (ex officio)
2008 Bett, Blair, Gepts, McClean, Miklas, **Porch** (Chair), Urrea, Welsh (ex officio)
2014 **Bett** (Chair), Ferreira, Gepts, Goncalves-Vidigal, Kalavacharla, Kelly, Kisha (ex officio), McClean, Osorno, Porch, Urrea
2018 Bett, Ferreira, Gepts, Goncalves-Vidigal, Kalavacharla, **Kelly** (Chair), Kisha (ex officio), McClean, Osorno, Porch, Urrea

BIC GENETICS COMMITTEE MEETING MINUTES

During the 2017 BIC Biennial Meeting

Meeting location: Kellogg Center, Michigan State University, East Lansing, MI
Date: Nov 1, 2017 Time: 11:30 AM

Committee Members and Guests present:

Phil Miklas - BIC president and acting Chair

Thiago Souza
Jim Kelly
Paul Gepts
Ted Kisha
Maria Celeste
Carlos Urrea
Frédéric Marsolais
Pedro Vidigal-Filho
Bodo Raatz
James Beaver
Juan Osorno
Gonçalves-Vidigal
Talo Pastor-Corrales

1. Old Business:

Approval of the Genetics Committee meeting minutes at the Niagara Falls Marriott Gateway Hotel, Niagara Falls, Ontario, Nov. 4, 2015 (AIF)

Genotype ID ontology – (update from Bodo Raatz) developing a universal naming convention for lines and cultivars to facilitate combining data and reducing duplicates from data sets is recommended but difficult to enforce –this effort remains a work in progress outside the purview of the BIC Genetics Committee.

Trait symbol ontology– (update by Jim Kelly) placed a list on the BIC website for QTL names used in the literature (http://bic.css.msu.edu/_pdf/Published_Symbols_used_in_Naming_QTL_in_Phaseolus_vulgaris.pdf) Symbols (abbreviations) used for naming various traits lack consistency across publications which creates confusion (eg. gy, sy, yld have been used to name QTL for seed yield) making it difficult to integrate results among studies and to conduct meta-analysis across combined data sets. The BIC Genetics Committee recommends researchers use previously published symbols and to work towards developing consensus for naming the same trait with a common symbol. Efforts to review BIC reports for use of established symbols will be made.

Updated "List of Genes- *Phaseolus vulgaris*"
***Co-16* gene symbol** (Maria C. Gonçalves-Vidigal) received final approval as a gene but its proximity to *Co-3* and other *anthracnose resistance loci* mapped in the same proximal region of Pv04 will be noted in the gene list.

The descriptions for ***Co-17, Phg-1, Phg-2, Phg2[2], Phg-3, Phg-4, Phg-5, Pkp-1*** gene symbols approved in 2015 were fine tuned in the updated "Gene List 2017".

2. New Business

Gene symbols to review:
Co-Pa (temporary symbol used by Maria C. Gonçalves-Vidigal) maps to *Co-1* locus but allelism tests show independence (this contradiction needs to be resolved before a gene symbol can be approved). Discussion about the difficulty in conducting allelism tests for genes within clusters led to review and revision of the "List of Evidence" necessary for reviewing a symbol (underlined sections b. iii and iv below)

a. The committee will evaluate the data to determine:
    i. if sufficient evidence exists to establish the inheritance hypothesis
    ii. whether any issue of potential allelism of the trait has been met
    iii. whether the proposed gene symbol has been previously assigned to another gene.
b. The evidence must include:
    i. data from one generation to formulate an hypothesis
    ii. data from subsequent generations to test that hypothesis
    iii. <u>for hyper-variable pathogens: family mean testing (F2:3 progenies, or recombinant inbred lines – RILs), and use of multiple, specific races of the pathogen to separate effects of individual genes in gene clusters</u>
    iv. <u>molecular marker data and genetic linkage map and physical map (preferred) positions when available</u>

In addition it was recommended that a section 'c' be added to the "List of Evidence" which addresses availability of materials with the described genes so that others will be able to conduct allelism tests with the new genes in the future.

c. Lastly – parent, germplasm line, or cultivar source of new genes accepted by the committee must be made publically available via seed deposit with the U.S. National Plant Germplasm System - Plant Introduction Station in Pullman, WA, as a Genetic Stock (this enables others to access the gene source for subsequent allelism tests, genetic studies, etc.). This requirement is unnecessary for widely known and easily accessible materials.

These edits and addition of gene source to the "List of Evidence" will be circulated to absent committee members for their input by the next meeting (AIF).

The review of a new symbol for powdery mildew resistance (was postponed; and awaits submission of evidence in summary form from the publication submitted by Juan Jose Ferreira).

3. Next meeting: UC-Davis (with P. Gepts as local host) sometime in late July – early August 2018, in conjunction with the W-3150 and PCGC meetings (AIF).

4. Membership: Kirstin Bett rotated off as Chair and was replaced by Jim Kelly as the new Chair – Michigan State University (AIF).

AIF = all in favor

THE BEAN IMPROVEMENT COOPERATIVE

Proudly Presents the

Frazier - Zaumeyer Distinguished Lectureship

to

David M. Kramer
Hannah Distinguished Professor
Photosynthesis and Bioenergetics at Michigan State University

Meritorious Service Award

Maria Celeste Gonçalves-Vidigal
Univeristy of Maringa
Maringa, Brazil

Gregory V. Varner
Michigan Dry Edible Bean Production Research Advisor Board
Breckenridge MI

Irvin E. Widders
Michigan State University
East Lansing MI

Distinguished Achievement Award

Deidré Fourie
ARC Grain Crops Institute
Potchefstroom, South Africa

Clare Mukankusi Mugisha
CIAT – Uganda
Kampala, Uganda

Technical Merit Award

Rian Lee
North Dakota State University
Fargo, ND

Evan M. Wright
Michigan State University
East Lansing MI

in recognition of outstanding accomplishments relating to bean (Phaseolus) improvement

DAVID M. KRAMER

Dr. Dave Kramer is Hannah Distinguished Professor in Photosynthesis and Bioenergetics at Michigan State University (MSU). He holds appointments in the MSU-DOE PRL, which is supported by the U.S. Department of Energy; the MSU Department of Biochemistry and Molecular Biology in the College of Natural Science. Dr. Kramer received his B.S. in Biology and M.S. in Cell Biology from the University of Dayton and his Ph.D. in Biophysics from the University of Illinois at Urbana-Champaign. After a postdoc in Paris, France, and then rising through the ranks to professorship at the Institute of Biological Chemistry, Washington State University, he accepted the John A. Hannah professorship at MSU in 2010.

Kramer's research focuses on understanding how the machinery of photosynthesis is integrated into living organisms, which is critical for improvements in its efficiency and robustness needed to meet our future food and energy needs. Kramer's lab has made key contributions to this understanding for some time, but on joining the DOE-supported Plant Research Laboratory at MSU, he had an opportunity to approach the problem in a new way. This led to formation of the Center for Advanced Algal and Plant Phenotyping (CAAPP) and PhotosynQ, a unique team of over 40 scientists, engineers, computer programmers, computational biologists and even social scientists and economists to develop a series of novel scientific platforms that allow scientists to peer into living plants and algae and see photosynthesis at work under the harsh conditions where they grow.

Connecting to these scientific platforms, Kramer and his teams have developed for plant analyses the Environmental Photo Bio Reactor (ePBR) for algae, the Dynamic Environmental Photosynthetic Imaging (DEPI) system and most recently hand-held instruments such as the MultispeQ, With these ingenious devices and accompanying data analysis software, it is now possible to study plants and algae under conditions more closely simulating the natural environment. The focus and the goal have been on making these tools broadly available and thus enabling the larger scientific community to answer basic and applied questions about photosynthesis in novel ways. Although these tools are now being used in hundreds of research labs around the world, his team has been pushing to make these tools even more widely available, resulting in the establishment of both a spin-off company, Phenometrics, and the PhotosynQ.org project that aims to make sophisticated scientific tools accessible to broader communities to solve critical agricultural questions, especially in the developing world. These tools are unique and sophisticated. In particular, MultispeQs, which are quite inexpensive and easy to use, are being rapidly implemented by a number of bean and cowpea breeding programs and physiology field and laboratories in both the developed and the developing world (e.g., in Africa, Asia, Central and South America) with the potential for real progress in identifying traits and unique markers related to stress tolerance and productivity. Dr. Kramer is actively involved in Legume Innovation Lab projects in the U.S., Uganda and Zambia.

Dr. Kramer is the 2016 recipient of the International Society of Photosynthesis Research Innovation Award and he was also recognized in 2016 with the prestigious Charles F. Kettering award for excellence in Photosynthesis Research by the American Society of Plant Biologists. Kramer's research is supported by a number of agencies, including the U.S. Department of Energy, U.S. Aid for International Development, National Science Foundation, the McKnight Foundation and the John A. Hannah Foundation.

MARIA CELESTE GONÇALVES-VIDIGAL

Professor Maria Celeste Gonçalves-Vidigal was born in Solidão, Pernambuco, in the Northeast region of Brazil. She received her Bachelor's degree in Agronomy from the Federal Rural University of Pernambuco in 1974. She worked from1975 to 1980 as Research Assistant in the Agronomy Institute of Pernambuco State (IPA). While still working at IPA she earned her Master's degree in 1979 in Plant Breeding and Genetics from the Federal University of Viçosa (UFV), a top Brazilian Agricultural Research University. In 1981, she began her academic career as an Assistant Professor in the Agronomy Department of the Maringá State University (UEM), Brazil. After teaching during eight years at UEM, Dr. Gonçalves-Vidigal went back to UFV to pursue a Ph.D. degree in Plant Breeding and Genetics, which she completed in 1993, working with Dr. Clivas Vieira, a renowned Brazilian common bean Scientist. She then returned to UEM where a year later she was promoted to Full Professor of Plant Breeding and Genetics.

Dr. Gonçalves-Vidigal has advised 10 B.Sc., 33 M.Sc., and 21 Ph.D. students, and supervised 12 post-docs. She is currently supervising two Post-Docs, eight Ph.D. and two M.Sc. students. She has published 120 journal articles with many of them in international journals including Crop Science, TAG, BMC genomics. She has also registered four genomic sequences of *Colletotrichum lindemuthianum* at the National Center for Biotechnology Information of Brazil. Dr. Gonçalves-Vidigal has been a proactive leader at the UEM. In addition to co-founding the Nupagri Agricultural Sciences Laboratory in 1994, she played a major role in the creation of the graduate Programs in Agronomy (1995) and Plant Breeding and Genetics (2002), where she served as a chair from 1995 to 1999, and from 2004 to 2008. These are highly ranked programs at the Brazilian national level that have offered affordable and high quality education to over 900 Graduate students. The success of these programs is testimony of Dr. Gonçalves-Vidigal's leadership and vision. Among her numerous contributions to the Brazilian academic community, she co-founded the Brazilian Plant Breeding Society (SBMP) and the Crop Breeding and Applied Biotechnology journal. She is currently the president of the SBMP.

Eager to keep up to date with scientific technologies and to expand her knowledge, Dr. Gonçalves-Vidigal was one of the first scientists from Agronomy Department-UEM to spend a sabbatical in the US. In 2002, she spent a one-year sabbatical as Visiting Scholar at Michigan State University in the laboratory of Dr. James Kelly. Later in 2008, she spent another year as a Visiting Scientist in the laboratory of Paul Gepts at the University of California, Davis. She has also had many other international collaborations with scientists in the U.S. including Drs. M. Melotto, Q. Song and M. A. Pastor-Corrales. These international collaborations have included student exchange and publications. The international exchange greatly benefited UEM graduate students, which were given the opportunity of working in collaboration with her colleagues.

Dr. Gonçalves-Vidigal's research is often cited and she continues to grow scientifically; her research is now expanding to include genomics. She has identified, named and mapped several genes in common bean, especially genes conferring resistance to anthracnose but also genes for resistance to angular leaf spot and rust. Ten of the anthracnose resistance gene are of Andean origin while four are Mesoamerican. She has also developed molecular markers tagging these resistance genes. Moreover, she has also released common bean cultivars including the carioca cultivar 'Flor Diniz UEM' and the black cultivar 'Awauna UEM', and has developed ten high performance lines. The new cultivars have been registered with public Brazilian institutions and their use provide profits to UEM.

GREGORY V. VARNER

Mr. Greg Varner is the Research Director for the Michigan Dry Edible Bean Production Research Advisory Board, a position that he has held since 1980. Greg grew up on a small-certified seed production farm in southern Midland County, Michigan. He graduated from with a B.S. in AgriScience Education in 1974 and earned a Masters in Crop Science from MSU in 1976. He worked for the MSU Cooperative Extension Service Crops Agent in Gratiot County, Michigan from 1976-1980 before assuming his present position as Research Director of the Michigan Dry Bean Industry serving as the Dry Bean Agronomist for the State of Michigan. He has been an active member of the Bean Improvement Cooperative since 1981.

In his current role, Greg conducts statewide bean variety trials, fungicide and insecticide trials and serves as a vital resource for both growers and the Michigan bean industry on all aspects of bean production and management. He works closely with breeders and researchers at MSU in a wide array of extension and educational roles and has participated in the release of over 40 new dry bean varieties. Greg runs the statewide testing program that provides a vital service in the evaluation of new bean lines from MSU and new varieties from other public and private breeding programs in North America. He has been involved in wide array of projects, working with seed industry on seed related problems, insect control, annual fungicide trials for white mold control, to bean desiccation trials, and resulting effects on canning quality to the use of bean powder as an ingredient in future food products. His greatest legacy to the bean industry has been his role in overseeing the dramatic changes in how beans are grown and harvested in Michigan. He has seen changes in acreage, seed types, improved productivity, and a major change to direct harvest and the subsequent modifications in planting, rolling, row widths, weed control, and crop desiccation prior to harvest. He has also overseen changes in market classes grown in Michigan going from a predominant navy bean state to a leader in black bean and organic bean production. Overseeing these changes entailed conducting numerous extension activities, field days, tours, meetings, combined with research directed toward best farming practices. In addition to working with the bean industry in the state, Greg has close ties with members of the canning industry and annually invites representatives of those industries to Michigan to participate in the canning evaluation of new bean lines from across the country. He has been successful in securing commodity block grants through the Michigan Department of Agriculture to ensure funding for research needs set annually by the industry. Through PRAB, he provides funding for research programs at MSU.

Greg serves on MSU Bean Commodity Committee, on student advisory committees and is actively involved with all canning evaluations conducted on campus. He has served as a member of the board of directors on the National Sclerotinia Initiative since its founding in 2002. He plays a vital role on this board ensuring that funding for bean research is equally and fairly represented among the programs supported by this initiative across the country. He serves as the industry representative on the Technical Management Advisory Committee - TMAC of the Legume Innovation Lab. His knowledge and experience of bean production and the role of applied research provides a valuable contribution and balance on this committee where diverse disciplines and represented. In addition to his research activities, Greg currently serves on the Isabella Bank Corporate Board of Directors and is Chairman of the Board of Directors of the Isabella Bank regional banks in Gratiot, Saginaw, and Midland Counties. Greg lives with his wife Joan in Gratiot County and owns farms adjacent to the original family farm. His service to the Michigan bean industry and the broader bean community has been outstanding and truly deserving of this award.

IRVIN E. WIDDERS

Dr. Irv Widders is the director of the USAID-funded Feed the Future Legume Innovation Lab managed at Michigan State University (MSU). Dr. Widders grew up on a small truck farm in Eastern Pennsylvania and received a Bachelor of Science in horticulture from Penn State in 1975. He continued his education at the University of California, Davis, where he received an MS in vegetable crops in 1977 and a PhD in plant physiology in 1982. He joined the Department of Horticulture at MSU as an assistant professor in 1982 and was promoted to professor in 1996.

Dr. Widders was appointed deputy director of the Bean/Cowpea CRSP (Collaborative Research Support Program) in 1998 and assumed the role of director in 2000. The Bean/Cowpea CRSP was renamed the Dry Grain Pulses CRSP in 2007 and the Feed the Future Legume Innovation Lab in 2013. Dr. Widders has continued to serve as director throughout this period to the present. In this role, he has continued MSU's legacy of engaging science and scientific leadership to address the seemingly insurmountable worldwide problems of hunger and poverty. He has overseen the management of dozens of long-term projects focused on advancing sustainable and secure agricultural developments through science research, technology, international collaboration, and capacity building programs in Sub-Saharan Africa, Central America, the Caribbean, and the United States. Under his leadership, smallholder farmers' bean and cowpea crop yields have improved significantly and sustainably due to environmentally friendly and affordable advances in pest management to reduce crop loss, improved seed varieties able to thrive in changing climates, improved soil management practices, and widespread education on agriculture and nutrition. As a result, household food security and income has increased in these regions, improving the health and lives of families and communities throughout the world.

Dr. Widders received the Globie Award for International Leadership and Service from MSU in 2012, the Ralph H. Smuckler Award for Advancing International Studies and Programs at MSU in 2015, and the LIL Lifelong Achievement Award for Excellence in Grain Legume Research Award in Zambia in 2016. He has been a BIC member since 2003 and during that time has engaged many of our BIC colleagues in international activities. As director of LIL, he has planned and organized many international meetings to bring together US and International partners to further scientific collaborations. Chief among these conferences is the upcoming Grain Legume Research Conference in Ouagadougou, Burkina Faso, in August 2017, the Joint Pan-African Grain Legume and World Cowpea Conference in Livingston, Zambia, in 2016, the Dry Grain Pulses CRSP Global Meeting in Kigali, Rwanda, in 2012, and in Quito in 2010. In addition to his role as director of the Feed the Future Legume Innovation Lab, Dr. Widders served as the Lead PI of USAID's associate award directed at the rapid technology dissemination and commercialization of disease-resistant bean varieties in Guatemala, Nicaragua, Honduras, and Haiti; the director of the Bean Health Research Program; coordinator of study abroad in Peru and at the EARTH University in Costa Rica, and as a consultant for a World Bank project in Uruguay. Other associate awards include MASFRIJOL; Technoserve; Mwe Gen Pwa, a $2M USAID bean seed relief project in Haiti's Hurricane Matthew affected areas; and a Gates Foundation Project on integrated pest management for smallholder cowpea farmers in West Africa; all of these awards have thrived under his management. Dr. Widders's leadership and work have advanced innovative, research-driven outreach, engagement, and economic development activities that have improved—and continue to improve— the quality of life for the world's most vulnerable peoples, and in recognition for this, Dr. Widders is truly deserving of the BIC Meritorious Service Award.

DEIDRÉ FOURIE

Dr. Deidré Fourie is a Plant Pathologist, with the Agriculture Research Council, Grains Crop Institute (ARC-GCI) in Potchefstroom, Republic of South Africa, working on pathology and breeding of dry bean. Dr. Fourie graduated with B.Sc. and M.Sc. degrees in Microbiology from the Potchefstroom University for Christian Higher Education in 1988 and 1992, respectively. She was hired in 1991 by ARC-GCI as a "Researcher" to work as a plant pathologist in support of the dry bean and sunflower breeding programs and has been there since. Deidre's Master's studies concerned bacteriology which skills she brought to her new job specialty for bacterial diseases of common bean and other crops. Although, Deidre has many accomplishments in sunflower, her major contributions in bean pathology and breeding are highlighted here. Her first efforts were to characterize the virulence diversity (races) in South Africa of the bacterial pathogens that cause halo blight and bacterial brown spot diseases of common bean. As she gained experience in the field as a plant pathologist in support of breeding programs, she also began working toward her Ph.D. at the University of Pretoria, which she received in 2003. By the time, she completed her Ph.D. studies on 'Bacterial diseases of dry beans in South Africa", she started to gain recognition worldwide as a pathology expert on bacterial bean diseases, and accordingly was promoted by ARC-GCI to Senior Scientist in 2002. Between 1999 and 2002, she attended seven international conferences and co-hosted another "The 3rd Bean Rust and 2nd Common Blight International Workshop" which contributed to her exposure and stature as a dry bean plant pathologist. She attended her first BIC Biennial meeting in Fargo in 2001 and has attended most meetings since. In 2009, she became the Director of the Dry Bean Breeding program at ARC-GCI.

In her current role, Dr. Fourie is responsible for all activities of the national dry bean breeding program in South Africa including crossing, selections, advanced regional and national trials, disease and quality evaluations, and cultivar release. Since 2009, she has released eight cultivars including five sugar (cranberry). two navy, and one dark red kidney Her plant pathology research contributed to the mapping and tagging with molecular markers of all the major R genes conditioning resistance to the halo blight pathogen and to the discovery of the new gene *Pse-6*, and QTL HB4.2 and HB5.1 which condition resistance to Race 6. Her research showed that QTLs SU91 and BC420 for CBB resistance did not limit yield in the absence of disease, and interacted in an epistatic manner such that BC420 did not affect resistance in the absence of SU91. Dr. Fourie contributed significantly to the initial development, and continued increase and distribution of the Andean Diversity Panel. This immense effort enables impactful research to be conducted by many others. She supports dry bean research across the continent of Africa with her involvement in committees, working groups, and as longtime member of the CIAT-led Pan African Bean Research Alliance and Southern African Bean Research Network. The "Common Bean Disease Workshop on Angular Leaf Spot and Root Rots" hosted by Dr. Fourie at Kruger Gate in 2015 was a fantastic success attended by 65 bean scientists from 14 countries. During that conference, Dr. Fourie was awarded the 'Certificate of Merit' from the USDA-ARS Chief Administrator whom recognized how critical her research efforts where to the success of the ARS-Feed-the-Future bean project. Her importance to the South African dry bean industry is evidenced by 30 popular press articles, 13 pamphlets, and three extension bulletins, in addition to her eight recent cultivar releases. Dr. Fourie has published 28 peer reviewed journal articles, one book chapter, and 15 technical reports, 44 presentations at international workshop/conferences and 38 regional/national presentations. This documented research and her approachable expertise are valuable resources for the global bean research community.

CLARE MUKANKUSI MUGISHA

Dr. Clare Mukankusi Mugisha, from Mbarara district, SW Uganda, completed a BSc degree in Agriculture in 1998 at Makerere University, Kampala, with her dissertation focusing on disease and insect pest resistance of dual-purpose cowpea. She completed an MSc in Agriculture, Makerere University in 2000 for which she conducted participatory research with women farmers in Kumi district, Bukedea sub county (E. Uganda) on the management of Rosette and Cercospora leaf spot of groundnut. She then worked as Government Agricultural Officer in Kabale district (SW Uganda) for a short period before joining CIAT as a Research Assistant to the regional plant pathologist, Dr. Robin Buruchara. At CIAT, she worked with small-scale bean seed producers in E. Uganda focusing on the recognition and management of major pests and diseases of common beans and producing good quality seed.

In 2003, she successfully qualified for an African Centre for Crop Improvement PhD scholarship to study Plant Breeding at the University of KwaZulu-Natal, Republic of South Africa. Her PhD focused on improving resistance to Fusarium root rot of common bean. While conducting her PhD research, she continued serving CIAT as a Research Associate (2005-2008) at CIAT-Uganda. On completion of her PhD in 2008, she was awarded a postdoctoral fellowship and served as network breeder for PABRA until 2013, when she was promoted to the full scientist position of "Bean Breeder" in the East and Central Africa Bean Research network.

Clare's work initially centered on projects aimed at understanding and improving resistance to key bean diseases but has since progressed to include projects on bio-fortification for iron and zinc, abiotic stress, and consumer traits such as cooking time. Clare offers oversight on research conducted in these areas and develops breeding lines targeting seven market classes of beans. Because of her outstanding scientific expertise and excellent mentor qualities, Clare is sought out by many bean scientists through different projects, consortiums and communities of practice. Some of these include the African Bean Consortium (ABC) of the Kirkhouse Trust foundation, Demand Led Breeding (DLB) project of the Alliance for Agricultural R&D for Food Security (consisting of the Syngenta Foundation for Sustainable Agriculture, the Crawford Fund and the Australian Centre for International Agricultural Research-ACIAR), Tropical Legume Project (BMGF), HarvestPlus, CGIAR program on Climate Change, Agriculture and Food security (CCAFS), USDA's National Institute of Food and Agriculture, Biotechnology and Biological Sciences Research Council(BBSRC) projects on bean root rots, The Regional Universities Forum for Capacity Building in Agriculture (RUFORUM), and Alliance for a Green Revolution in Africa funded plant breeding capacity building program of Makerere University.

Clare maintains and distributes breeding lines developed by herself and the CIAT bean programs in Colombia and Malawi and germplasm from NARS and other partners. Clare is very keen on capacity building and has so far directly supervised three PhD students and 12 MSc students that completed successfully and advised many others**.** She has trained five interns at Diploma, BSc and MSc level in different areas of interest. She also helps train the NARs partners in disease phenotyping and breeding techniques. She acts as a direct supervisor of four research associates and six technical staff. In addition, she is the acting Country coordinator of the CIAT Uganda office comprised of 30 staff members. She has co-authored over 20 peer-reviewed publications and book chapters. In summary, Dr. Clare Mukankusi Mugisha has made significant contributions in her short career to the improvement of *Phaseolus* beans in Eastern Africa and has rapidly become an important collaborator and mentor to many African and overseas collaborators and students.

RIAN LEE

Mr. Rian Lee has been an instrumental member of the bean genetics and molecular genetics research group at North Dakota State University for the past 18 years since joining Dr. Phillip McClean's group at NDSU in 1999. Rian graduated from North Dakota State University with a BS in Biotechnology in 1993. He has recently taken advanced degree courses in the Genomics and Bioinformatics program at NDSU. After receiving his BS degree, Rian worked as a research molecular genetics technician from 1993 to 1999 with Biogenetic Services Inc. and Mycogen.

After joining Dr. McClean's group, he quickly became a leading mentor to undergraduate students, and valued colleague to other graduate students, research technicians, and postdoctoral scientists at NDSU and elsewhere. He supported not only bean researchers, but researchers working on all crops at NDSU. He provided valuable technical skills and leadership for the BeanCAP project and Phaseolus genome sequence project. This included providing the expertise necessary to develop the two-enzyme genotyping-by-sequencing methodology widely used in the bean research community. The Middle American Diversity panel GBS libraries were a critical components of the GWAS output from the BeanCAP project. Rian provided all of the DNA and RNA source materials used for the genome sequencing project. Rian has contributed to numerous research projects over the years, and has worked with bean breeders and geneticists, molecular geneticists, and genomicists gaining experience and contributing to a range of projects that has helped in basic as well as applied research in common bean.

A major contribution was the discovery of the *Crg* locus for *Ur-3*-mediated rust resistance along with Venu (Kal) Kalavacharla, and Phil McClean. Rian has also technically led projects on white mold and multiple disease resistance loci. Recently, Rian has provided bioinformatics support for common bean by developing comprehensive data resources that detail: 1) the suite of gene models in common bean; 2) the Pfam families for all gene models; 3) the Arabidopsis orthologs for the bean gene models; and 4) the Glycine orthologs for the bean gene models. Rian was the first to assemble bean EST sequences into CDS sequences that formed the basis for gene calling during the reference genome sequencing project. Those assembles were also found in the first bean databases developed by Phytozome and the Legume Information System. Because of these significant research contributions, Rian has been a co-author on 20 manuscripts not only from Dr. McClean's group, but other research groups from around the world. Rian is truly a very skilled research with a patient personality that allows him to take the time to carefully work with others while generating valuable scientific information that has supported bean research worldwide.

EVAN M. WRIGHT

Mr. Evan Wright is a research technician working in the dry bean breeding and genetics program at Michigan State University. He grew up on a cash crop farm near Muncie, Indiana where he acquired a strong agricultural background. As an undergraduate student at Central Michigan University, Evan was actively involved in biological research. Using transmission electron microscopy, Evan worked with Dr. Daniel Wujek on freshwater algal taxonomy. His work with the Wujek lab resulted in four publications, including a paper describing a species new to science, *Mallomonas weei*. Evan graduated from CMU with a bachelor's of science degree in Plant Biology in 2004 and briefly worked on resistance to soybean cyst nematode with Dr. Brian Diers at the University of Illinois before returning to the state of Michigan to continue his education. At Michigan State University, Evan joined Dr. Jim Kelly's dry bean breeding & genetics lab in 2005, where he conducted research on quality traits of black beans. By developing a population of black beans and screening it with molecular markers, he was able to map several QTL governing yield and color retention. While working on his degree, Evan gained valuable experience in all aspects of dry bean breeding, including crossing, planting, selecting, and harvesting. After earning a master's of science degree in Crop and Soil Sciences in 2008, Evan continued to work in the dry bean-breeding program as a part-time technician. When the previous technician retired, Evan began his current position as MSU's dry bean breeding program's full-time technician, although he also performs all technician duties for the USDA's bean breeding program as their *de facto* technician.

Evan exemplifies all the positive attributes of a dedicated technician ensuring that all aspects of the breeding program are running smoothly by anticipating and rectifying potential problems before they arise. He works amiably with members of the university staff, colleagues, and industry representatives, and he actively assists and advises students with their research needs. During field season, Evan's efforts in preparing seed, coordinating trials, planting fields, controlling weeds, taking notes, selecting, and harvesting are invaluable. He is extremely familiar with the crop, which allows him to make selections on advanced breeding lines in Michigan and across the country in addition to scouting seed fields for off-types. He provides invaluable advice on parental selections, advancing lines and using his computer skills to streamline and classify record keeping. In addition to managing field operations for both programs, he also assists both programs with canning quality evaluation, an extremely time- and labor-intensive endeavor that involves the cleaning, weighing, canning, and scoring of hundreds of genotypes. Furthermore, Evan makes crosses for all market classes, set up field books and nurseries, performs statistical analysis for all field and canning data, maintains and transports equipment across the state, and streamlines all aspects of the program when possible.

Evan has been involved in the development of many dry bean varieties coming from the MSU program including Bellagio, Rosetta, Eldorado, Snowdon, Powderhorn, Alpena, Zenith, Desert Song, Gypsy Rose, and Samurai varieties. He has received a number of awards including the G.O. Mott Meritorious Graduate Student Award, Crop Science Society of America, in 2007, the Jonathon Baldwin Turner Fellowship-University of Illinois in 2004 and the Academic Excellence Award-College of Science and Technology Residential College, from Central Michigan University in 2003. He is a member of the Bean Improvement Cooperative. Evan has published a number of refereed journal articles from both his BS and MS research, numerous variety registrations, and additional publications with graduate students he assisted as a technician. He is most deserving of the BIC technical merit award.

IN MEMORY OF MICHAEL HUGH DICKSON

Dr. Michael Hugh Dickson passed away on March 28th, 2018. Mike was born in London, England on April 2, 1932. He was the son of Dr. Hugh and Eranee Dickson and was from a long line of rose breeders. He spent his first three years of life in Egypt where his father was a plant scientist working on King Tutt's tomb. He grew up in turbulent times in England during World War II and graduated from Charterhouse School. In 1950 he left England to complete his B.S. degree at McGill University (MacDonald college) and then his M.S. and Ph.D. degrees in Plant Breeding at Michigan State University. He was a professor at Ontario Agricultural University in Guelph, Ontario for six years before moving to the New York State Agricultural Experiment Station (NYSAES) at Cornell University in Geneva NY as a professor in 1964. He established a world class breeding program in common beans and crucifers, resulting in many scientific papers, awards and mentoring of graduate students. He was the President of the Bean Improvement Cooperative (BIC) from 1977 to 1986, and was an active member of the bean research community throughout his career. He received the Meritorious Service Award from the BIC in 1987, and was elected Fellow of the American Society for Horticultural Science before retiring in 1995.

Mike was widely respected and well known for doing cooperative, multi-disciplinary research leading to the development of disease and insect resistance in several crops, including common beans. His work included the development of beans with high levels of resistance to root rots and white mold, and heat and cold tolerance. His recurrent snap bean breeding lines with white mold resistance have been widely dispersed. Mike was also well known for developing new techniques to support breeding efforts including approaches to test for bean seed-coat shattering and the straw test to evaluate plants for white mold resistance in beans. The straw test method, published as a two-page BIC report in 1996, has more than 100 citations, and is still used worldwide. He developed 'persistent white' cauliflower that allows curds to remain white in direct sunlight without self-wrapping leaves or being tied. He also developed and advanced the orange cauliflower. He developed cabbage breeding lines with glossy leaves which reduce damage from the diamondback moth, selected materials resistant to soft rot with Dr. Jianping Ren and developed broccoli with tolerance to high temperatures. Among his most influential efforts was the development of cabbage with resistance to black rot, the world's most damaging disease of *Brassica* vegetables. These materials have been utilized by seed companies worldwide and have made significant contributions to yield stability of cabbage and food security.

Mike married his college sweetheart, Jean Hamilton, in 1958. They would have celebrated their 60th wedding anniversary in August. Over the years they entertained many guests from around the world. In their retirement, he and Jean travelled the world, spending fifteen winters in their Tucson, Arizona home and visiting with their children and families. Mike stopped by the W1150 Regional Project meeting held in Tucson in 2011 and received a standing ovation from the participants which attested to the respect for him by the bean community. Mike had many hobbies including gardening, sailing, skiing, reading and painting in oil and watercolors. He was a long-term member of the Presbyterian Church in Geneva, a former board member of the Geneva Public Library, Commodore of the Seneca Yacht Club and a member of several other Geneva organizations. He is survived by his beloved wife Jean; his daughters Nancy, Jane and Roslyn; and his grandchildren Mario, Isabel, Blake and Drew.

IN MEMORY OF SILVIO HUGO OROZCO SARRIA

The BIC community received with sadness the news of the death of Dr. Silvio Hugo Orozco Sarria on January 27, 2018 in Guatemala City. Dr. Orozco was a breeder of the CIAT Bean Program for more than 15 years, where he demonstrated a great capacity to lead work teams in which he proved to be positive, kind and generous with his knowledge of beans. He always stood out for his humanity and high research quality, which led him to become one of the most outstanding bean breeders in Colombia and Latin America. He worked on the development of several of the best bean varieties in the world such as Diacol Calima, Diacol Nima, ICA Pijao, ICA Bunsi, among others.

Dr. Orozco served as National Director of the Grain Legume Program at the Colombian Agricultural Institute (ICA) from 1972 to 1974. Subsequently, he was an expert in Tropical Agriculture of FAO in Honduras from September 1974 to May 1977. In 1977, he joined the CIAT Bean Program as a plant breeder and developed numerous contributions to bean science in Guatemala. He was also an agronomist for the regional bean program "PROFRIJOL" of Central America and the Caribbean from 1981 to 1990 in Guatemala. In 1991, he served as Coordinator of the Regional Bean Project for Central America, until his retirement on December 31, 1992.

In 1993, the Department of Agricultural Research of the Ministry of Natural Resources of Honduras released a new variety of beans at the national level under the name of "Don Silvio", in honor of his distinguished research efforts and exceptional breeding lines. "Don Silvio" was derived from the improved line DOR482, which was released as one of the first varieties with a high level of resistance to the *Bean golden yellow mosaic virus*, that even under strong virus pressure maintained high yield. This line was early maturing, and was also tolerant to common bacterial blight and had a red seed coat coloration that was widely accepted by both producers and consumers.

Dr. Silvio Hugo's breeding work and good nature projected CIAT favorably in Central American. He toured the region in his pickup truck, sharing his time and experience with colleagues in all countries, both in the field and in moments of rest, and leaving tangible results in the varieties he helped create. His cheerful and optimistic personality left fond memories for everyone. So important was his contribution in the varieties he developed that today, the Calima variety became the standard and gave its name for an entire commercial class of grain worldwide.

Dr. Orozco Sarria was the author or co-author of more than 100 international publications and book chapters and received numerous awards in Latin America for research and services provided to the international agricultural research and development community. He will be remembered for his enormous professional achievements, which advanced the knowledge of bean breeding from its early stages to maturity, paving the way for important contributions to better livelihoods for bean farmers around the world.

IN MEMORY OF MILDRED ZAPATA

Mildred Zapata Serrano, Professor of Plant Pathology in the Department of Agroenvironmental Sciences at the University of Puerto Rico, Mayaguez Campus passed away on 13 February 2018. She is survived by her brother Ricardo, sister Maria, niece Sara and nephew Ricardo.

Dr. Zapata was born in Mayagüez, Puerto Rico on April 7, 1952. She obtained a B.S. degree in Biology in 1974 and an M.S. degree in Crop Protection in 1982 from the University of Puerto Rico, Mayaguez Campus. Mildred earned a Ph.D. degree in Plant Pathology at the University of Nebraska under the supervision of Dr. Anne Vidaver. Her Ph.D. dissertation dealt with common bacterial blight resistance of tepary beans.

Dr. Zapata began work with common beans in the 1970's as a technician in the laboratory of Dr. Nader Vakili at the USDA-ARS Tropical Agricultural Research Station (TARS) in Mayaguez, Puerto Rico. She conducted research to improve greenhouse and field screening techniques for evaluating beans for resistance to common bacterial blight. Dr. Zapata collaborated with Dr. George Freytag, former USDA/ARS Research Geneticist at TARS and Dr. Robert Wilkinson, Professor at Cornell University, in the development of bean lines with common bacterial blight resistance derived from scarlet runner beans (*P. coccineus*). This led to the release of five bean germplasm lines that have high levels of resistance to common bacterial blight. One of these lines was used as a progenitor of 'Verano' which is currently the most popular white bean in Puerto Rico. Dr. Zapata also collaborated with Dr. Freytag and Dr. Mark Bassett in the development and release of the common bacterial blight interspecific breeding line XR 235-1.

Dr. Zapata was active in the PROFRIJOL project in Central America and the Caribbean (CA/C). This provided an opportunity to make a collection of strains of the common bacterial blight pathogen from several different countries in the CA/C. A differential response was observed when some sources of common bacterial blight resistance were screened with different strains of *Xap.* This provided evidence that physiological races of the common bacterial pathogen may exist. In February 1996, the University of Puerto Rico hosted the First International Workshop on Common Bacterial Blight. Scientists from the Americas, Europe and Africa, with expertise in working with common bacterial blight of bean, gathered to discuss the state of the art of research dealing with this important disease. Following discussions at the Workshop, Dr. Dermot Coyne proposed minimum standards to designate races of *Xap*. Dr. Zapata proposed the use of a common set of bean differentials and standard inoculation and evaluation methods to produce uniform and reproducible data from the greenhouse screening of common bean leaves to *Xap*. In 2010, Dr. Zapata and collaborators reported that a single dominant gene, *Xap-1*, that conferred resistance to *Xap* strain 3353 had been identified. The SCAR SAP6 marker, located on B10, was found to co-segregate with the resistant phenotype. In recent years, Dr. Zapata has participated in the development and release of several breeding lines and cultivars that have enhanced levels of resistance to common bacterial blight.

In addition to beans, Dr. Zapata researched bacterial diseases of coffee and cocoyams. She taught a phytobacteriology course at the UPRM and trained numerous graduate and undergraduate students. She was an active researcher until her untimely death.

EFFECTS OF HIGH NIGHT TEMPERATURE STRESS ON REPRODUCTIVE STRUCTURES OF LIMA BEAN (*Phaseolus lunatus)*

Ernest EG*, Wisser RJ, and Johnson GC

Department of Plant and Soil Sciences, University of Delaware (emmalea@udel.edu)

INTRODUCTION

Heat stress reduces yields of May and early June-planted lima bean (*Phaseolus lunatus*) in the Mid-Atlantic Region of the U.S. High night temperatures during flowering and seed development can reduce or delay pod set, resulting in delayed harvest, lower yield and split pod sets. Breeding heat tolerant baby and Fordhook type lima beans is one goal of the University of Delaware lima bean breeding program. Past experiments showed that genotypes which shed higher amounts of pollen onto the style under heat stress produced higher yield under heat stress. Additional experiments were undertaken to characterize additional effects of high night temperatures on lima bean reproductive structures.

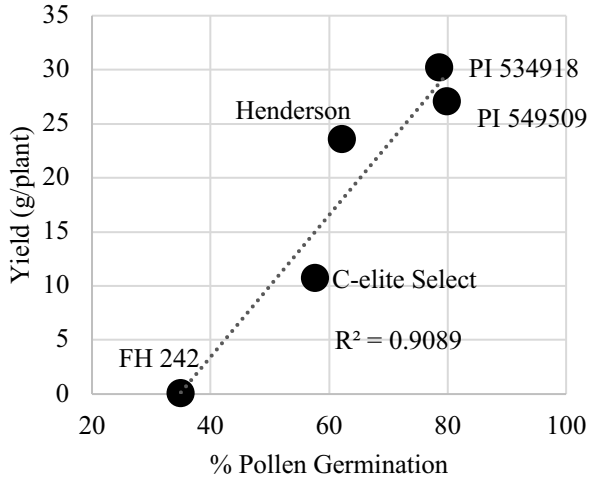
MATERIALS AND METHODS: Greenhouse experiments were used to characterize the response of lima bean genotypes to high versus ideal night temperatures in order to better understand the mechanism by which high night temperatures reduce yield. Lima bean genotypes with different responses to heat stress were grown in two climate controlled chambers inside of a greenhouse under hot and cool night temperature regimes. Each experiment was arranged in a randomized complete block design with 5 replications of each genotype. Target night temperatures were 27°C in the hot chamber and 18°C in the cool chamber. Target daytime temperatures for both chambers were 32-35°C. Pollen from the style of newly opened flowers was germinated by the sitting drop method in pollen germination medium containing 400 mg/l H_3BO_3, 600 mg/l $CaNO_3$, 400 mg/l $MgSO_4$, 400 mg/l KNO_3 and 40% sucrose (1). Pollen was incubated in the pollen germination medium for 4-5 hours at 25°C, then stained with acetocarmine and viewed at 100x. Germinated (pollen tube exceeds the pollen grain diameter) and ungerminated pollen grains were counted. For flower morphology experiments, newly opened flowers were dissected and viewed at 30x. To test processes downstream of pollination, hand pollinations were made on heat sensitive genotypes grown in the hot chamber with pollen collected from plants grown under ideal conditions. Selfed flowers at the same developmental stage and on the same raceme as the hand pollinations were marked as controls. Yield components (number of pods, number of seeds per pod, total number of seeds, total weight of seeds) and dry shoot weight were measured for each plant.

RESULTS

Heat Stress Effects on Yield Components: In heat sensitive genotypes there were significantly fewer pods per plant and significantly lower weight of seeds per plant for plants grown under high night temperature conditions than those grown under ideal temperature conditions. For all genotypes (heat sensitive and heat tolerant) there were significantly fewer seeds per pod for plants grown under high night temperature conditions than those grown under ideal temperature conditions. For three genotypes, including the most heat sensitive ones, dry weight of leaves and stems was significantly higher under high night temperature conditions than under ideal temperature conditions. When total above-ground dry biomass (leaves, stems, seeds and pods) was compared there were no differences between plants grown in high versus ideal night temperature conditions.

Pollen Germination: Genotype averages for in vitro pollen germination for unstressed plants ranged from 80 to 93%. For heat stressed plants genotype averages ranged from 35 to 80%. For some genotypes, in vitro germination of pollen collected from heat stressed plants was significantly lower than of pollen collected from unstressed plants. The rate of in vitro pollen germination from heat stressed genotypes was correlated to that genotype's yield under heat stress (Figure 1).

Figure 1. Heat Stress Yield vs Heat Stress Pollen Germination



Flower Morphology: In three of four genotypes tested (Fordhook 242, G27525, PI 549509) heat stress did not significantly affect the number ovules present at anthesis. In one genotype (C-elite Select) there was a significantly lower number of ovules in flowers produced on heat stressed plants.

In one very heat sensitive genotype, Fordhook 242, 88% of flowers produced by heat stressed plants had anomalous morphology which may interfere with pollination and fertilization such as anthers or stigma protruding from the keel or a hooked or bent stigma. There were significantly more deformed flowers in heat stressed plants than those grown under ideal conditions for this genotype (13%).

Processes Downstream of Pollination: When flowers on heat stressed C-elite Select (a heat sensitive genotype) plants were hand pollinated with pollen from unstressed PI 534918, 50% of the crosses produced a pod and 40% of the crosses produced mature seed. A significantly lower percent of the tagged selfed flowers produced a pod (5%) or produced mature seed (3%). In a parallel experiment with Fordhook 242 as the female parent 40% of crosses with PI 534918 pollen produced a pod and 10% produced mature seed. None of the tagged, selfed Fordhook 242 flowers produced mature pods or seed.

CONCLUSIONS: High nighttime temperatures reduce yield in some lima bean genotypes by reducing the number of pods set and the number of seeds per pod. High night temperatures do not reduce overall aboveground biomass, indicating that this type of temperature stress may have little effect on photosynthate availability. High night temperature has a significant effect on pollen production, release and viability, and in some genotypes results in changes to flower morphology that could interfere with pollination and fertilization. Hand pollination of a moderately heat sensitive genotype with highly viable pollen resulted in fertilization and seed set, but was not as successful in a very heat sensitive genotype, indicating that in very heat sensitive genotypes, pollen quantity and viability may not be the only limiting factor in yield under heat stress.

REFERENCE

(1) Gurusamy, V., Vandenberg, A. and Bett, K. E. (2007), Manipulation of in vivo pollination techniques to improve the fertilization efficiency of interspecies crosses in the genus Phaseolus. Plant Breeding, 126: 120–124. doi:10.1111/j.1439-0523.2007.01337.x

A REPEATABLE PROTOCOL FOR FUSARIUM ROOT ROT PHENOTYPING OF COMMON BEAN

Zitnick-Anderson K[1], Modderman C[1], Hanson LE[2], and Pasche JS[1]*

[1]North Dakota State University, Department of Plant Pathology *(Julie.Pasche@NDSU.edu)
[2]USDA-ARS, East Lansing, MI

INTRODUCTION: Pathogens within the *Fusarium solani* species complex (FSSC) cause root rot on common bean worldwide. Infection of root tissue by these pathogens results in damage to the vascular system, reducing seed yield. While integrating cultural management practices including crop rotation, planting high-quality seed and cultivation can reduce severity of Fusarium root rot, the most effective method of managing the disease is incorporating host resistance. A crucial step in developing resistant varieties is a repeatable protocol to screen for pathogen resistance under controlled conditions. However, phenotyping for Fusarium root rot can be hampered by a lack of adequate disease pressure and inconsistent results.

METHODS: A protocol was developed utilizing a watering regime that allowed for the soil matric potential to decrease until just above wilting point before soil was saturated to field capacity. Temperature (25 ± 2°C) and relative humidity (≥90%) were tightly regulated in a walk-in growth room throughout the experiment. Five genotypes were evaluated using five plants per replicate and three replicates per trial. The inoculation experiment was performed four times. Nine FSSC isolates pathogenic on common bean and confirmed via amplification of the *translation elongation factor 1α* (*EF-1α*) gene (O'Donnell et al. 1998) were utilized for inoculation. When the hypocotyl arch broke the soil surface, soil was saturated to field capacity and five mL of a 1×10^6 macroconidia/mL suspension was dispensed directly to the base of each plant within 30 minutes of watering. Plants were rated for Fusarium root rot severity (RRS) 14 days after inoculation on a 1 to 9 scale, where 1 represents no disease (Van Schoonhoven and Pastor-Corrales 1987.). The Andean (274 lines) and Middle-American (289 lines) Diversity Panels were evaluated in replicated trials using the procedure described for the five control lines.

RESULTS: Results indicate the RRS from each of five genotypes evaluated over 4 trials could be combined based on Levene's test for homogeneity of variance ($\alpha = 0.05$). Across the four trials, a consistent rank from susceptible to resistant was observed for three of five control genotypes. VAX3 was consistently the most resistant genotype (Figure 1). Talon and Dynasty interchanged rankings across trials, but did not display significantly different RRS in any of the trials. These two genotypes were considered intermediate based on mean RRS across the four trials. Cabernet was more susceptible than these two cultivars, and Montcalm was consistently most susceptible to Fusarium root rot.

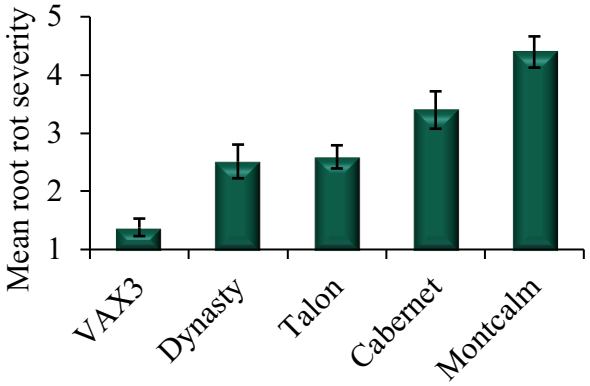


Figure 1. Mean root rot severity (1 to 9) for 5 control lines inoculated with *Fusarium solani* across four greenhouse experiments.

Mean RRS in the ADP and MDP ranged from 1.0 to 5.5 and 1.0 to 4.8, respectively, across two trials for each panel (data not shown). Eighteen and 10 MDP and ADP genotypes, respectively, were equally or more resistant to Fusarium root rot than the resistant control, VAX3, under greenhouse conditions (Table 1). Analyses to identify genetic regions associated with the resistance observed here are currently underway.

Table 1. Identification number (ID), genotype, and seed color for lines in the Middle-American (MDP and Andean Diversity Panels (ADP) displaying root rot severity (RRS; 1 to 9 scale) equal to or less than the resistant control line VAX3 under greenhouse conditions.

ID	Genotype	RRS		ID	Genotype	RRS
MDP 126	Loreto	1.0		ADP 511	Canario_1	1.0
MDP 140	Ember	1.0		ADP 514	Mantegaamarela	1.1
MDP 186	GN Harris	1.0		ADP 4	Kilombero	1.2
MDP 216	I9365-31	1.1		ADP 94	Lushala	1.3
MDP 349	Harrowhawk	1.1		ADP 513	Canario_2	1.3
MDP 9	AC Redbond	1.2		ADP 1	Rozi_koko	1.4
MDP 60	Swan Valley	1.2		ADP 2	W6_16444	1.4
MDP 131	Pink Floyd	1.2		ADP 111	Uyole98	1.4
MDP 403	McHale	1.2		ADP 214	G5087	1.4
MDP 87	Condor	1.3		ADP 429	Pr9920_171	1.4
MDP 96	Cornell 49-242	1.3			**VAX3**	**1.4**
MDP 113	Fargo	1.3				
MDP 133	Medalist	1.3				
MDP 134	Navigator	1.3				
MDP 142	ROG 312	1.3				
MDP 319	Reliant	1.3				
MDP 331	CDC Expresso	1.3				
MDP 332	CDC Jet	1.3				
	VAX3	**1.3**				

CONCLUSIONS: Consistent reactions to Fusarium root rot were observed across the five control lines evaluated. Our recommendation is that these lines are included in future Fusarium root rot evaluations to aid in assay reproducibility and enable comparisons across studies. Sources of resistance to Fusarium root rot were identified in both the ADP and MDP lines evaluated. This protocol for consistent, repeatable phenotyping will aid in further identifying reliable sources of resistance to Fusarium root rot in dry beans.

LITERATURE CITED

Van Schoonhoven, A. and Pastor-Corrales, M. 1987. Standard system for the evaluation of bean germplasm. Pp. 36-38 In: Centro Internacional de Agricultura Tropical. Cali, Colombia.

O'Donnell K, Kistler HC, Cigelnik E, Ploetz RC. 1998. Multiple evolutionary origins of the fungus causing Panama disease of banana: concordant evidence from nuclear and mitochondrial gene genealogies. Proc Natl Acad Sci USA 95:2044–2049.

ANGULAR LEAF SPOT RESISTANCE – GWAS OF FIELD AND GREENHOUSE SCREENINGS IN COLOMBIA

Nay MM[1,2]*, Buendia HF[2], Portilla AE[2], Studer B[1], Raatz B[2]

[1] ETH Zürich, Switzerland, Molecular Plant Breeding (michelle.nay@usys.ethz.ch)
[2] CIAT Cali, Colombia, Bean Program

INTRODUCTION: Angular leaf spot (ALS) caused by the fungus *Pseudocercospora griseola* is an important disease in common bean and causes high yield losses in the tropics and subtropics. ALS resistant common bean lines have been characterized and resistance loci were repeatedly found on chromosomes 1, 4, 8 and 10 (Carvalho et al. 1998; Mahuku et al. 2009; Sartorato et al. 1999; Oblessuc et al. 2012). However, due to the high diversity of the pathogen and the pathotype-specificity of the resistance in common bean, efficient and durable resistance is difficult to achieve (Mahuku et al. 2002). This work aims at finding resistance loci specific to Colombian isolates of *P. griseola* in the field and greenhouse.

MATERIALS AND METHODS: To study ALS pathotype-specificity, a panel of 316 common bean lines, named extBALSIT, was evaluated in the greenhouse with a single isolate and in the field with a mix of isolates. In the greenhouse, a single isolate of the race 63-47 was used and in the field, a mix of five isolates previously collected in Darien with the races 63-15, 31-47 (2x), 31-39 and 15-44 was used. The panel was assembled to contain a collection of the most resistant plant material available at CIAT, including the Bean ALS International Trial (BALSIT) panel of previously characterized resistance sources, breeding material with phenotypic variability for ALS response and susceptible checks. Disease severity was evaluated with the CIAT standard scale ranging from 1 (no disease symptoms) to 9 (very severe disease symptoms and defoliation). The bean lines were genotyped-by-sequencing (GBS) using the restriction enzyme ApeKI (Elshire et al. 2011) and SNPs were extracted using the NGSEP pipeline (Perea 2016), filtering for a minimum quality score of Q40, maf <0.5, 20% missing data and removing heterozygote values. Genomic positions correspond to the v2.1 of the *Phaseolus vulgaris* reference genome. Genome-wide association studies (GWAS) were conducted with the TASSEL 5.0 MLM model using PCA to correct for population structure and the K matrix to correct for kinship (Bradbury et al. 2007).

RESULTS AND DISCUSSION: The extBALSIT panel was tested in the greenhouse with race 63-47 of *P. griseola,* and in the field in Darien (Colombia). Phenotypic results showed different distribution of resistant and susceptible lines between the experiments, indicating that the resistance in the panel is pathotype-specific (Figure 1).

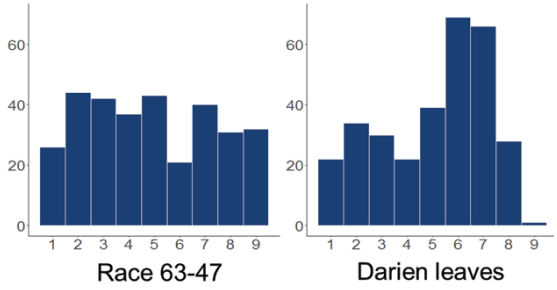


Figure 1: Histogram of mean ALS scores of the 316 lines in the extBALSIT panel tested in the greenhouse and in the field.

After filtering, GBS resulted in 22,765 SNPs that were tested for their association to disease resistance scores. A major resistance locus on chromosome 8 was identified in both trials (Figure 2). This locus at the end of chromosome 8 coincided with the previously characterized resistance locus *Phg-2,* found in the Mesoamerican bean background (Sartorato et al. 1999). Our results demonstrate the high importance of the *Phg-2* locus in conferring ALS resistance to Colombian pathogen isolates.

Resistant and susceptible lines were best distinguished with the T/G SNP at position 61,901,182 on chromosome 8 (Figure 3). The T allele at this position originated from genotype G10474 and was contained in several MAB lines (MAB 348-351, 354, 373). These lines showed broad-spectrum resistance to race 63-47 in the greenhouse and to a mix of five races in the field in Darien. The locus, however, did not explain the resistance completely and there were probably other resistance loci present, which could not be detected using this panel. This SNP constitutes a promising candidate for the development of molecular markers and its use in marker-assisted selection.

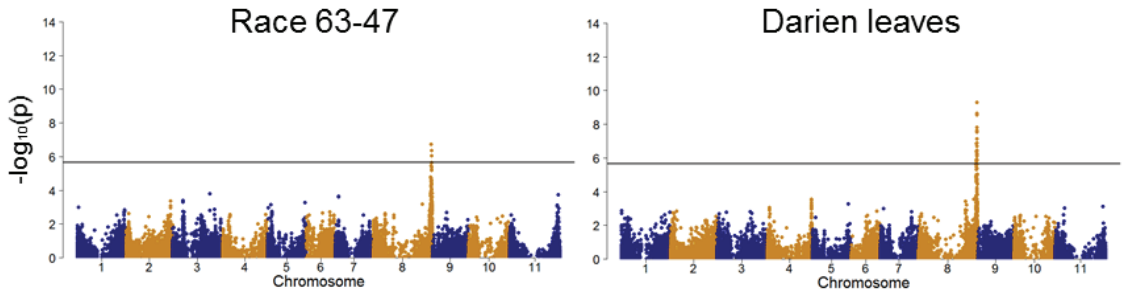


Figure 2: GWAS results of ALS disease resistance on leaves with race 63-47 in the greenhouse and with a mix of races in the field in Darien. The significance threshold was Bonferroni-corrected with α = 0.05.

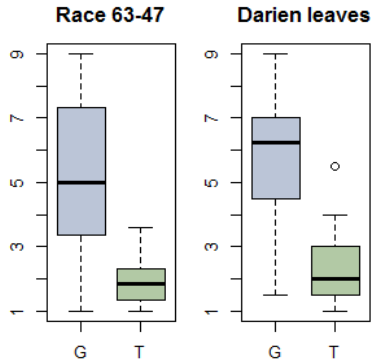


Figure 3. Selected SNP marker tagging the resistant lines. Shown are ALS scores of the lines with nucleotide G (n=200) and T (n=25) at position 61,901,182bp on chromosome 8.

REFERENCES:

Bradbury, PJ et al., 2007. TASSEL: Software …. *Bioinformatics,* 23, pp.2633-2635.

Carvalho, GA et al., 1998. Inheritance of resistance of the Andean bean line AND-277 to race 63-23 of Phaeoisariopsis griseola and identification of a RAPD marker linked to the resistance gene. *Fitopatologia Brasileira,* 23(4), pp.482–485.

Elshire, RJ et al., (2011) A Robust, Simple Genotyping-by-Sequencing (GBS) Approach for High Diversity Species. *PLoS ONE,* 6(5): e19379

Mahuku, GS et al., 2002. Genetic variability within Phaeoisariopsis griseola from Central America and its implications for resistance breeding of common bean. *Plant Pathology,* 51(5), pp.594–604.

Mahuku, GS et al., 2009. Genetics of angular leaf spot resistance in the Andean common bean accession G5686 and identification of markers linked to the resistance genes. *Euphytica*, 167(3), pp.381–396.

Oblessuc, PR et al., 2012. Mapping of angular leaf spot resistance QTL in common bean (Phaseolus vulgaris L.) under different environments. *BMC Genetics*, 13(50).

Perea, C et al., 2016. Bioinformatic analysis of genotype by sequencing (GBS) data with NGSEP. *BMC Genomics*, 498(17, supl 5), pp.539-551.

Sartorato, A et al., 1999. SCAR Marker linked to Angular Leaf Spot Resistance Gene in Common Bean. *Annual Report of the Bean Improvement Cooperative*, pp.23–24.

UROMYCES APPENDICULATUS PREVALENCE IN DRY BEAN FIELD IN NORTH DAKOTA

Cecilia Monclova-Santana[1], Samuel G. Markell[1], Maricelis Acevedo[3] and Julie S. Pasche[1]

[1]Department of Plant Pathology, [2] Department of Plant Sciences, North Dakota State University, Fargo, ND, USA 58108. [3]Cornell University Department of International Programs

INTRODUCTION North Dakota is the leading dry-bean producing state in the US. North Dakota farmers grow approximately 32% of the total dry beans produced in the country, with production concentrated in the northeast corner of the state. Common bean rust caused by the biotrophic fungus, *Uromyces appendiculatus* (Pers.:Pers.) Unger, has been a major disease of dry beans in North Dakota. In 1994, the rust epidemic resulted in 16% yield loss in North Dakota (Venette et al., 1998). The subsequent incorporation of the resistance gene *Ur-3* in new varieties greatly reduced rust incidence throughout the 1990's and 2000's. In 2008, *U. appendiculatus* race 20-3 was reported; this race is virulent on resistant genes *Ur-3*, *Ur-6*, *Ur-7*, and unknown gene from Montcalm (Markell et al., 2009). Genetic resistance is the primary tool to control dry bean rust; however, current grower-preferred varieties are susceptible to most prevalent rust races 20-3 (Knodel et al., 2017; Markell, 2008). The objective of this research is to identify dry bean rust races currently present in North Dakota with the aim to provide direction to future dry bean breeding strategies.

MATERIALS AND METHODS Leaves containing rust pustules were collected from surveyed fields in the primary dry bean regions of North Dakota in 2015 and 2016. A total of 62 samples from 26 fields and 53 samples from 35 fields were collected in 2015 and 2016, respectively. Urediniospores from field samples were inoculated on susceptible pinto UI14 using Soltrol and a 0.05% Tween 20 solution to generate single pustule isolates. Single pustule isolates were increased and race-typed using the standard differential set (Steadman et al. 2002). Pustule size was directly measured using a comparator 14 days after inoculation using a 0-6 scale, where a phenotype equal to or greater than 4,3 was considered a virulent/susceptible reaction (Stavely, 1984).

RESULTSA total of 61 and 27 single pustule *U. appendiculatus* isolates obtained in 2015 and 2016 were characterized for virulence phenotype, respectively. Approximately 80% of isolates from 2015 were identified as race 20-3, (Table 1). Ten percent of isolates were identified as race 28-3. Races 20-2, 20-11, 21-3, 28-11 and 29-3 were identified, but each of these races were represented by less than 2% of isolates. In 2016; almost 60% of isolates were identified as 20-3 and 20% of isolates were race 21-3. Races 16-3, 24-3, 28-3, 29-3, 29-7, 29-31, 31-7 were each identified in less than 2% of isolates. New races identified overcome known resistance gene *Ur-9, Ur-12, Ur-4, Ur-13*: and just a handful of isolates were virulent to *Ur-5* and *Ur-3+*.

CONCLUSIONS Bean rust has become more prevalent in North Dakota since 2014 growing seasons. Among the 88 *U. appendiculatus* isolates tested across 2 years, 99% were virulent on the widely deployed gene *Ur-3*, emphasizing the great need for the incorporation of additional resistance genes in North Dakota.

Table 1: Virulence phenotype of nine bean rust races found in North Dakota 2015 and 2016.

Differential line	Resistance gene	Race[a]									
		16-3	20-2	20-3	20-11	21-3	31-7	28-11	28-3	29-3	29-31
Early Gallatin	*Ur-4*	2	2	2	2	4,3	5,4	2	2	5	4,5
Redlands Pioneer	*Ur-13*	2	1	1	1	2,3	4,3	1	2,3	2,3	1
Montcalm	Unknown	3,4	4,3	4,5	5	4,5	4,3,2	4	4	4	5
PC-50	*Ur-9, Ur-12*	1	2	2	2	3	6,3	4,3	4	5,4	5,4,3
Golden Gate Wax	*Ur-6*	5,4	6	5,6	5,6	5,6	6	5	6,5	6	6,5
PI 260418	Unknown	3,4	1	1	1	2,3	2	2,3	3,2	3,2	2
GN1140	*Ur-7*	6	3,4	6	5	6	6	6	6	6	6
Aurora	*Ur-3*	5,4	5	4	5,6	4,5	6	4	6,5	5,6	6
Mexico 309	*Ur-5*	1	1	1	1	2,3	5,6	2,3	2,3	1	6
Mexico 235	*Ur-3+*	3,4	2,3	3,2	4,3	3,2	3	4,3	3,2	1	4
CNC	Unknown	3,2	1	1	1	1	3,2	3,2	2,3	1	4,3
PI 181990	*Ur-11*	1	1	1	1	1	1	1	1	1	1

[a] Reaction grades 1= No visible symptoms; 2= Necrotic spots without sporulation; 2,3= Reaction 2 with few type 3; 3,2= reaction type 3 with few type 2; 3= Uredinia <0.3 mm in diameter; 3,4= reaction 3 with few type 4; 4,3= Reaction 4 with few type 3; 4= Uredinia 0.3-0.49 mm in diameter; 4,5= Reaction 4 with few type 5; 5,4= Reaction 5 with few type 6; 5= Uredinia 0.5-0.8 mm in diameter; 5,6= Reaction 5 with few type 6; 6,5= Reaction 6 with few type 5; 6= Uredinia 0.8-1.2 mm in diameter.

REFERENCES

Knodel, J.J., Beauzay, P.B., Endres, G.J., Franzen, D.W., Kandel, H.J., Markell, S.G., Osorno, J.M., Pasche J.S. and Zollinger, R. K. (2017). 2016 Dry Bean Grower Survey of Production, Pest Problems and Pesticide Use in Minnesota and North Dakota. Fargo, ND.

Markell, S.G., M.A. Pastor-Corrales, J.G. Jordahl, R.S. Lamppa, F.M., Mathew, J.M., Osorno, and R.S. Goswami. 2009. Virulence of *Uromyces appendiculatus* to the resistance gene Ur-3 identified in North Dakota in 2008. *Annu. Rep. Bean Improv. Coop. 52*:82–83.

Stavely, J.R. 1984. Pathogenic specialization in *Uromyces phaseoli* in the United States and rust resistance in beans. *Plant Dis. 68*:95-99

Steadman, J.R., Pastor-Corrales, M.A., and Beaver J.S. 2002. An overview of the 3rd bean rust and 2nd bean common bacterial blight international workshops. *Annu. Rep. Bean Improv. Coop. 45*: 120-124.

Venette, J.R., Gross, P.L., and Grafton, K.F. 1998. Bean rust in North Dakota. *Ann. Rpt. Bean Improv. Coop. 41*:76:77

VIRULENCE DIVERSITY OF *COLLETOTRICHUM LINDEMUTHIANUM* IN GUATEMALA AND GWAS TO IDENTIFY GENOMIC REGIONS ASSOCIATED WITH ANTHRACNOSE RESISTANCE IN COMMON BEAN

Maldonado-Mota CR[1, 2]., Pastor-Corrales MA[4], Hurtado-Gonzales OP[4], Moghaddam SM[5], Schroder S[1], McClean PE[1], Pasche J[3], Lamppa R[3], Tobar-Piñon MG[1,2], Villatoro-Merida JC[2], Miranda AN[2], Moscoso JR[2], Agreda K[2], JM Osorno[1]

[1] Department of Plant Sciences, North Dakota State University, Fargo, ND
(carlos.maldonadomota@ndsu)
[2] Instituto de Ciencia y Tecnología Agrícolas (ICTA), Guatemala.
[3]Department of Plant Pathology, North Dakota State University, Fargo, ND.
[4]USDA-ARS Soybean Genomics and Improvement Laboratory
Beltsville Agricultural Research Center, MD.
[5]Department of Plant Biology, Michigan State University, East Lansing, MI.

Anthracnose, caused by *Colletotrichum lindemuthianum* (Sacc. and Magnus) Briosi and Cavara is a fungal disease that affects common bean worldwide. Climbing beans in Guatemala represent the main source of protein for the habitants of this region (9.4 kg/person/year). Unfortunately, anthracnose threatens climbing bean production in the region. Six races were found among samples collected in Guatemala Highlands using the standard common bean differential lines. Also, a germplasm collection from ICTA Guatemala was evaluated for resistance to *C. lindemuthianum* race 73, which is the predominant race in the U.S. Approximately 10% of 369 climbing bean accessions showed no symptoms (score of 1). GWAS results using 78754 SNP markers indicated that genomic regions for resistance to *C. lindemuthianum* exist in Pv04 and Pv07.

MATERIALS AND METHODS: During the 2016, anthracnose samples from Guatemala were sent to USDA-ARS Beltsville-MD. To determine *C. lindemuthianum* races from Guatemala, the standard set of 12 race differentials was used (Pastor Corrales, 1991). The climbing bean collection from ICTA in Guatemala was used to identify potential accession with resistance to anthracnose. 369 accessions of *P.vulgaris* were used in this study. Race 73 was used to evaluate the collection of climbing beans at the NDSU greenhouse complex. The genotypic data were provided by the Dry Bean Genomics Lab at NDSU. 78754 SNP markers were used. Least Square (LS) Means were calculated using the phenotypic data of the three replicates. The output data from the LS Means was used, continuous results used were the values that showed resistance (1-3 values) and susceptibility (4-9values). A genome wide association analysis was performed using GAPIT and multiple test were used, to control structure and relatedness.

RESULTS AND DISCUSSION: In this study, 6 races were characterized based on the response of the differential set, the most frequent among the isolates was race 585, Table 1 only show 6 isolates from 16 rescued. The races identified displayed more Mesoamerican virulence genes than Andean virulence genes. Results suggest that cultivars and/or sources of resistance genes of Andean origin are needed to develop common bean cultivars with broad and durable resistance to *C. lindemuthianum* in the highlands of Guatemala or other regions

Table 1. Reaction of common bean differential lines to 6 isolates of *Colletotrichum lindemuthianum* from Guatemala.

Differentials Cultivars	Gene Pool	Anthracnose Resistant Gene	Linkage Group of *Phaseolus vulgaris*	Binary code**	CLC-13-1	CLQ-1-2	CLQ-30-4	CLC-19-1	CLC-5-1	CLC-6-2	CLC-4-1
Michelite	MA	*Co-11*	Pv03	1	1	9	9	9	9	9	5
MDRK*	A	*Co-1*	Pv01	2	1	1	1	1	1	1	1
Perry Marrow	A	*Co-1*^{3}	Pv01	4	7	1	1	1	1	1	6
Cornell 49242	MA	*Co-2*	Pv11	8	9	9	1	9	9	9	9
Widusa	A	*Co-1*^{5}	Pv01	16	1	1	1	1	1	1	1
Kaboon	A	*Co-1*^{2}	Pv01	32	9	1	1	1	1	1	1
Mexico 222	MA	*Co-3*	Pv04	64	1	7	1	7	7	7	3
PI 20762	MA	*Co-3*^{3}, *Co-4*^{3}	Pv04, 08	128	1	1	7	1	8	8	9
TO	MA	*Co-4*	Pv08	256	1	1	9	1	9	9	9
TU	MA	*Co-5*	Pv07	512	9	9	9	9	9	9	9
AB 136	MA	*Co-6, co-8*	Pv07, NA	1024	1	1	1	8	8	8	7
G2333	MA	*Co-4*^{2}, *Co-3*^{5}, *Co-5*^{2}	Pv04,08, 07	2048	1	1	1	1	1	1	5
				Race	**556**	**585**	**897**	**1609**	**1993**	**1993**	**3981**

*Differential Andean cultivar MDRK=Michigan Dark Red Kidney

**Binary value: The designation of the race results from the addition of the binary value of each susceptible differential

R: (1-3) resistant reaction; S: (4-9) susceptible reaction

NA: Not available

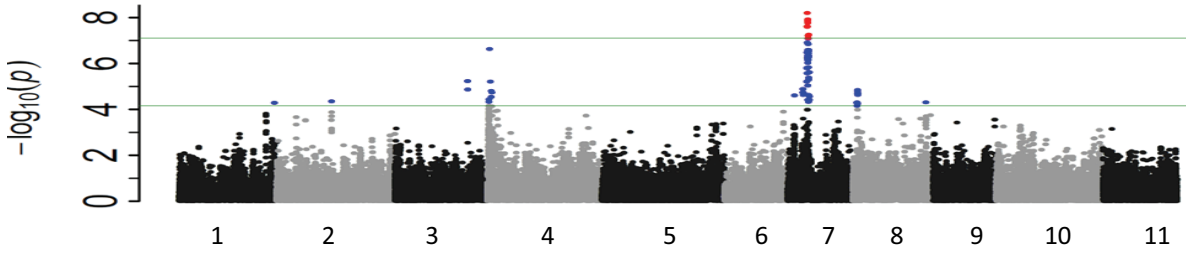


Figure 1. Manhattan plot using Efficient mixed model analysis (EMMA) for *C. lindemuthianum* resistance to race 73. The green line is the cut-of value to call a peak significant. SNPs above the 0.01 percentile are highlighted in red, while those above 0.1 are highlighted in blue above the yellow line. Numbers below the Manhattan plot represent chromosomes.

Also, our research suggests that climbing bean germplasm collected in highlands from Guatemala is a good potential source of resistance against race 73. Ten percent of the population is symptomless (score of 1) and 56% of the population is resistant (scored of 2-3). However, GWAS results for resistance of the climbing bean germplasm from Guatemala showed that the most significant chromosomal region involved in the resistance to *C. lindemuthianum* is located in Pv07 and Pv04 (Figure 1).

REFERENCES:

Pastor-Corrales, M.A. 1991. Estandarización de variedades diferenciales y de designación de razas de *Colletotrichum lindemuthianum*. (Standardizing differential varieties and races designation of *Colletotrichum lindemuthianum*) Phytopathol. 81:694.

SYMBIOTIC NITROGEN FIXATION IN THE MESOAMERICAN GENEPOOL OF COMMON BEAN

Jennifer Wilker[1*], Alireza Navabi[1], Timothy Porch[2], Philip McClean[3], K. Peter Pauls[1]

[1] Department of Plant Agriculture, University of Guelph (jwilker@uoguelph.ca)
[2] United States Department of Agriculture, Agriculture Research Service Puerto Rico
[3] Department of Plant Sciences, University of North Dakota

INTRODUCTION. Legume species form symbiotic associations with nitrogen-fixing bacteria wherein the *Rhizobium* spp. convert atmospheric nitrogen into useable plant forms. Common beans (*Phaseolus vulgaris*, L) are generally considered ineffective at nitrogen fixation (Bliss, 1993). As a quantitative trait, nitrogen fixation is controlled by multiple genetic as well as non-genetic factors. Discovery of quantitative trait loci (QTL) can be accomplished through association mapping methods, often carried out using the progeny of bi-parental populations segregating for trait(s) of interest (recombinant inbred lines, RILs) or diversity panels, as in genome wide association studies (GWAS). Recent association mapping studies have reported wide variation for traits associated with nitrogen fixation in common bean RILs and diversity panels (Farid, 2015; Kamfwa, 2015). The present study aims to investigate the phenotypic diversity for nitrogen fixation within the Mesoamerican genepool of common bean and to identify QTL associated with the trait using GWAS.

MATERIALS AND METHODS. The 300-genotype Mesoamerican Diversity Panel (MDP; BeanCAP) and 19 AAFC-UofG Mesoamerican cultivars were inoculated with *Rhizobia* and grown in an alpha-lattice design (two replicates/location) at three low-nitrogen field sites in Ontario (2014, 2015) and Puerto Rico (2015-2016). Various agronomic and symbiotic nitrogen fixation (SNF)-related parameters were measured in the field and post-harvest. Isotope analyses were performed on ground seed samples and results (ΔN values) were used to quantify nitrogen-fixing capacity using the natural abundance method (Shearer & Kohl, 1986). The non-nodulating common bean mutant R99 (Park & Buttery, 1988) was used as the non-fixing genotype in percent nitrogen derived from the atmosphere (%Ndfa) calculations. Analysis of variance and multi-variate analyses were performed in SAS 9.3 (SAS Institute). Single nucleotide polymorphism (SNP) genotyping was performed on the MDP at North Dakota State University using the BARCBean6K_1 and BARCBean6K_2 SNP chips (Moghaddam *et al*, 2016). SNP genotyping of the Canadian genotypes was performed at the Québec Innovation Center with the Illumina BARCBean6K_3 BeadChip (Hyten *et al*, 2010). SNP data was pooled for all genotypes, the common SNPs retained, then further filtered for 0.05% minor allele frequency (MAF) and filtered for presence in 90% of the panel; consequently, GWAS was performed using 253 individuals and 3527 SNPs. GWAS analysis was performed in TASSEL5 (Bradbury *et al*, 2007).

RESULTS AND DISCUSSION. Diversity for traits associated with SNF was found within the Mesoamerican genepool of common bean. Significant differences (P=0.0001) were found for %Ndfa among genotypes and ranged from 9% (*cv*. Bandit) to 79.6% (*cv.* ABCP-8) at Elora, 2014 (Fig. 1). Greater variation (3.6-98.2%) for %Ndfa was found in the Andean Diversity Panel of common bean (Kamfwa, 2015), whereas less variation (11-51%) for %Ndfa was found in a RIL population segregating for the trait (Farid, 2015). Nodule count was significantly different (P<0.0001) among genotypes and ranged from 1.1 (*cv*. Gala) to 55.3 (*cv.* Lightning) at Isabela, 2016 (Fig. 2). A study of seedling nodulation found a smaller range (18-32) among 7

Mesoamerican genotypes (Farid, 2015). Genotype by trait bi-plot analysis (Fig. 3) revealed that genotype performance differed between races (Mesoamerica *vs.* Durango-Jalisco). At Elora (Fig. 3, A) yield and %Ndfa were closely associated, whereas at Belwood (Fig. 3, B) these two traits were not associated. Leaf chlorophyll content measurements were not associated with %Ndfa at Elora, while these traits were negatively associated at Belwood. Days to flowering was not associated with %Ndfa at either location. Preliminary GWAS analysis found a QTL associated with %Ndfa on Pv08 (data not shown). Further GWAS studies will be carried out to find and confirm QTL associated with SNF in the Mesoamerican gene pool of common bean.

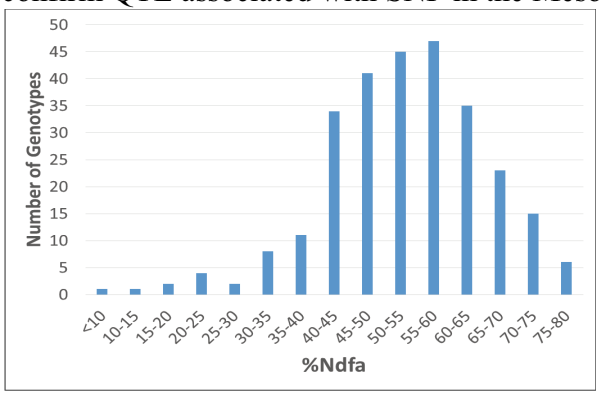


Figure 1. Distribution for %Ndfa (lsmeans) for 275 genotypes grown at Elora, 2014. Mean 52.9% ± 1.43%.

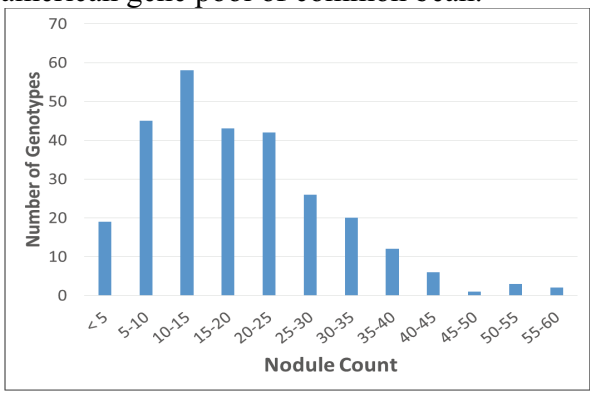


Figure 2. Distribution for Nodule Count (lsmeans) for 276 genotypes grown at Isabela, 2015-2016. Mean 18.8% ± 1.23%.

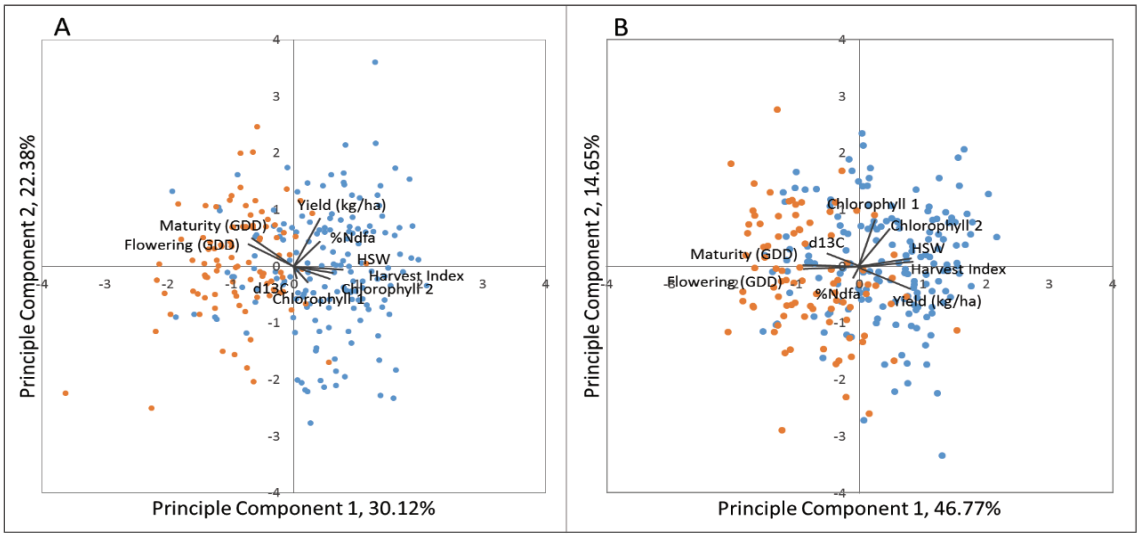


Figure 3. Genotype by trait biplots for the MDP + 18 Canadian genotypes grown at (A) Elora, 2014 and (B) Belwood, 2015. Durango-Jalisco race genotypes are denoted by blue dots (●) and Mesoamerica race genotypes are denoted by orange dots (●).

REFERENCES:

Bliss. 1993. Plant Soil 152:71-79. Bradbury et al. 2007. Bioinformatics 23:2633-2635. Farid. 2015. University of Guelph, Canada. Hyten et al. 2010. BMC Genomics 11:475. Kamfwa. 2015. Michigan State University, United States. Moghaddam et al. 2016. Plant Genome Volume 9. Park & Buttery. 1988. Can. J. Plant Sci. 68: 199-202. SAS Institute Inc. 2012. United States. Shearer & Kohl. 1986. Aust. J. Plant Physiol. 13:699-756.

USING HIGH TEMPERATURES TO ASSIST IN SCREENING *PHASEOLUS* SPP.

Traub J[1]*, Porch T[2], Naeem M[3], Austic G[4], Kelly JD[5], Loescher WH[1]

[1]Michigan State University (MSU), Department of Horticulture (traubjes@msu.edu), [2]United States Department of Agriculture - ARS Tropical Agriculture Research Station, [3]The Islamia University of Bahawalpur, Cholistan Institute of Desert Studies [4]MSU, MSU-DOE Plant Research Laboratory [5]MSU, Department of Plant, Soil and Microbial Sciences

INTRODUCTION

Heat stress negatively impacts the yield of common beans (*Phaseolus vulgaris* L.) and prevents their cultivation in certain areas. Furthermore, under field conditions, heat stress often coincides with and exacerbates the effects of drought stress. This research examined a variety of methods for screening large numbers of bean germplasm exposed to heat stress.

MATERIALS AND METHODS

Evaluated in this experiment were ten common bean lines from a drought stress panel used in previous field experiments, four common bean cultivars used in previous greenhouse drought experiments, and the stress tolerant tepary bean (*Phaseolus acutifolius* A. Gray) line TB1. Because of space considerations, bean plants were originally grown in the greenhouse until the third trifoliate leaf was mature, at which point plants were moved to a growth chamber for a week. There, temperatures increased from 35/30 °C (day/night) to 45/40 °C in 5 °C increments. After two days of acclimation at each temperature, photosynthesis, chlorophyll fluorescence, and visual rating was measured for each plant using a LI-COR 6400XT, a MultispeQ, and the human eye, respectively. Plantings were staggered to ensure each plant was the same maturity at the time of measurement.

RESULTS AND DISCUSSION

Bean plants exposed to temperatures of 45 °C for two days showed signs of heat stress as measured by gas exchange, chlorophyll fluorescence, oxidative stress, and visual ratings. At lower temperatures, no signs of heat stress were apparent, but at 45 °C, a gradient of responses appeared for the traits measured. TB1 showed the greatest tolerance to heat stress, with smallest reductions in photosynthesis and chlorophyll fluorescence and little visual damage to its leaves. Other genotypes like SB787 ranked consistently low for all measurements at 45 °C. Gradually raising temperature was useful for screening a large group of germplasm for heat tolerance, but this heat tolerance only partially related to drought tolerance observed in the field. Despite the coincidence of drought and heat stress in the field, tolerance to these stresses must be independently screened for. Gas exchange, chlorophyll fluorescence, and visual inspection methods are all useful for screening for heat stress; the resources available can dictate which method researchers use. Plant breeders can utilize some of the methods described here to supplement field data and further characterize the stress tolerance of later generation bean lines.

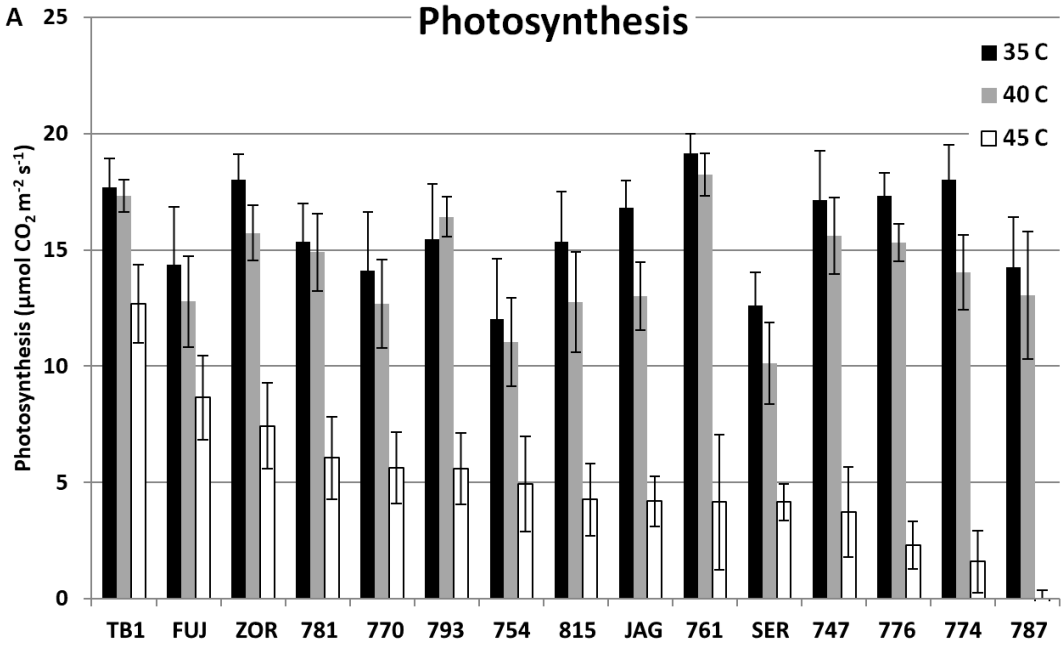


Figure 1. The photosynthetic rates of fifteen bean genotypes measured at 35, 40, and 45 °C. Measurements were taken with a LI-COR 6400XT after two days of acclimation at each temperature in a growth chamber.

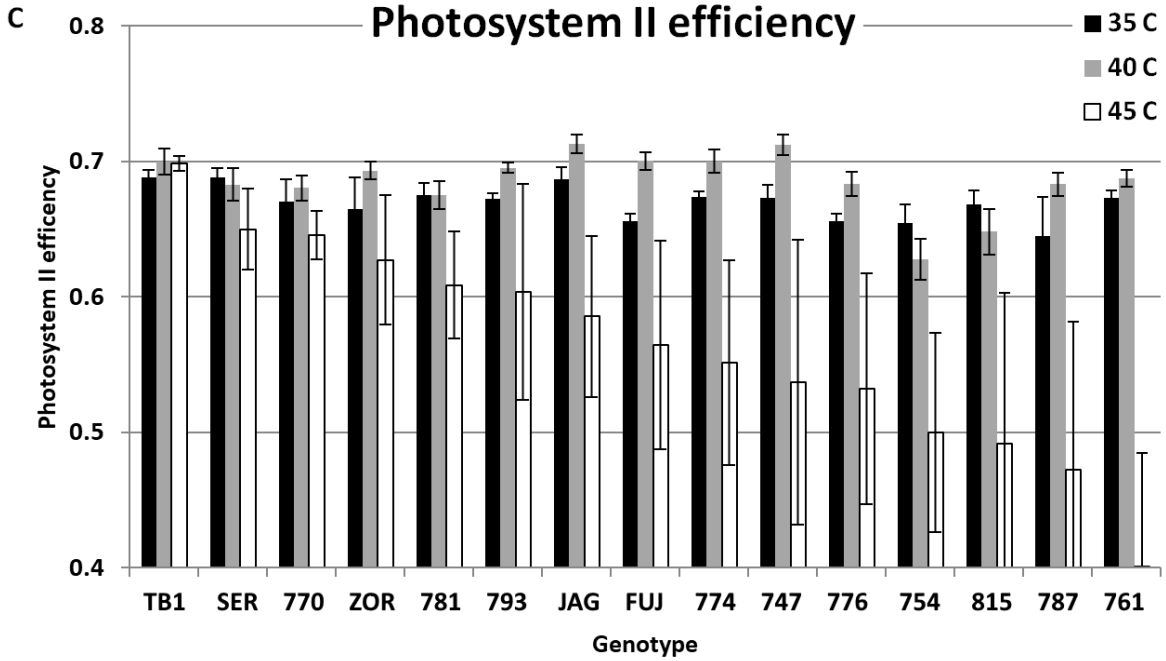


Figure 2. The photosystem II efficiency, a chlorophyll fluorescence measurement, of fifteen bean genotypes measured at 35, 40, and 45 °C. Measurements were taken with a MultispeQ after two days of acclimation at each temperature in a growth chamber.

FIELD SCREENING AND SELECTION OF COMMON BEAN CARIOCA SEEDED PROGENIES WITH MULTIPLE RESISTANCE TO BCMV, BGMV AND CPMMV

Thiago Lívio P. O. Souza[1]*, Rodrigo S. Silva[2], Josias C. Faria[1], Adriano M. Knupp[1], Marcelo S. Aguiar[1], Helton S. Pereira[1], and Leonardo C. Melo[1]

[1]Embrapa Arroz e Feijão, Santo Antônio de Goiás, GO 75.375-000, Brazil;
[2]Universidade Federal de Goiás (UFG), Goiânia, GO 74.690-900, Brazil;
*Corresponding author: thiago.souza@embrapa.br

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is among the most important edible legume crops in the world, mainly because of its social and economic importance as well as its high nutritional value. Brazil is the world's largest producer and consumer country of common bean (FAO, 2017). Approximately 70% of beans consumed by Brazilians come from the carioca market class (light beige seeds with light brown stripes, nearly full elliptical shape, opaque, not shiny), making it the most popular commercial class in Brazil, although it has very restricted consumption in other countries (CONABE, 2017). Despite the genetic progress obtained in the last decades, the common bean crop still present an average seed yield below its yield potential in Brazil. One of the factors that compromise the yield performance and reduce the commercial quality of seeds is its susceptibility to a large number of diseases. Among these diseases, viruses caused by *Bean common mosaic virus* (BCMV), *Bean golden mosaic virus* (BGMV) and *Cowpea mild mottle virus* (CPMMV) are major concerns, mainly in Central Brazil. Seed yield losses ranging from 40 to 100% are being reported, depending on the rate of occurrence, time of sowing, time of plant infection, and cultivar choice (Faria *et al.*, 2016). The main goal of the present work was to evaluate common bean elite progenies from the carioca market class in the field and select those with multiple resistance to BCMV, BGMV and CPMMV.

MATERIAL AND METHODS

Two field nurseries were carried out at Embrapa Arroz e Feijão (Santo Antonio de Goiás, GO, Brazil) during the rainy growing season of 2016 and the dry growing season of 2017. Thirty-nine elite progenies were evaluated, all harboring the transgenic event Embrapa 5.1 which confers resistance to BGMV, as well as three carioca seeded control cultivars (BRS Estilo, BRS FC402 and BRS FC401 RMD). Of these 39 progenies, 10 derived from the cross BRS Estilo × CNFCT 16206, on generations $BC_4F_{4:6}$ and $BC_3F_{5:7}$, and 29 from the cross BRS Sublime × BC_3F_1 (BRS Estilo × CNFCT 16206), on generations $F_{3:6}$ and $F_{4:6}$. Both nurseries were carried out in a randomized block design with three replicates, using the regular technologies recommended for the crop but without control of diseases and pests. The plots consisted of four 4.0 m–rows with 0.5 m between rows and 10–12 plants/m. The scoring scale used to evaluate virus severity ranged from 1 (absence of disease symptoms and signs of pathogens) to 9 (80-100% disease severity or plant death) (Melo, 2009). Individual and combined variance analyses ($p < 0.01$) were performed using the F test. The comparison of means were accomplished by the Scott-Knott method ($p < 0.05$).

RESULTS AND DISCUSSION

The combined analysis for virus severity showed variability between progenies, environments, and the presence of G × E interaction ($P \leq 0.01$). As expected, this differential response of the progenies to the environments is because of the highest natural pressure of viruses in the dry growing season (Souza *et al.*, 2017). All progenies showed effective resistance to BCMV and BGMV, exhibiting mean severity scores of 1.0, whereas the conventional controls were susceptible to BGMV, exhibiting mean severity score ≥6.0. It was not possible to evaluate the severity of CPMMV in the control cultivars since the symptoms were totally confused or hidden by the symptoms of BGMV. For this reason, as previously reported by Souza *et al.* (2017), the severity of CPMMV was evaluated only in elite progenies. Twelve progenies showed mean score for CPMMV severity ≤3.0 and, therefore, they were selected as resistant to BCMV (gene *I*), BGMV (event Embrapa 5.1) and CPMMV (resistance gene(s) under characterization). Individual plants were selected from these 12 progenies to develop carioca seeded inbred lines homozygous for the resistance to the three viruses. This breeding step is being aided by marker-assisted selection and artificial inoculation. The resulting inbred lines will be further tested in final field trials for a wide agronomic performance evaluation beginning next year.

ACKNOWLEDGMENTS

This work is supported by Brazilian Agricultural Research Corporation – Embrapa (Grant No. 02.14.01.004.00). T.L.P.O. Souza, H.S. Pereira and L.C. Melo are supported by Brazilian Council for Scientific and Technological Development (CNPq).

REFERENCES

CONABE – Companhia Nacional de Abastecimento. 2016. Indicadores da agropecuária. http://www.conabe.gov.br (accessed on 29 Nov. 2017).

FAO – Food and Agriculture Organization of the United Nations. 2016. Faostat. http://faostat3.fao.org/browse/Q/QC/E (accessed on 29 Nov. 2017).

Faria J.C., Aragão F.J.L., Souza T.L.P.O., Quintela E.D., Kitajima E.W., and Ribeiro S.G. 2016. Golden mosaic of common beans in Brazil: management with a transgenic approach. APS Features. doi:10.1094/APSFeature-2016-10.

Melo L.C. 2009. Procedimentos para condução de ensaios de valor de cultivo e uso em feijoeiro-comum. Embrapa [Documentos, 239]. Embrapa, Santo Antônio de Goiás, GO.

Souza T.L.P.O., Faria J.C., Aragão F.J.L., Del Peloso M.J., Faria L.C., Wendland A., Aguiar M.S., Quintela E.D., Melo C.L.P., Hungria M., Vianello R.P., Pereira H.S., and MELO, L.C. 2017. Agronomic performance and yield stability of the RNAi-based Bean golden mosaic virus resistant common bean. Crop Science, in press (doi:10.2135/cropsci2017.06.0355).

SENSORY CHARACTERISTICS OF ANDEAN DRY BEANS

Amber N Bassett[1], Karen A Cichy[12] and Daniel Ambechew[3]

[1]Department of Plant, Soil, and Microbial Sciences, Michigan State University, East Lansing, MI, USA; [2]Sugar beet and Bean Research Unit, USDA-ARS, East Lansing, MI, USA; [3]Southern Agricultural Research Institute, Hawassa, Ethiopia

INTRODUCTION

Dry beans are a nutrient-rich food that display a range of sensory characteristics. Sensory quality includes important traits considered when consumers select dry beans and when food companies include dry beans in products, but these traits have largely been overlooked by breeders. This is partly due to the expense and complexity involved in incorporating sensory traits in breeding programs, but even for breeders interested in incorporating sensory traits in their programs, few resources are available concerning which traits are associated with consumer preference and how these traits are genetically controlled. Using the Andean Diversity Panel (ADP)[1], we have developed a protocol to assess sensory characteristics in dry beans. This information can enable breeders to target specific sensory profiles in their programs, as well as allow agronomic traits of dry bean varieties to be improved without sacrificing desirable flavor. Future work is needed to address which traits are most associated with consumer preference and which sensory profiles are most suited for use in new products using dry beans as ingredients.

MATERIALS AND METHODS

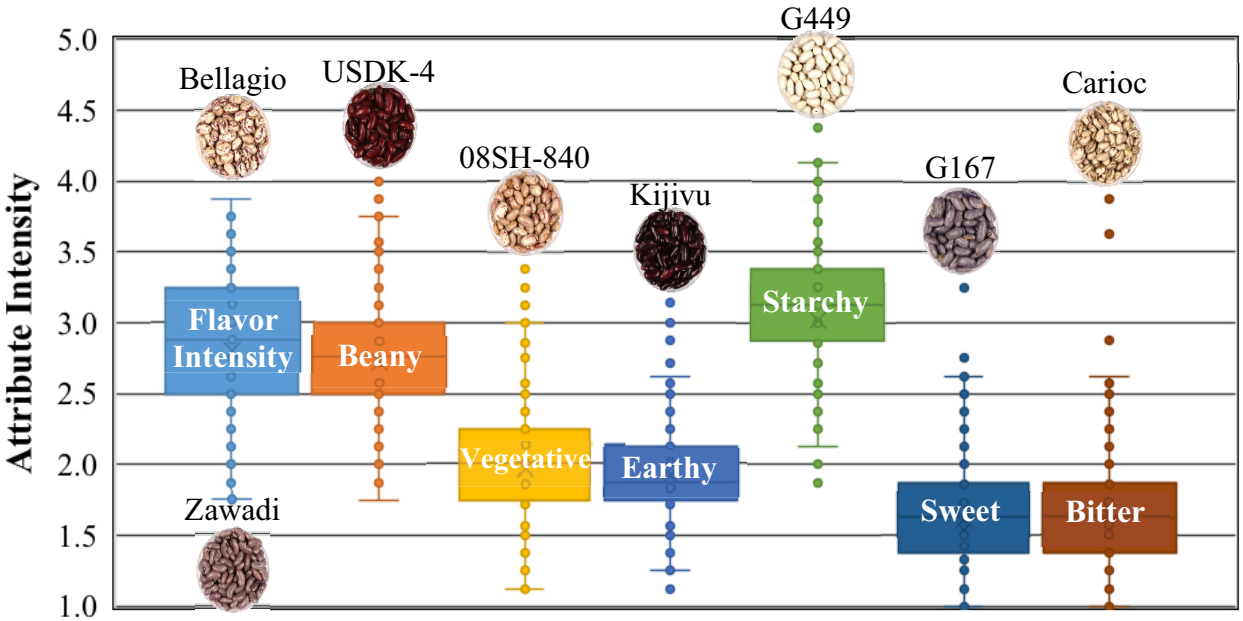
388 genotypes of the ADP were grown in Hawassa, Ethiopia in 2015 by the Southern Agricultural Research Institute and evaluated for sensory characteristics via a quantitative descriptive analysis approach using a trained panel. Panelists at Michigan State University were trained using a diverse set of dry beans exhibiting a range of attribute intensities. The sensory traits evaluated using 5-point attribute intensity scales (low → high intensity) included beany, vegetative, bitter, sweet, starchy, earthy, and total flavor intensity as well as seed coat perception and cotyledon texture. The 80% cooking time of soaked beans cooked in distilled water as determined by Mattson cookers[2] was used as the time required to cook each genotype to completion. A Quantitative Descriptive Analysis (QDA) approach was taken for ADP evaluation, in which each panelist independently evaluated samples to limit group bias. Each genotype was evaluated in two separate sessions with four panelists per session for a total of eight evaluations per genotype. The resulting ratings form the sensory profiles for the ADP, which were used to compare genotypes, select genotypes for further study, and to explore the genetic control of each trait using a GWAS approach in TASSEL[3].

RESULTS AND DISCUSSION

Across the ADP, a broad range of attribute intensities was observed for all sensory characteristics (Figure 1, texture traits not included). Statistically significant differences ($\alpha = 0.05$) were found for all traits except beany and earthy at the genotype level. This reflects the range of sensory characteristics present in the ADP and provides information regarding which beans may be useful as ingredients, as extremes for future sensory evaluations, or even as potential breeding material. A range of sensory characteristics also exists within each seed type (data not shown). This suggests that currently, seed type does not define the flavor or texture of a dry bean, but presents an opportunity to target consistent, desirable sensory profiles when breeding dry beans for current and new market classes and uses. Additionally, little correlation was observed ($-0.3 \leq r \leq 0.3$, $\alpha = 0.05$) among attribute intensities, which suggests that sensory characteristics can be combined in

many ways through breeding to achieve target sensory profiles. Using association mapping, significant markers associated with attribute intensities were identified for multiple traits, including the bitter trait, which had associated markers on Pv03, Pv05s Pv07, and Pv10 meeting the Bonferroni correction threshold ($\alpha = 0.05$) and additional markers on Pv03, Pv04, and Pv07 meeting the FDR threshold ($\alpha = 0.05$) (data not shown). Breeders can be reasonably confident that bitterness is negatively associated with consumer preference, and bitterness may be a reasonable entry-level trait to consider for those breeders interested in incorporating sensory traits in their breeding programs. A second year of phenotyping will reveal which markers are stable and useful for selection. This will set the foundation for marker-assisted selection of sensory traits in dry beans to allow for improvement of agronomic traits without sacrificing desirable sensory quality and for the targeting of specific sensory profiles to address market class consistency, consumer preference, and industry demand.

Figure 1: Attribute intensities of the Andean Diversity Panel for the sensory traits evaluated by a trained panel using a Quantitative Descriptive Analysis (QDA) approach. The names and images of genotypes identified as extremes for each trait are included.



REFERENCES

1. Cichy, K. A. *et al.* A Phaseolus vulgaris diversity panel for andean bean improvement. *Crop Sci.* **55,** 2149–2160 (2015). 2. Wang, N., Daun, J. K. & Malcolmson, L. J. Relationship between physicochemical and cooking properties, and effects of cooking on antinutrients, of yellow field peas (*Pisum sativum*). *J. Sci. Food Agric.* **83,** 1228–1237 (2003). 3. Bradbury, P. J. *et al.* TASSEL: software for association mapping of complex traits in diverse samples. *Bioinformatics* **23,** 2633–2635 (2007).

BEAN PURCHASING PREFERENCES AMONG LOW-INCOME HISPANIC AND NON-HISPANIC WOMEN IN ARIZONA AND IOWA

Donna M. Winham

Food Science and Human Nutrition
Iowa State University, Ames, IA

Dry grain pulses such as common beans are promoted across most federal nutrition agencies including the Dietary Guidelines for Americans, the Special Supplemental Food Program for Women, Infants and Children (WIC), the Supplemental Nutrition Assistance Program (SNAP), the Expanded Food and Nutrition Education Program (EFNEP), and the National School Lunch Program (NSLP). These agencies advocate to improve the nutrition status of limited resource or otherwise at-risk adults and children to reduce disparities in food access, improve nutrition knowledge, and ultimately improve health.

Despite the federal nutrition agency support for consumption, there is little published knowledge on consumer preferences for dry or canned beans in the United States. Private industries have marketing research data, but this information is not publicly available, is seldom reported outside of a given organization, and often represents views of more affluent consumers, not low-income groups. This lack of information interferes with the ability of bean researchers to direct crop improvements and seed characteristics that are of importance to consumers in general as well as for niche markets. To address some of these issues, two surveys with low-income Hispanic and non-Hispanic white women were conducted in Arizona (2012) and Iowa (2016). The study purpose was to describe purchasing patterns, determine preferred characteristics of dry beans and canned beans, and assess attitudes and perceptions towards beans in the diet. Information on knowledge of bean health benefits has been reported elsewhere.[1,2]

Women were eligible to complete the survey if they were current participants in EFNEP, WIC, or SNAP, and were 18-65 years old. Only women who self-identified as Hispanic or non-Hispanic white were included in data analysis because the number of women of other racial backgrounds was small at both study sites. Participants completed questions on Hispanic ethnicity, and the Bidimensional Acculturation Scale (BAS) that categorizes individuals by cultural affiliation as Hispanic-dominant, bicultural, or English-dominant. The English-dominant category included non-Hispanic white women too. Survey questions on preferred bean characteristics were adapted from previous research by Martínez,[3] and Cichy.[4]

Participants in both studies had similar characteristics of age, number of children in household, income, and education. Hispanic-dominant women tended to have less education, more children, be in a relationship, and have greater food security regardless of income. Hispanic-dominant women consumed beans significantly more often than bicultural or English-dominant colleagues ($p=.000$). Almost 100% of the Hispanic-dominant women purchased dry beans. Participants were given a list of eight dry bean characteristics and asked to select all that were important to them. By percent mentioned, the top three were the same for both sites: price (59% AZ, 54% IA), tradition or always buy this brand (37% AZ, 34% IA), and quality (26% AZ, 35% IA). Other traits varied slightly in importance by state: taste of beans (21% AZ, 28% IA), nutritional value (25% AZ, 22% IA), color of beans (25% AZ, 22% IA), cook quickly (16% AZ, 18% IA), brand (15% AZ, 19% IA), and shape (5% AZ, 10% IA). Some of these characteristics by acculturation status were markedly different. For the cook quickly trait, 21% AZ and 24% IA of the Hispanic-dominant respondents felt this was important in contrast to 6% AZ and 8% IA of bicultural, and 23% AZ and

15% IA of the English-dominant women. Thirty percent of IA Hispanic-dominant women chose beans on shape, but only 3% of English-dominant women did.

In Iowa, nearly double of the English-dominant participants (53%) agreed or strongly agreed that dry beans take too long to prepare in comparison to their Hispanic-dominant (28%) peers. Arizona responses were similar with 35% of English-dominant in agreement compared to 22% of Hispanic-dominant women.

Most women did not care to buy beans from a specific country (68% AZ, 91% IA), but 29% of Arizonans and 7% of Iowans did prefer a Latin American country. Iowa women were asked if they would like to buy dry beans from Iowa. Overall, 40 % said yes with 54% of the English-dominant, 29% of bicultural, and 31% of Hispanic-dominant women in favor of local dry beans.

Hispanic-dominant women in Arizona (32%) and Iowa (51%) bought canned beans less often than English-dominant women (80% Arizona; 70% Iowa). The Iowa Hispanic-dominant women had less favorable views of canned beans as indicated by a significantly lower score on the canned bean attitude scale in comparison to their peers (2.67 vs. 3.06 bicultural vs. 3.23 English-dominant; p=.000). In general, a significantly higher percentage of Hispanic dominant women in both states agreed their families would not eat canned beans (42% AZ, 41% IA), that canned beans were not true to their culture (13% AZ, 25% IA), and that canned beans do not taste good (49% AZ, 48% IA).

In conclusion, significant differences in bean purchasing, preferences, and attitudes exist across low-income Hispanic and non-Hispanic white women. Identification of the barriers, attitudes, and current consumption patterns can elucidate ways to resolve these issues and increase consumption. Nutrition education programs that take into account stigmas and perceived qualities of beans could foster positive cultural practices and proactively address the myths and misconceptions that hinder the inclusion of these healthy foods. Although there have been global efforts in developing countries to identify preferred bean traits, e.g. CGIAR, Feed the Future, little data exists for US consumers. Larger scale survey data as described on consumer attitudes, knowledge, and preferences of bean traits in dry and canned forms would be useful for breeders, growers, and other industry partners to inform practices in the US.[5]

1. Winham DM, Florian TL, Thompson SV. Low-income US Women under-informed of the specific health benefits of consuming beans. PloS One. 2016 Jan 28;11(1):e0147592.
2. Palmer SP, Winham DM, Hradek C. Knowledge gaps of the health benefits of beans among low-income women. Am J Health Behav, 2018, 42, 27-38 DOI: https://doi.org/10.5993/AJHB.42.1.3
3. Martínez L, Bernsten R, Zamora M. Estrategias de mercado para el frijol Centroamericano. Market strategies for Central America dry beans. Agronomía Mesoamericana. 2004;15(2):121-30.
4. Cichy KA, Wiesinger JA, Mendoza FA. Genetic diversity and genome-wide association analysis of cooking time in dry bean (*Phaseolus vulgaris* L.). Theoretical and Applied Genetics. 2015 Aug 1;128(8):1555-67.
5. Cichy KA, Fernandez A, Kilian A, Kelly JD, Galeano CH, Shaw S, Brick M, Hodkinson D, Troxtell E. QTL analysis of canning quality and color retention in black beans (Phaseolus vulgaris L.). Molecular breeding. 2014 Jan 1;33(1):139-54.

GENETIC CHARACTERIZATION OF THE NON-DARKENING TRAIT IN COMMON BEAN

Erfatpour M*[1] and Pauls KP[1]

[1]University of Guelph, Department of Plant Agriculture (merfatpo@uoguelph.ca)

INTRODUCTION: Seed appearance (colour, pattern, and gloss), size, and shape are three major quality characteristics that determine the market class and quality of beans. The light seed coat background in pinto beans and cranberry beans turns brown in mature seeds and with age during a process known as postharvest darkening (PHD) (Prakken 1970). Darkened beans receive low market value due to their perceived association with low quality beans. Three distinct categories have been identified for seed coat darkening in beans including: regular darkening (RD), slow darkening (SD), and non-darkening (ND) (Elsadr et al. 2011). In order to prevent the occurrence of PHD, the non-darkening (ND) trait has been introgressed into pinto and cranberry beans. It is believed that the *J* gene controls PHD in common bean (Prakken 1970). This model proposes that plants with a dominant *J* allele express the trait, but beans which are homozygous recessive at this locus (*jj*) do not darken, at all. Yet, the molecular mechanism by which *J* regulates the accumulation of brown pigments in the seeds coat remains unclear. Identification of the gene responsible for the ND trait may lead to developing a gene-based marker for marker-assisted selection (MAS) in early stages of plant breeding program for ND beans; furthermore, the biochemical process involved in PHD might be understood. The objective of this study was to characterize the genetic basis of non-darkening trait in common bean.

MATERIALS AND METHODS: A population of 128 F_2-derived RILs was developed from a cross between 'Wit-rood boontje' (a cranberry-like bean as the source of the ND trait) and 1533-13 (a SD pinto bean) and advanced to F_5 generation by single seed descent. DNA was isolated from F_5 plants and genotyped with an Illumina BeadChip (BARCBEAN6K_3) containing 5398 SNPs. The mapping population was also genotyped with STS marker $OL4S_{500}$ linked to the *J* locus (McClean et al. 2002) and a SSR marker (Pvsd-0028) linked with the *Sd* locus (Felicetti et al. 2012). The genotyping data were combined with two additional markers for non-darkening and slo

F_6 seeds were characterized for seed coat phenotype with a colorimeter (Konica Minolta CM-3500D, Osaka, Japan). A principal component analysis (PCA) was performed in EXCEL using XLSTAT software to identify the parameter that best explains the phenotypic variation.

The genotypic data were combined with the phenotypic data to map regions of the genome containing QTL associated with the ND trait. QTL mapping was performed in MapQTL 6 using Composite Interval Mapping (CIM).

A library of amplicons was prepared from the candidate genes of 6 genotypes including 'Wit-rood boontje', 1533-15, RIL29 (ND), RIL81 (RD), Othello (RD pinto bean variety), and Etna (RD cranberry bean variety), and resequenced using an Illumina MiSeq System. The sequencing results were analyzed with CLC Genomics Workbench to identify the possible polymorphism(s) associated with the ND trait.

RESULTS: A genetic linkage map was constructed using 1327 informative SNPs plus the phenotypic markers, STS marker ($OL4S_{500}$), and SSR marker (Pvsd-0028), previously associated with the *J* gene and *Sd* gene, respectively. The map consisted of 11 linkage groups and was 1253.2 cM large. In PCA analysis the parameter a* AUV was determined as the best parameter for

separation of seed coat colour classes in our mapping population (Fig. 1). Thus its values were included for the QTL analysis.

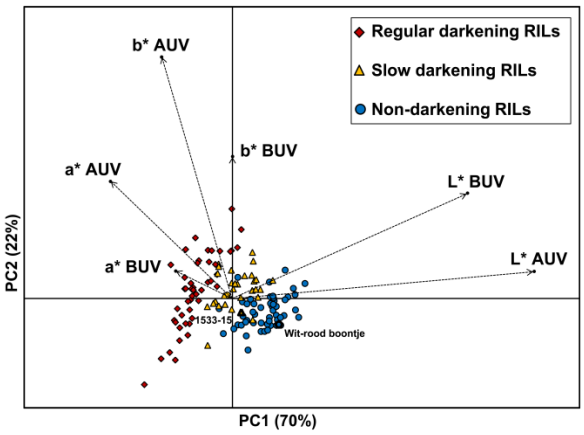


Fig. 1 The biplot of observations and variables obtained from colorimeter readings of the seed surface of 128 RILs before and after exposure to UV light. Blue dots, yellow dots, and red dots represent ND genotypes, SD genotypes, and RD genotypes, respectively. BUV: before exposure to UV, AUV: after 24 h exposure to UV, PC1: the first principal component, and PC2: the second principal component.

A major QTL for the non-darkening trait was flanked by SNP 715646341 and SNP 715646348 on chromosome 10 (Fig. 2). The region, which spanned 13.2 cM, explained 48% of the phenotypic variation for seed coat darkening. Forty candidate genes were identified in the QTL interval. Resequencing of the candidate genes which were most likely to be involved in the ND trait, including two MYB-like DNA-binding genes (Phvul.010G130500 and Phvul.010G130600), a transcription initiation factor (Phvul.010G130700), and a MYB family transcription factor (Phvul.010G131400), revealed a single nucleotide deletion in the exon region of one the candidate genes.

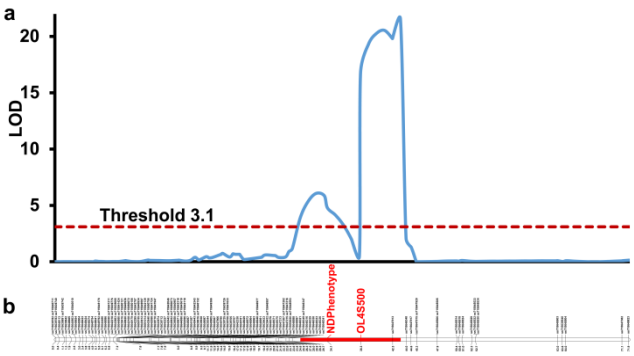


Fig. 2 ND QTL mapped to linkage group 10 with the mapping population derived from 'Wit-rood boontje' × 1533-33 cross. (a) The LOD profile for the ND trait on linkage group 10. The dashed line indicates the significance threshold (3.1). (b) The order and positions of markers in linkage group 10. Marker names and positions are shown above and below the linkage group. The red horizontal bar defines the limits of the QTL, between 30.7 cM and end 43.3 cM. SNPs ss715646341 and ss715647913 flanked the QTL to the left and to the right, respectively.

A dominant marker was developed for the polymorphic region and screened over the mapping population including 46 RD RILs, 31 SD RILs, and 51 ND RILs. The marker was also screened over 36 RD, SD, and ND commercial varieties and breeding lines of pinto beans and cranberry beans. No recombination event was observed for the marker among the individuals in each phenotypic class indicating that the identified polymorphism is most likely to be responsible for the ND trait in common bean.

REFERENCES: Elsadr et al. 2011. Theor Appl Genet 123:1467-1472. Felicetti et al. 2012. Crop Sci 52:1600-1608. McClean et al. 2002. J Hered 93:148-152 Prakken. 1970. Med. Landbouwhogeschool Wageningen 70:1-38

QTL ANALYSIS OF COOKING TIME IN AN ANDEAN RECOMBINANT INBRED LINE POPULATION

Berry M[1], Nchimbi-Msolla S[3], and Cichy K*[1,2]

[1]Michigan State University, Plant Breeding, Genetics, and Biotechnology, [2]Michigan State University, USDA-ARS, Sugarbeet and Bean Research Unit, East Lansing, MI, [3]Sokoine University of Agriculture, Morogoro, Tanzania

Dry bean is a dietary staple in many parts of the world including Africa and Latin America. Beans contain high amounts of fiber but also protein leading it to be called "the poor man's meat" in some parts of the world. Despite its nutritive value, one factor limiting dry bean consumption is the length of time required for cooking. Cooking time in dry bean has been studied for decades, however, research in this area has focused on storage conditions, field conditions, or seed treatment immediately prior to or during cooking, but not on the underlying genetic causes of cooking time differences. This study found eleven quantitative trait loci (QTL) on seven linkage groups involved with cooking time.

The recombinant inbred line (RIL) population used in the QTL analysis was developed using TZ-27 (Incomparable) and TZ-37 (W6 16488) as parents. The RIL population and parental lines were grown in Morogoro and Arusha in Tanzania in 2016 and 2017. Seed was soaked for 12 hrs. and cooked in distilled water with a Mattson (pin drop) cooker (Customized Machining 246 and Hydraulics Co., 247 Winnipeg, Canada). The elapsed cooking time was recorded as the time required for 80% of the pins to pierce the seeds.

Eleven QTL for cooking time were discovered on linkage groups 1, 3, 4, 5, 6, 10, and 11 (Fig. 1). A dearth of information exists for cooking time QTL, but one research group found a QTL ($CT1.1^{CL,TT}$) on linkage group 1 that explained 21.4% of the variation for the trait (Garcia, 2012). $CT1.1^{CL,TT}$ was also found in this work in both years at the Arusha location, and it explained 5.4% of cooking time variation. $CT1.1^{CL,TT}$ contains a gene for magnesium transport, which could effect cooking time as higher levels of magnesium or calcium in the seed has been linked to longer cooking times. On linkage group 6, $CT6.1^{WI}$ explained 14.5% of the variation for cooking time in Arusha during 2017. This QTL contains a gene for wax ester biosynthesis and a gene for sodium transport. Increasing amounts of sodium have been linked to faster cooking times, and heat penetration of the seed during cooking could be effected by the prevelance of wax in the seed coat. Other candidate genes in the remaining QTL include a calcium transporter, pectin lyase, cellulase, and polygalacturonase.

Fine mapping could be performed on these regions to discover which gene in each QTL is effecting cooking time. The QTL can also be used in marker assisted selection so that the time-consuming step of phenotyping for cooking time can be minimized during the early stages of selection. A better understanding of the genetics behind cooking time may lead to greater consumption of beans worldwide.

Garcia, R. A. V., Rangel, P. N., Bassinello, P. Z., Brondani, C., Melo, L. C., Sibov, S. T., & Vianello-Brondani, R. P. (2012). QTL mapping for the cooking time of common beans. *Euphytica*, *186*(3), 779-792.

Table 1. QTL for cooking time for the TT RIL population grown in Morogoro and Arusha in 2016 and 2017.

QTL Name[a]	Year	Location	LOD Max[bc]	r^2(%)[c]	Additive[cd]	Physical QTL Size (kb)	QTL range (cM)
CT1.1^{CL,TT}	2016	Arusha	2.8	5.4	2.8	89.4	26.3 - 34.6
CT1.1^{CL,TT}	2017	Arusha	2.7	4.1	1.45	89.4	30.2 - 37.4
CT1.4^{TT}	2017	Arusha	3.0	4.5	1.53	73.4	20.4-30.2
CT1.5^{TT}	2017	Morogoro	3.1	4.1	2.25	968.7	67.6 – 72.0
CT3.1^{TT}	2017	Morogoro	6.3	10.1	3.5	339.8	27.1 – 29.9
CT3.2^{TT}	2017	Morogoro	10.2	15.3	4.2	2056.7	41.6 – 43.6
CT3.2^{TT}	2017	Arusha	8.0	20.5	-14.3	1895.9	42 – 43.2
CT4.1^{TT}	2017	Arusha	2.9	3.5	1.4	825.8	56.6 – 58.3
CT5.1^{TT}	2017	Arusha	3.3	3.9	1.3	323.7	33.4-35
CT6.1^{TT}	2017	Arusha	9.7	14.5	2.6	353.9	85.6 – 87.5
CT6.2^{TT}	2017	Morogoro	6.9	9.7	3.4	365.9	87 – 89.3
CT10.1^{TT}	2017	Morogoro	4.2	6.3	2.8	1324.6	20.9 - 25
CT10.1^{TT}	2016	Morogoro	3.5	7.8	5.9	1324.6	20.9 - 25
CT11.1^{TT}	2016	Arusha	6.3	15.0	6.2	75.6	21.9 – 23
CT11.2^{TT}	2017	Arusha	3.5	5.1	1.6	404.4	232.5 – 236.5

[a]The first number in the QTL name is the linkage group, and the second number indicates the order of the QTL on the linkage group.
[b]QTL Cartographer was used to perform composite interval mapping on the TT RIL population. LOD thresholds of 2.76 (Arusha, 2016), 2.91 (Morogoro, 2016), 2.55 (Arusha, 2017) and 2.69 (Morogoro, 2017) were determined by running 1000 permutations in QTL cartographer. Composite interval mapping was used to generate QTL regions and Phytozome was used to locate QTL on the *P. vulgaris* genome.
[c]The largest LOD, r^2, and additive score within the QTL were reported.
[d]Positive values indicated the allele was contributed by TZ-27, and negative values indicated TZ-37 contributed the allele.

EVALUATING SPECIALTY SUCCULENT LIMA BEANS (*PHASEOLUS LUNATUS*) AS ALTERNATIVE CROPS IN DELAWARE

Johnson GC* and Ernest EG

Department of Plant and Soil Sciences, University of Delaware (gcjohn@udel.edu)

Lima bean (*Phaseolus lunatus*) is the most widely grown vegetable crop in Delaware. Green seeded baby lima bean types predominate (over 95% of the crop). In addition, three specialty lima beans are also grown on limited acres: a speckled type, a light colored butterpea type, and a large seeded green Fordhook type. These specialty lima beans currently represent less than 5% of the lima beans grown in Delaware. The University of Delaware initiated a breeding program in 2005, focusing on green baby lima beans and green Fordhook types. In the breeding program, diverse lima bean germplasm is used for crosses and a wide variety of colors, forms, and qualities result in the progeny. While the focus of the program is on green types, there is potential for developing and introducing new specialty types adapted to Delaware. We currently have a grant project seeking to identify and evaluate new specialty lima beans from the Delaware breeding program to be grown as succulent lima beans for freezing and canning. In addition, specific selections from US and international germplasm collections are being increased and evaluated.

METHODS

Seed of specialty succulent lima beans already collected or developed as offshoots of the UD green baby lima breeding program have been identified and increased for use in 2018 field trials. This includes three red speckled baby types, two white seeded baby types, 5 multicolored baby types, and one striped Fordhook type.

Seed of 12 additional fixed breeding lines from the UD breeding program not previously evaluated and selected germplasm from USDA germplasm collections have been identified and shipped to Puerto Rico for increase to use in small plot trials in 2018 and further increase. In addition, an additional 16 indeterminate specialty pole types have been identified and will be used for seed increase in 2018. We are also currently seeking additional black seeded types for increase.

A collection of diverse lines maintained by the UD lima bean breading program has been obtained and will be evaluated for cooking and eating characteristics for use in further breeding of specialty limas. These evaluations will be conducted in winter 2017-2018.

To evaluate consumer acceptance of available specialty succulent lima beans from the UD breeding program, each breeding line was grown to a succulent stage in 2017 in 25' row plots, the succulent seed was shelled from pods, and was then blanched and frozen. Initial consumer tests were conducted on December 8, 2017. There were 16 testers in the preliminary test consisting of extension agriculture educators at the University of Delaware. Eight succulent and two dry samples were cooked and then stored under refrigeration for 24 hours. Samples were reheated and labeled randomly from 1-10. A Hedonic ranking test was performed on the following attributes: Overall Appearance, Color, Taste, and Texture. Rankings were 5 Like a lot, 4 Like a little, 3 Neither like/dislike, 2 Dislike a little, 1 Dislike a lot. Tasters were also encouraged to list descriptors including: buttery, nutty, bland, sweet, metallic, or bitter. Results were analyzed using a one-way ANOVA. Frequency analysis will be performed after additional tests. Additional consumer tests will be conducted in January and February 2018.

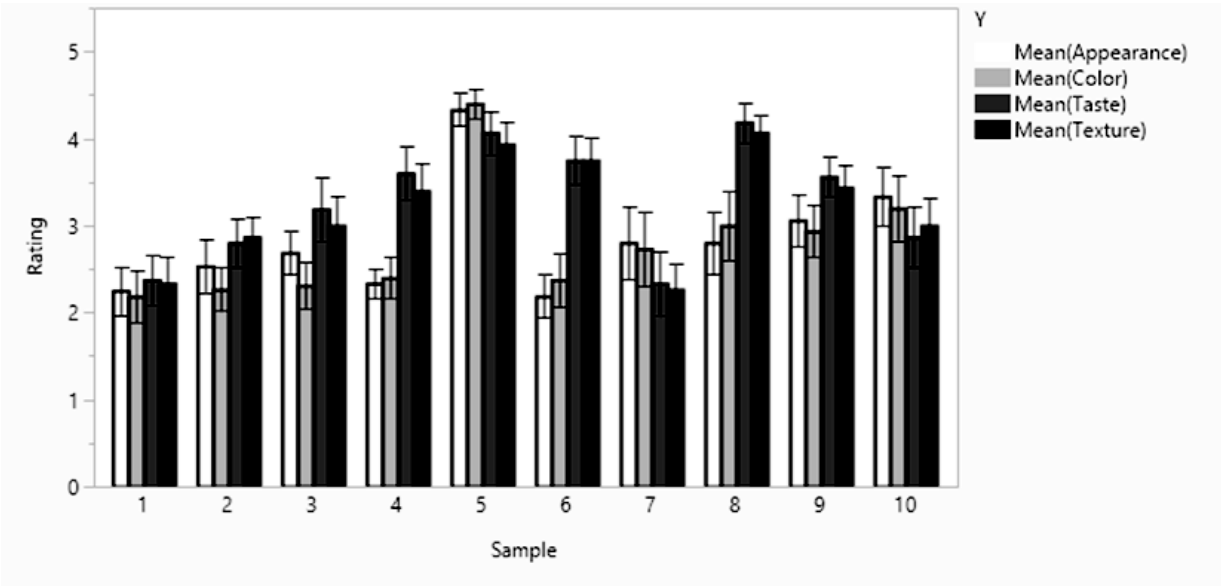
Table 1. Specialty Lima Bean Lines Used for Organoleptic Ratings, Dover, DE, December 8, 2017

Sample	Identification	Succulent	Dry	Cooked Color
1	DE1002303A Speckled	no	yes	brown
2	DE900604A Dark Red	no	yes	brown
3	Cave Dweller	yes	no	light purple
4	Dwarf Florida Butter	yes	no	green purple mix
5	DE0901201B	yes	no	green white mix
6	DE1002303A	yes	no	light purple
7	DE0900604 Dark Red	yes	no	chocolate
8	DE0900604 Light Red	yes	no	light purple green mix
9	DE0801802B	yes	no	cream
10	PA German	yes	no	light purple

PRELIMINARY RESULTS

The specialty lima bean line DE0901201B, which produced a blend of green and white seed when cooked, had the highest overall oganolepic ratings (appearance, color, taste, and texture, Figure 1). Two additional lines DE1002303A (light purple mix when cooked) and DE0900604 Light Red (light purple green mix cooked) had high ratings for taste and texture but lower ratings for overall appearance and color. Dark red and speckled selections that cooked brown to chocolate in color had the lowest ratings for all properties. Additional trials will be conducted to get 120+ total tasters.

Figure 1. Mean Ratings for Four Organoleptic Attributes of Specialty Lima Beans, Dover, DE, 2017



FINE MAPPING OF GENES CONFERRING RESISTANCE TO RUST AND ANTHRACNOSE OF COMMON BEAN

O.P. Hurtado-Gonzales[1], G. Valentini[2], T.A.S. Gilio[2], C. Quigley[1], Q. Song[1], M.C. Gonçalves-Vidigal[2], M.A. Pastor-Corrales[1]

[1]Soybean Genomics and Improvement Laboratory, ARS-USDA, Beltsville, MD 20705,
[2]Universidade Estadual de Maringá, PR, Brazil, Departamento de Agronomia

INTRODUCTION

Rust and anthracnose are major diseases of common bean in the world, especially in the Americas and Africa. Both diseases are caused by pathogens with extensive and shifting virulence diversity. Disease resistance is the most cost-effective strategy to manage these pathogens. Molecular markers are essential tools for marker-assisted selection (MAS); they accelerate the development of cultivars combining various disease resistance genes needed to attain effective and hopefully durable resistance. Several published molecular markers tagging common bean rust and anthracnose resistance genes often yield false positive and false negative results. Additionally, many of these markers are gel-based and labor intensive. There is a need in breeding programs to have molecular markers that are closely linked to the disease resistance genes and that accurately tag the genes of interest. The objective of this study was to use fine mapping to develop DNA markers that are closely linked to rust and anthracnose resistance genes in common bean and that accurately tag these genes when used in MAS.

MATERIALS AND METHODS

The fine mapping approach included accurate phenotyping with specific races of the rust and anthracnose pathogens, inheritance of resistance studies, bulk segregant analysis (BSA) combined with high-throughput genotyping using the SNP chip BARCBEAN6K_3, haplotype identification from resequenced bean lines, and customized SNP marker development. Races of the bean rust pathogen included 15-1, 31-1, 22-6, 31-22, 6-15, and 22-52. Races of the anthracnose pathogen included 73, 2037, and 3481. Crosses between resistant and susceptible cultivars were made for the inheritance of resistance studies. These crosses included Pinto 114 (susceptible to rust) x Aurora (*Ur-3*); Pinto 114 (S) x Mexico 235 (*Ur-3* and unknown rust resistance gene or genes); Early Gallatin (*Ur-4*) x Mexico 309 (*Ur-5*); Rudá (S) x Ouro Negro (*Ur-14* and *Co-3*^{4}); Amendoim Cavalo (resistant parent with new anthracnose resistance gene) x PI 2070262 (S). Segregating F_2 and $F_{2:3}$ populations, the resistant and susceptible parents, and cultivars used as other controls were inoculated with races of the rust and anthracnose pathogens. The BSA was performed using new trifoliate leaves of the resistant and susceptible parents and of F_2 or $F_{2:3}$ populations. Each bulk was generated with DNA from at least eight F_2 or $F_{2:3}$ plants. These were screened with 5,398 single nucleotide polymorphism (SNP) markers in the BARCBEAN6K_3 Illumina BeadChip. The results in the BeadChip were visualized using the Illumina BeadArray Reader and the Genome Studio software was used for allele calling. Allele calls were visually inspected for errors. Additional methodologies were used for developing SSRs and KASPs markers, for target resequencing. Fine mapping of various loci, statistical analysis, linkage and recombination analysis were described in by Hurtado-Gonzales et al. 2017.

RESULTS AND DISCUSSION

Fine mapping determined the precise physical location of several resistance loci used in this study. Specifically, we have conducted the fine mapping of Andean *Ur-4* and Mesoamerican *Ur-3*, *Ur-5*,

and *Ur-14* rust resistance genes (Table 1). In addition, we completed a high-resolution mapping for the Mesoamerican anthracnose resistance locus *Co-3*^{4} and the recently discovered anthracnose resistance gene in the Andean landrace Amendoim Cavalo (Table 1). This gene of Andean origin, provisionally named *Co-AC*, confers broad resistance to highly virulent Mesoamerican races of the anthracnose pathogen. The molecular markers developed using fine mapping will be most useful in MAS to develop bean cultivars combining two or more disease resistance genes. They will also significantly reduce the time and labor associated with current phenotypic detection of these rust and anthracnose disease resistance genes.

Table 1. Fine mapping of four rust and two anthracnose resistance genes in common bean

Resistance gene	***Ur-3***	***Ur-4***	***Ur-5***	***Ur-14***	***Co-3^4***	***Co-AC***
Donor bean genotype	Aurora	Early Gallatin	Mexico 309	Ouro Negro	Ouro Negro	Amendoim Cavalo
Type of resistance reaction[1]	2,2+	2,2+	3,f2	3,f2	1-3	1-3
Number of F_2 plants	129	393	393	178	178	113
Number of $F_{2:3}$ plants	281	570	100	2361	1932	660
Race of the rust or anthracnose pathogens used to phenotype $F_{2:3}$ plants	31-1 (53)	22-52 (108)	31-1 (53)	31-1 (53)	73	3481
Size of region identified using bulk segregant analysis (Mb)	2.1	1.68	0.7	2.0	2.0	1.85
Size of fine mapped genomic region (Kb)	46	20	67	30	87.3	10
Number of candidate genes[2]	6	3	6	2	8	3

[1]Rust and anthracnose phenotype according to scales from Stavely and Pastor-Corrales (1989) and Van Schoonhoven and Pastor-Corrales (1987), respectively. [2]Based on the reference genome of *Phaseolus vulgaris* version 1.0.

REFERENCES

Stavely JR, Pastor-Corrales MA (1989) Rust. In: Schwartz HF, Pastor-Corrales MA (eds) Bean production problems in the tropics. CIAT, Cali, Colombia, pp.159-194

Hurtado-Gonzales, O.P. et al. (2017) Fine mapping of *Ur-3*, a historically important rust resistance locus in common bean. G3 7:557-567.

Valentini, G. et al. (2017). High-resolution mapping reveals linkage between genes in common bean cultivar Ouro Negro conferring resistance to the rust, anthracnose, and angular leaf spot diseases. *TAG* 130:1705-1722.

Van Schoonhoven and Pastor-Corrales (1987) Standard system for the evaluation of bean germplasm. Centro Internacional de Agricultura Tropical (CIAT), Cali, Colombia, 54 p.

WHY WAX BEANS LACK CAROTENOIDS

James R. Myers[1], Joel Davis[1], Haidar Arkwazee[1], Lyle Wallace[1], Rian Lee[2] Samira Mafi Moghaddam[2] and Phil McClean[2]

[1]Oregon State University, Department of Horticulture, Corvallis, OR 97331, [2]North Dakota State University, Department of Plant Science, Fargo, ND

The yellow pod or wax bean trait of *snap* beans was first described in the 19th century then genetically characterized by Currence (1931) who found it was controlled by a single recessive gene designated *y*. This trait was among the genes used to construct the first pre-molecular marker linkage maps in common bean (Bassett 1991). In a map constructed in 1996 with RFLP markers using a wax bean x wild bean cross, *y* was mapped to the distal end of Pv02 (Koinange et al., 1996).

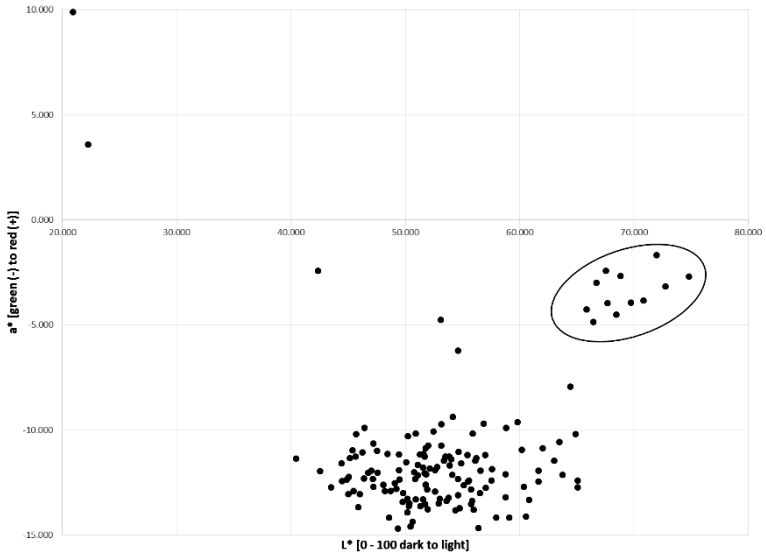


Figure 1. Plot of L* vs. a*for fresh pod color of the Bean CAP Snap Bean Diversity Panel. Points in the circle are wax beans.

During evaluation of the Bean CAP Snap Bean Diversity Panel (SBDP) we observed that wax beans along with Refugee and Romano types had among the lowest amounts of carotenoids in their pod tissues. This panel was also characterized for pod color using the L*a*b* color scale and we found that L* and a* (Fig. 1) provided clean separation of wax beans from all others (mean L* = 73.0 and mean a* = -3.5 v. green bean means of L* = 55.5 and a* = -12.7).

We were able to refine the linkage map location of *y* using two bi-parental populations ('Unidor'/'OSU 5630' and 'Serin'/'OSU 5630'), and through association mapping in the Bean CAP Snap Bean Diversity Panel (SBDP). Two bi-parental populations were classified for pod color as a qualitative trait, and the SBDP had previously been characterized for pod color and carotenoid levels as quantitative traits. The physical position (genome assembly version 1) of SNPs from these three sources bracketed a 532 kb region on the distal end of Pv02 harboring the gene conditioning pod color. This interval contains approximately 43 gene models.

Within this region, we used RNA seq data to search for gene models associated with relatively high expression in young and mature pods compared to expression in vegetative tissues. One gene model (Phvul.002G004400) met these criteria. The sequence corresponds to a pentatricopeptide repeat (PPR) containing protein in Arabidopsis and other organisms. PPR proteins are modular RNA-binding proteins that often control gene expression in both mitochondria and chloroplasts. In the mitochondria, they are associated with fertility restoration of cytoplasmic male sterility. In rice, a null PPR targeted to chloroplasts produces albino seedlings (Su et al., 2012).

Our working hypothesis is that a PPR protein targeted to chloroplasts in pods is responsible for the wax phenotype. Carotenoids accumulate in chloroplasts, and if the null PPR in bean prevents normal plastid ontogeny, then this may account for the low carotenoid levels observed in wax beans. Sequencing of Phvul.002G004400 in six wax bean and eight green bean cultivars revealed two haplotypes (Table 1): one of which corresponded primarily to wax, refugee and Romano types, and the other corresponding to round podded green beans. One exception (Unidor) to this correspondence was observed.

Table 1. Common bean accessions from the Bean CAP Snap Bean Diversity Panel for which Phvul. 002G004400, a putative pentatricopeptide repeat protein, and candidate gene for the wax trait was sequenced.

Accession	Type	Haplotype
Brittle Wax	Wax	1
Kentucky Wonder Wax	Wax	1
Refugee wax	Wax/refugee	1
Roc D'or	Wax	1
Serin	Wax	1
Unidor	Wax	2
Bronco	Green	2
Castano	Green	2
Kentucky Wonder	Green	2
Golden Sands	Green*	2
OSU 5630	Green	2
Renegade	Green	2
Roma II	Romano	1
US Refugee #5	Refugee	1

*Original description is of a wax bean, but when phenotyped, was found to be a green bean.

Further work is needed to validate Phvul.002G004400 as the candidate gene for *y. P. vulgaris* PPR sequence should be transformed and expressed in Arabidopsis or another model plant system to verify expression. An examination of chloroplast ultrastructure would be useful, and a cataloging of haplotypes for this locus may be useful in revealing the organization and expression of this gene.

REFERENCES

Bassett, M.A. 1991. A revised linkage map of common bean. Hortscience 26: 834-836.
Currence, T. M. 1931. A new pod color in snap beans. J. Hered. 22:21–23.
Koinange, E.M.K., Singh, S.P. and Gepts P. 1996. Genetic control of the domestication syndrome in common bean. Crop Sci. 36:1037-1045.
Su, N., Hu, M.L., Wu, D.X., Wu, F.Q., Fei, G.L., Lan, Y., Chen, X.L., Shu, X.L., Zhang, X., Guo, X.P. and Cheng, Z.J. 2012. Disruption of a rice pentatricopeptide repeat protein causes a seedling-specific albino phenotype and its utilization to enhance seed purity in hybrid rice production. Plant Physiol. 159:227-238.

ANTI-OBESOGENIC ACTIVITY OF COMMON BEAN

Henry J. Thompson, John N. McGinley, and Elizabeth S. Neil and Mark A. Brick[2]

Colorado State University, Fort Collins, CO 80523, USA

In developed countries which are at the epicenter of the obesity pandemic, pulse crop consumption is well below recommended levels. In a recent systematic review and meta-analysis of 21 randomized controlled clinical trials, pulse consumption was associated with improved weight control and reduced adiposity, although the underlying mechanisms were a matter of speculation. Common bean (*Phaseolus vulgaris*, L.) is the most widely consumed pulse crop and was the focus of this investigation. Using outbred genetic models of dietary induced obesity resistance (OR) and dietary induced obesity sensitivity (OS) in rat, the impact of bean consumption was investigated on the efficiency with which consumed food was converted to body mass (food efficiency ratio), body fat accumulation, adipocyte morphometrics, and patterns of protein expression associated with lipid metabolism. Cooked whole bean as well as a commercially prepared cooked bean powders were evaluated.

Effect of bean on body weight and visceral fat deposition under ad libitum feeding conditions Two ad libitum feeding studies were conducted. In the first study, cooked whole bean was fed so that 75% of dietary protein was provided by bean and the remainder by casein, an animal based protein source. Bean consumption resulted in rats that were 10.5% lighter. Visceral adipose depots were normalized to tibia length to adjust for differences in body weight as is commonly done in rodent obesity studies. There was a overall difference of 65.3% between total visceral adiposity in rats fed control versus bean containing diet. The greatest reductions in fat pad mass were observed in the parametrial and retroperitoneal fat pads. Since there is a growing trend to increase bean consumption via its incorporation into various food products, commerically available bean powders and cooked whole bean were incorporated into the diet at the same level. The responses of the rats to bean consumption of the two bean products were remarkably similar. There was an average 13.5% difference in body weight and 43.7% difference in visceral adiposity between rats fed bean and the standard diet.

Effect of bean on feed efficiency, visceral fat disposition, and hepatic lipid metabolism under paired-feeding conditions. The ad libitum feeding approach used in the first two experiments indicated that dietary bean had effects on palatibility and/or satiety that impacted weight gain, but such effects could be explained by differences in the bioavailability of calories from a bean containing diet. To evaluate this possibility, we used the commerically available bean powder in a paired-feeding study and evaluated how bean feeding affected the conversion of consumed food to body mass (food efficiency ratio). Contrary to the suggestion of differences in energy bioavailability, the food efficiency ratio was the same for control and bean fed rats whether they were obesity sensitive or obesity resistant. However, despite the fact that both groups had the same weight gain and final body weight, the bean fed rats had reduced visceral adiposity. The magnitude of the difference in total visceral adiposity between control and bean fed rats was 27.8% in the OS rat strain and 43.4% in the OR rat strain. While bean consumption reduced the mass of the three visceral fat pads that were measured, the parametrial and perirenal fat pads were reduced in mass to the greatest extent. To further examine the nature of this effect, we evaluated adipocyte morphmetrics in the parametrial fat pad. Bean fed rats had smaller adipocytes in both OS and OR rats.

Tissue from this experiment was used for mechanistic studies since the effects of bean feeding were independent of differences in body weight gain and food efficiency. A focus on fatty acid

oxidation in liver was prompted by reports that differences in fatty acid oxidation in OR versus OS rats accounted for resistance versus sensitivity to obesity in these rats strains. Analyses were performed using a western blot based immuno-nanocapillary electrophoresis system to quantify differences in protein expression. Four proteins involved in fatty acid oxidation were assessed, acyl CoA synthase (long chain fatty acid isoform 4) (ACSL4), acyl CoA dehydrogenase for long chain fatty acids (ACADL), fatty acid translocase (CD36), and carnitine palmitoyl transferase 1 (CPT1). Bean consumption increased expression of all four proteins with the difference in CPT1 being statistically significant. These same data were subjected to multivariate analysis of variance to assess if the overall capacity for β-oxidation was increased by bean feeding. With adjustment of the model for effects due to animal pairings and differences in food efficiency among pairs, the Hotelling statistic was highly significant for an overall increase in β-oxidation associated with bean feeding ($p = 0.001$). Focusing on CPT1 which is the rate limiting step in β-oxidation and that was significantly different between control and bean fed rats, proteins that regulate CPT1 expression were assessed. Amount of acetyl CoA carboxylase (ACC) and ^{Ser79}pACC (which is inactive) were significantly lower in bean fed rats, a finding consistent with CPT1 induction. Since ACC is regulated by AMP activated protein kinase (AMPK), we also assessed its activity and it was higher in bean fed rats, but the difference from control rats was not statistically significant. We predicted that serum triglycerides would be reduced by bean feeding. *We found that* circulating triglyceride was lower in both bean fed groups and was significantly different from control in the OS rats and circulating triglyceride was positively associated wit*h visceral adiposity (*r = 0.729, $p = 0.0002$). CPT1 is a rate limiting step in β-oxidation and has been suggested to account for difference in obesity sensitivity. To assess this idea, a series of regression analyses were performed. CPT1, which bean feeding induced, was negatively correlated with *plasma triglyceride (*r = -604, $p =$ 0.005) and with total *visceral adiposity (*r = -656, $p = 0.002$). We also assessed liver triglyceride content and found the same pattern of reduction in response to bean feeding as observed with serum triglyceride and hepatic CPT1.

Summary While bean consumption did not affect food efficiency ratio, bean reduced visceral adiposity and adipocyte size in both obesity sensitive and resistant rats. In liver, bean consumption increased carnitine palmitoyl transferase 1, which is the rate limiting step in long chain fatty acid oxidation and also resulted in lower levels of circulating triglycerides. Collectively, our results are consistent with the clinical finding that pulse consumption is anti-obesogenic and indicate that one mechanism by which cooked bean exerts its bioactivity is oxidation of long chain fatty acids.

Pulses such as common bean are neglected staple food crops in many Western societies that are experiencing a pandemic increase the in prevalence of obesity. The WHO sponsored "Year of the Pulse" promoted renewed interest in this highly available and affordable food source. The data reported herein indicate that common bean has specific anti-obesogenic activity which could lessen the impact of obesity on chronic diseases in individuals who are already overweight or obese and potentially reduce the risk of adult weight gain by inhibiting accumulation of lipid in visceral fat depots. Additional work is required to establish the specific mechanisms that account for the effects of common bean on lipid metabolism and to determine if the anti-obesogenic activity of common bean is found in all pulse crops.

TOTAL DIETARY FIBER CONTENT OF DRY BEAN, DRY PEA, CHICKPEA, AND LENTIL CULTIVARS USING AOAC 2011.25 INTEGRATED ASSAY

Chen Y[1], Thompson HJ[1], Brick[2] MA, Vandemark GJ[3], McGee RJ[3], and B. Ogg[2].

[1]Colorado State University, Department of Horticulture and Landscape Architecture
[2]Colorado State University, Department of Soil and Crop Sciences
[3]USDA/ARS, Pullman WA

INTRODUCTION

The human health benefits of dietary fiber (DF) in food crops are well documented. Pulse crops were highlighted in 2016 during the "Year of the Pulse" for their contribution to food security worldwide and health beneficial effect (Brick and Fisher, 2016). However, the consumption of DF in the human diet is lower than recommended, and the gap between actual consumption and recommended intake represents an unrecognized health risk. Many dieticians propose an increase in pulses as a practical way to close the dietary fiber gap. A systematic measurement of the contents of DF and its constituents in pulses has not been conducted since the AOAC 2011.25 method for measuring DF was adopted in 2011. A better understanding of DF content and its components in pulse crops may promote pulse consumption as whole grain and as an ingredient in pulse crop flour.

MATERIALS AND METHODS

Total dietary fiber was measured in 26 cultivars of dry bean (*Phaseolus vulgaris* L.), 11 cultivars of dry pea (*Pisum sativum* L), 13 cultivars of lentil (*Lens culinaris* Medik) and 24 cultivars of chickpea (*Cicer arietinum* L), each grown at two locations. Dietary fiber analysis was determined using the AOAC 2011.25 method as modified by Kleintop et al. (2013).

RESULTS

Mean insoluble dietary fiber (IDF), soluble dietary fiber (SDF), oligosaccharide (OLIGO) and total dietary fiber (TDF) content varied among the four pulse crops (Table 1). Mean TDF content for chickpea was 21.8%, dry bean 25.8%, dry pea 24.7%, and lentil 20%. The range in the components of DF also varied among pulse crops (Table 2). Total dietary fiber among cultivars within crop ranged from 19.5 to 24.9% for chickpea, 24.1 to 27.4% for dry bean, 22.3 to 28% for dry pea, and 18.4 to 21.3% for lentil. Dietary fiber components (IDF, SDF, and TDF) also varied ($P < 0.05$) among cultivars within pulse crops (data not shown). Location effects were not significant for most components of DF. These results indicate that genetic variation occurs among pulse crops and within cultivars of the four pulses. In general, all four pulses had high TDF relative to cereal crops and possessed adequate genetic diversity to improve DF by selection. Consumption of daily servings of any of these pulse crops would reduce the dietary fiber gap that exists today. In fact, one serving per day would supply approximately 25 to 30% of the minimum daily requirement in the human diet. This data will also contribute to a better understanding of the variation for DF in pulse crops when used as whole grains or as ingredients in processed food.

Table 1. Mean insoluble dietary fiber (IDF), soluble dietary fiber (SDF), oligosaccharide (OLIGO), total dietary fiber (TDF) content, and g of total dietary fiber in one serving of pulse crop.

Crop	% IDF[a]	% SDF[a]	% OLIGO[a]	% TDF[a]	TDF/serving g
Chickpea	15.8^{A}	3.5^{A,C}	2.5^{C}	21.8^{B}	7.1
Dry bean	13.9^{B}	7.7^{B}	4.2^{A}	25.8^{A}	8.1
Dry pea	16.2^{A}	3.9^{A,C}	4.6^{A}	24.7^{A}	4.3
Lentil	13.6^{B}	3.1^{A}	3.3^{B}	20.0^{C}	6.0
p-value	<0.001	<0.001	<0.001	<0.001	

[a]Values are expressed as percent of dry weight total. Means with different superscripts within a column are significantly different ($P<0.05$). Abbreviations: OLIGO = raffinose + stachyose + verboscose; TDF = IDF + SDF + OLIGO.

Table 2. Range in percent insoluble dietary fiber (IDF), soluble dietary fiber (SDF), total oligosaccharides (OLIGO), and total dietary fiber (TDF) among four pulse crops grown at two locations.

Crop	% IDF[a]	% SDF[a]	% OLIGO[a]	% TDF[a]
Chickpea	14.4 to 17.1	2.0 to 5.8	1.0 to 3.5	19.5 to 24.9
Dry bean	12.3 to 15.7	5.8 to 9.8	3.6 to 5.2	24.1 to 27.4
Dry pea	14.2 to 19.8	3.3 to 5.3	4.0 to 5.4	22.3 to 28.0
Lentil	12.3 to 4.7	2.7 to 3.9	3.0 to 3.7	18.4 to 21.3

[a]Values are expressed as percent of total seed dry weight. Abbreviations: OLIGO = raffinose + stachyose + verbascose; TDF = IDF + SDF + OLIGO.

REFERENCES:

1. Brick, M., and M. Fisher. 2016. 2016 is the International Year of Pulses. Crops Soils 49:38-38. doi:10.2134/cs2016-49-1-9.

2. Kleintop AE, Echeverria D, Brick LA, Thompson HJ, Brick MA. Adaptation of the AOAC 2011.25 integrated total dietary fiber assay to determine the dietary fiber and oligosaccharide content of dry edible beans. J.Agric.Food Chem. 2013 Oct 9;61(40):9719-26.

3. Chen, Y., R. McGee, G. Vandemark, M. Brick, and H.J. Thompson. 2016. Dietary Fiber Analysis of Four Pulses Using AOAC 2011.25: Implications for Human Health. Nutrients, 8: 829; doi:10.3390/nu8120829.

BEAN IMPROVEMENT IN THE EASTERN AFRICA BEAN CORRIDORS-CHALLENGES AND OPPORTUNITIES

Mukankusi Clare*, Rubyogo Jean Claude, Robin Buruchara, Birachi Eliud, Steve Beebe

International Centre for Tropical agriculture (CIAT)-Pan Africa Bean Research Alliance (PABRA) Corresponding author: c.mukankus@cgiar.org

ABSTRACT

The CIAT breeding program aims to develop elite lines with multiple traits responding to the complex production constraints and market demanded traits of bean clients (producers, consumers and processors) in Africa. The East Africa region is the highest bean producing region in Africa with four of the PABRA member countries (Ethiopia, Kenya, Tanzania and Uganda) being among the leading global producers. Bean breeding in PABRA has been organized to respond to clients' needs using the bean corridor approach. The approach stemmed from studies of the major bean flows in the region. Though the corridor approach offers great opportunities for breeders to reach more users as well as get feedback on their products and in so doing become more market driven, it calls for more timeliness and efficiency in the breeding process. Adoption of more efficient breeding tools is key in addition to tracking of the costs through proper data management.

INTRODUCTION

Greater than >90% of bean trade in East and Southern Africa is informal and >60% of traders do not have access to information on bean prices (Birachi et al., 2017). Intensification of economic development is underway, for example the Southern Agricultural Growth Corridor of Tanzania (SAGCOT). However, persistent systemic failures hinder smallholder involvement across the value chain, e.g. cross-border trade restrictions, low tradable volumes and high transaction costs. The bean corridors which are areas of bean economic intensification are characterized by flow of products from source to destination, all linked up into a network (Birachi et al., 2017). They are characterized by clear production and supply pathways, clusters (institutions) with enabling infrastructure, geopolitics, cultures and preferences. The corridor approach provides context for public and private sector investment, and market support services, clear links between project intervention and intended outcomes at a larger scale (for targeted volumes) and ensures continuous or sustainable flow of products and services across regions. It also helps to better target new varieties while offering opportunity of linking smallholder farmers to major local regional and international markets and stimulates growth of diverse business opportunities in seed, grain, processing, financial products, information among others (Birachi et al., 2017)

EAST AFRICA BEAN CORRIDORS

Red mottled beans, yellow beans and red beans are the most preferred types in these countries with the exception of Ethiopia and Madagascar where white beans rein. Other bean types are still grown though at lower levels. Four bean corridors have been defined; they include; EAREM (East Africa red mottled bean; Kenya, Uganda, Rwanda), YEBECO (yellow bean; W Tanzania, DR. Congo and Burundi), Ethiopia red bean, Ethiopia white bean), and Madagascar white bean (Table 1). Market driven breeding approaches are promoted in the corridors, such as; visioning and Foresight for setting breeding goals, understanding clients' needs, new Variety Design and product profiling, variety development strategy and stage plan, monitoring, evaluation and learning (Persley and Anthony, 2017).

Table 1. Description of corridor mapped for East Africa

Corridor	Driving market class	Countries
1. East Africa Red mottled (EAREM)	Red mottled beans	Kenya, Rwanda, Uganda, N. Tanzania
2. YEBECO (Yellow bean corridor)	Yellow bean	Western Tanzania, Burundi, Congo
3. Ethiopia white bean	White pea bean	Ethiopia
4. Madagascar white bean	White bean	Madagascar
5. Ethiopia red bean	Red bean	Ethiopia

Source: Birachi et al, 2017

Fig 1. Major bean flows in East Africa

What are the implications to the African Bean Breeder?

Capacity building and targeted research is conducted to respond to the identified priority of each corridor. The involvement of multiple stakeholders along the bean value chain at all stages of the breeding process is being re-enforced with more involvement of specifically the private sector to help drive the business of bean breeding. Positive results have been noted, such as the inclusion of untraditional traits in breeding pipelines such as short cooking time, canning quality and Fe and Zn content that are demanded by the market in addition to resilience traits. Enhanced interest in beans as a commercial crop is leading to development of financial products such as crop insurance to respond to the private sector needs. However, with this approach, public bean breeding programs will need to enhance their timeliness in releasing high quality market demanded varieties that have ready market to be able to meet demands of the business. The efficiency (time, cost and quality) of the breeding process (phenotyping, genotyping), tackling un-traditional traits (seed bronzing, canning, pod quality, taste and flavor, flatulence etc.) in addition to producing the required volumes and quality (Atlin et al., 2017) will be key. Modern data management to track progress and tracking the cost and efficiency of breeding program will be paramount. However, the need to maintain genetic diversity while responding to market demands may be challenging but should be part and parcel of the breeding tasks.

REFERENCES

Persley, G.J. and Anthony V.M. (Eds) (2017). The Business of Plant Breeding. Market-led Approaches to New Variety Design in Africa. ISBN-13: 9781786393814

Atlin, G.N., Cairns, J.E., Das, B. 2017. Rapid breeding and varietal replacement are critical to adaptation of cropping systems in the developing world to climate change. Global Food Security 12 (2017): 31-37

Birachi, E. Buruchara, R., Odhiambo, C., Kalemera, S., and J. C., Rubyogo, 2017. Bean corridors: A novel approach to scale up national and regional trade in Africa. https://cgspace.cgiar.org/bitstream/handle/10568/80540/PABRA20_Bean_Corridors_BRIEF.pdf?sequence=5

GENETIC DIVERSITY OF THE GUATEMALAN CLIMBING BEAN COLLECTIONS

Tobar-Piñón MG[1,3*], Moghaddam SM[2], Lee RK[1], Villatoro-Merida JC[3], Osorno JM[1], and McClean PE[1]

[1] Department of Plant Sciences, North Dakota State University, Fargo ND (gabriela.tobarpinon@ndsu.edu)
[2] Department of Plant Biology, Michigan State University, East Lansing, MI
[3] Instituto de Ciencia y Tecnología Agrícolas (ICTA), Guatemala.

INTRODUCTION

Since common bean is the most important legume crop for human consumption around the world, bean breeders are challenged to increase the production of beans while facing new problems like climate change. Guatemalan climbing beans have been suggested to represent race Guatemala, a newly identified race in the Middle American gene pool that may represent an untapped source of alleles for bean improvement. This study confirmed the existence of race Guatemala in the Middle American gene pool and its differentiation from other races. The low population structure found within these Guatemalan beans also makes this population ideal for the discovery of candidate genes for important traits. We demonstrate that the Guatemalan population was useful to provide candidate genes for previously reported genetic factors like the *V* gene for flower color, and the *Asp* gene for seed coat luster. The important relationship between flowering time and altitudinal adaptation of beans was also emphasized.

MATERIALS AND METHODS

Population structure was analyzed using ~78,000 SNPs and 629 accessions from the collections GUA_1966-82 and GUA_2015 of Guatemalan climbing beans (*P. vulgaris*) using the Software STRUCTURE, principal components analysis and a maximum likelihood tree. Accessions from races Mesoamerica, Durango-Jalisco (Mesoamerican Diversity Panel) and Nueva Granada (Andean Diversity Panel) were used for comparison. Also, expected heterozygosity (*He*) and polymorphic information content (PIC) were calculated using Power Marker. Intra accession diversity was calculated using InDel markers. GWAS analysis for 19 traits of economic/agronomic importance were evaluated in both collections using GAPIT with four different models for controlling structure and relatedness.

RESULTS AND DISCUSSION

The genetic diversity and population structure analysis of the Guatemalan climbing bean collections allowed the confirmation of race Guatemala in the Middle American gene pool and its differentiation from race Mesoamerica and race Durango-Jalisco (Figure 1). Therefore, it represents a new source of alleles for breeding programs. It is recommended that seeds of the Guatemala populations GUA_1966-82 and GUA_2015 should be retained in the germplasm bank both among and within accessions. Single seed descendant is recommended before starting the pre-breeding process. Lower population structure was found within the Guatemalan climbing beans, this makes the population ideal for the discovery of genetic factors and candidate genes for important traits In this study, we found genomics regions associated to flower color in Pv06 (Figure 2), pod color in Pv08 and seed coat luster in Pv07. We also found that flowering time is an important factor for the altitudinal adaptation of the Guatemalan climbing beans.

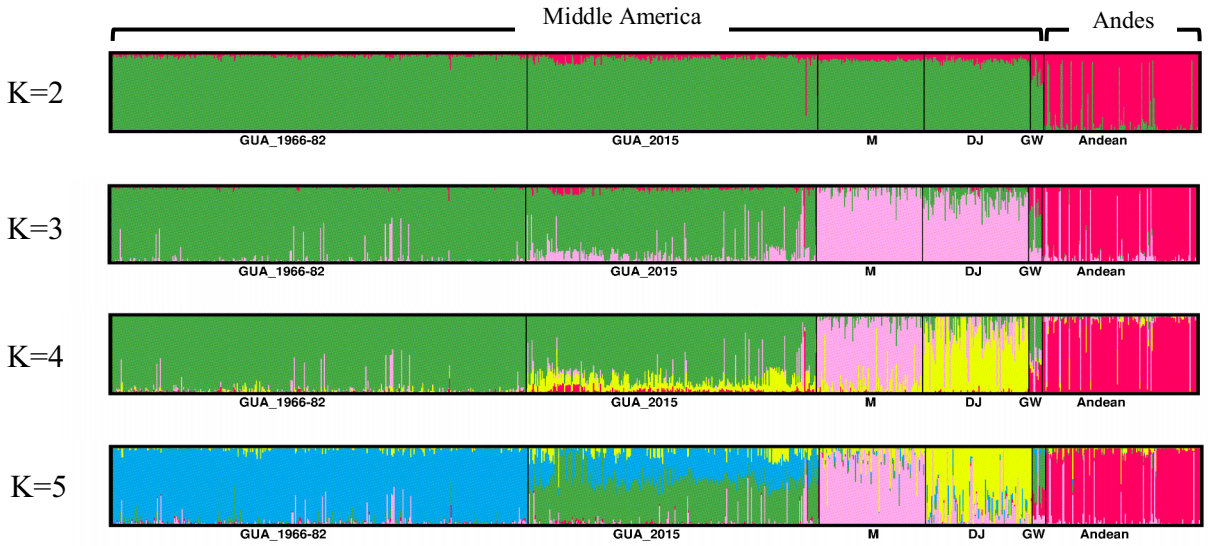


Figure 1. Results of population structure analysis with K values ranging from 2 to 5. M= race Mesoamerica, DJ= race Durango_Jalisco, GW= Guatemalan wilds.

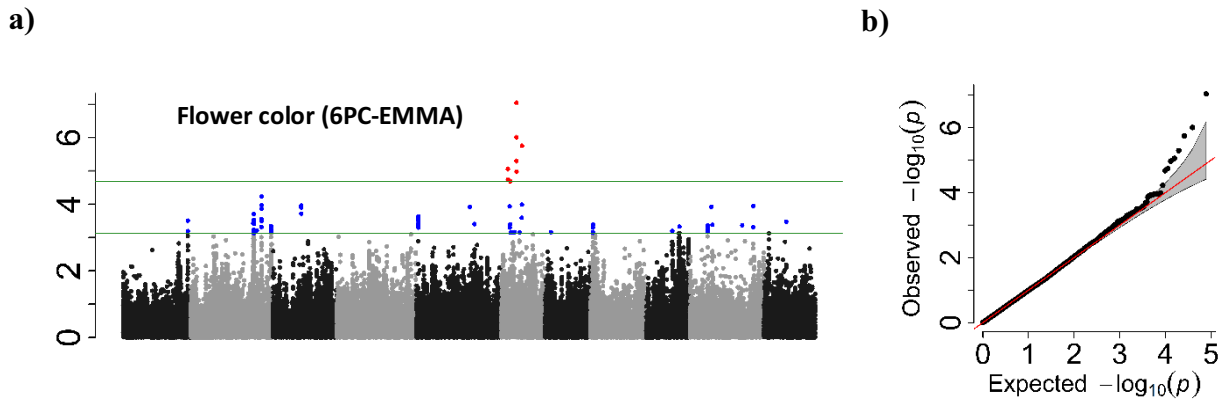


Figure 2. a) Manhattan plot for flower color of GUA_2015. b) quantile-quantile plot for the best model. Green lines represent the cutoff values for 0.1 and 0.01 percentiles. Markers significant for the 0.01 and 0.1 percentiles are colored in red and blue, respectively. Best model is indicated in parenthesis.

REFERENCES

Beebe, S., P.W. Skroch, J. Tohme, M.C. Duque, F. Pedraza, and J. Nienhuis. 2000. Structure of genetic diversity among common bean landraces of Middle American origin based on correspondence analysis of RAPD. Crop Sci. 40: 264–273.

IDENTIFICATION OF VIRUS INFECTING CULTIVATED AND WILD PHASEOLUS IN THE CENTRAL-WEST REGION OF MEXICO

Chiquito-Almanza E[1*], Caballero-Pérez J[2], Acosta-Gallegos JA[3], Guevara-Olvera L[1], Cuellar W[4], and Anaya-López JL[3]

[1] Instituto Tecnológico de Celaya, Department of Biochemestry, Molecular Biology Lab.
[2] Cinvestav, Langebio U.G.A., Molecular and Developmental Complexity Lab.
[3] CEBAJ-INIFAP, Bean and Chickpea Breeding Program (anaya.jose@inifap.gob.mx).
[4] Centro Internacional de Agricultura Tropical, Virology Unit.

INTRODUCTION

Virus diseases are a significant threat to agriculture production. Common bean (*Phaseolus vulgaris* L.) is likely the most susceptible plant species to virus infection in the *Leguminosae* (Morales, 2006) and one of the most widely cultivated legumes in the world (FAOSTAT, 2017). In Latin America, at least 20 virus species transmitted by different vectors cause diseases of economic importance on common bean (Morales and Castaño, 2008). In dry bean, common mosaic and top necrosis, caused by *Bean common mosaic virus* (BCMV) and *Bean common mosaic necrosis virus* (BCMNV), respectively, are frequent. Both viruses are transmitted in a non-persistent form by several species of aphids, but the most efficient mean of dissemination and above all for long distances is via seed, where it remains latent for more than 30 years (Sastry *et al*., 2013). Recently in Mexico a high presence from both viruses in almost all bean-producing states has been detected (Flores-Esteves *et al*., 2003; Lepe-Soltero *et al*., 2012); nevertheless, in these studies 52 % of the evaluated symptomatic samples, were negative to presence from BCMV and/or BCMNV. These results suggest that there are other viruses causing similar symptoms as those elicited by BCMV and BCMNV in common beans. The aim was to identify the viruses infecting bean plants and its distribution, in the Mexican states of Guanajuato, Jalisco and Nayarit.

MATERIAL AND METHODS

During 2013-2015 leaf samples from wild *Phaseolus* spp (WB) and cultivated common bean *Phaseolus vulgaris* (CB) plants with and without symptoms of virus damage were collected in the states of Guanajuato, Jalisco and Nayarit, Mexico. The identification of the virus infecting the samples was performed by high throughput sequencing and assembly of total small RNAs (small RNA sequencing and assembly; sRSA). Total RNA from different CB and WB samples was used to construct 53 sRNA libraries: 26 from CB (*P. vulgaris*), and 27 from WB that included samples from *P. vulgaris*, *P. coccineus* and *P. leptostachyus*.

RESULTS AND DISCUSSION

In the libraries from CB we identified six virus species: BCMV, BCMNV, *Cowpea mild mottle virus* (CPMMV), *Phaseolus vulgaris endornavirus 1* (PvEV-1), *Phaseolus vulgaris endornavirus 2* (PvEV-2), and *Bean golden yellow mosaic virus* (BGYMV). Likewise, in the libraries of WB four virus were identified: BCMV, BCMNV, PvEV-1 and *Peanut mottle virus* (PeMoV). All identified viruses were confirmed by PCR or RT-PCR. While in all libraries of CB viral sequences were identified, only six libraries of WB displayed these sequences. In total, nine complete genomes and 36 partial genomes of the identified viral species were reconstituted throughout alignment with a related virus genomes. These sequences and the virus isolates will be useful for understanding viral genomic diversity and their biological characteristics. With the exception of BGYMV, all the virus species identified are seed borne. Due to controversies with respect to seed

transmission of CPMMV, studies are needed to assess the phytosanitary risk of this virus. BCMV and BCMNV represent high phytosanitary risk due to a high rate of transmissibility; whereas PvEV-1 and PvEV-2 are considered non-pathogenic, and PeMoV with a transmissibility rate below 3%, represent low risk. The abundance of these viruses in cultivated beans suggests the absence of genetic resistance or varietal deterioration combined with the repeated use of grain as seed; thus, among other traits, new cultivars must include resistance against these viruses, and need to be grown with production practices that include the use of clean seed and the control of vectors.

AKNOWLEDGEMENTS

This work was supported by CONACYT (Mexico) (PDCPN 2013-1 No. 214641 and a doctoral fellowship to E. Chiquito-Almanza 226668) and INIFAP (Mexico) (10242732533).

REFERENCES

FAOSTAT (Food and Agriculture Organization of the United Nations) Available online: http://www.fao.org/faostat/es/#data/QC (accessed on Sep 2017).

Flores-Estéves, N., Acosta-Gallegos, J.A., Silva-Rosales, L. 2003. Bean common mosaic and Bean common mosaic necrosis virus in Mexico. Plant Dis. 87:21-5.

Lepe-Soltero, D., Sánchez-García, B.M., Jiménez-Hernández, Y., Salinas-Perez, R.A., García-Neria, M.A., González de León, D., Becerra-Leor, N.E., Acosta-Gallegos J.A., Silva-Rosales L. 2012. Presence of BCMV and BCMNV in five dry bean-producing states in Mexico. Tropical and Subtropical Agroecosystems. 15: 313-321.

Morales, J.F. Common beans. In Natural resistance mechanisms of plants to viruses, 1st ed.; Loebenstein, G.; Carr, J.P., Eds.; Springer: Netherlands, 2006, pp. 367-382.

Morales, J.F., Castaño, M. Enfermedades virales del frijol común en América Latina. Ed: Morales, F.J.; Escobar-Correa F.J., 2008, pp. 7-83.

Sastry, K.S. Plant virus and viroid diseases in the tropics. Volume 1. Introduction of plant viruses and sub-viral agents, classification, assessment of loss, transmission and diagnosis. Springer: Netherlands, 2013, pp. 109-189.

SPATIAL AND TEMPORAL SCALES OF RANGE EXPANSION IN WILD *PHASEOLUS VULGARIS*

Andrea Ariani, Jorge Carlos Berny Mier y Teran, and Paul Gepts

Department of Plant Sciences, University of California Davis

Among the five domesticated species, two species – *Phaseolus vulgaris* (common bean) and *P. lunatus* (lima bean) – have wild progenitors with an extraordinarily wide distribution, from Mexico in the northern hemisphere to Argentina in the southern hemisphere, covering a distance of some 10,000 km. This distribution is not continuous but is marked by several gaps due to low or high altitude and/or high and low temperature or humidity, respectively. Examples of such gaps are the Isthmuses of Tehuantepec and Panama and the switch between the eastern Andes (Colombia and Venezuela in the northern Andes; southern Peru, Bolivia, and northwestern Argentina in the southern Andes) and western Andes (Ecuador and northern Peru).

The question is therefore: How and when did such a broad distribution of the wild ancestor come into being, given that the genus *Phaseolus* originated in Mexico (Delgado Salinas et al. 2006)? Such a question raises ancillary questions such as the mode of dispersal (discrete events or a gradual spread) and the vector assuring this dispersal?

To answer these questions, a genomic approach was initiate to study the DNA diversity in a large sample of wild *P. vulgaris* (n = 246), including 157 genotypes of the Mesoamerican (including wild populations in Mexico, Central America and Colombia MW), 77 of the southern Andean (including populations in southern Peru, Bolivia, and Argentina, AW), and 12 of the Northern Peru-Ecuador (PhI) gene pools. A single plant per accession was analyzed by genotyping-by-sequencing (GBS) using the pipeline developed by Ariani et al. (2016). This pipeline uses the restriction enzyme *Cvi*AII for the GBS procedure as it provides a larger number of restriction fragments (close to the number of predicted genes in the genome) and a better genome distribution, including the pericentro-meric areas of each chromoso-me (Ariani et al. 2016), compared to the standard enzyme *Ape*KI.

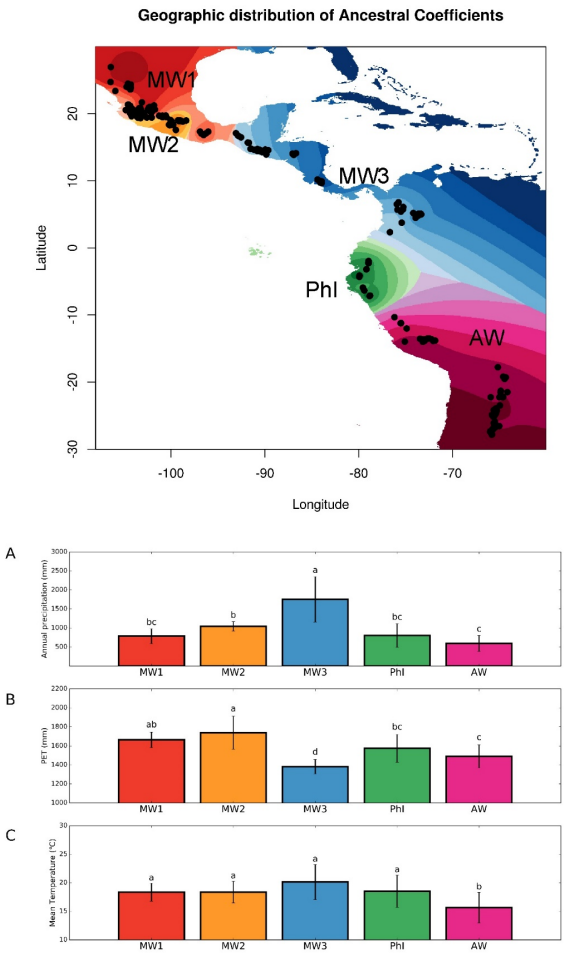
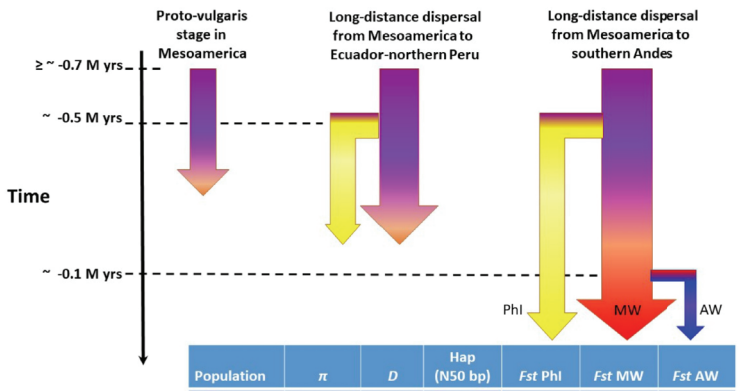


Figure 1. Top: Geographic distribution of five populations identified by TESS3 among wild *Phaseolus vulgaris* for K = 5. Bottom: Average climate variables for K = 5 populations (same color as in top graph): A: Precicipitation; B: Potential evapo-transpiration; C: Temperature.



Population	π	D	Hap (N50 bp)	Fst PhI	Fst MW	Fst AW
MW	1.42e-5	1.52	951 (246)	0.30	0.00	0.34
PhI	1.04e-5	0.28	27 (694)	0.00	0.30	0.62
AW	6.46e-6	-0.45	245 (921)	0.62	0.34	0.00

Figure 2. Phylogeny of major wild *Phaseolus vulgaris* gene pools, comparative genetic diversity and distance, and ages of major long-distance dispersal events. AW: southern Andean wild; MW: Mesoamerican wild; PhI: Ecuador-northern Peru. *Π*, nucleotide diversity; *D*, Tajima's *D* statistic; *Fst* population differentiation. Hap, number of haplotype blocks; N50, median block length. Width of arrows are approximately proportional to nucleotide diversity.

Results of diversity analyses show the same overall pattern of population divergence obtained previously with markers other than SNPs, such as phaseolin (Kami et al. 1995), allozymes (Koenig and Gepts 1989), RFLP (Becerra Velasquez & Gepts 1994), RAPD (Freyre et al. 1996), AFLP (Tohme et al. 1996), SSRs (Kwak and Gepts 2009), an DNA sequencing (Rendón-Anaya et al. 2017b). A population structure analysis using the TESS3 program identified five populations: three Mesoamerican ones (MW1-3), one southern Andean (AW), and one for the intermediate group (PhI) (Fig. 1). Whereas the MW gene pool (MW1 + MW2 + MW3) had the highest nucleotide diversity (Fig. 2), the PhI gene pool was a close second, especially considering its much smaller size. The AW gene pool had the lowest nucleotide diversity, consistent with earlier observations (Schmutz et al. 2014). The lower diversity of the Andean gene pools (PhI and AW) is consistent with the narrowness of the wild bean habitat there leading to a strong correlation (r = -0.9) with latitude, as well as their origin in rare dispersal events from the Mesoamerican core distribution. In contrast, the Mesoamerican area shows both latitudinal and longitudinal gradients, consistent with a much larger overall population size compared to the Andean populations. The dispersal events led to changes in climatic environment: e.g., MW3 is distributed in a warmer, more humid environment, where AW is located in a drier environment, but with lower potential evapotranspiration (Fig. 1), predictive of the adaptation of the Andean domesticates.

The rare dispersal events leading to the Andean gene pools were probably mediated by migratory birds, the most likely animal vector capable of bridging the significant gaps in the wild distribution. In Ecuador, wild bean populations are named as "dove "or "pigeon" bean (Debouck et al. 1993). Several dove species have distributions overlapping that of wild beans (DeGraaf & Rappole 1995). These rare long-distance dispersal events are thought to have taken place some 0.5 Myrs (PhI) and 0.1 Myrs (AW) ago (Ariani et al. 2017). The distinctness of the PhI population at the DNA sequence and metabolomic levels (Kami et al. 1995; Delgado-Salinas et al. 1999; Chacón et al. 2007; Rendón-Anaya et al. 2017b) has led to the naming of the PhI group as *Phaseolus debouckii*, a cryptic sister species of *P. vulgaris* (Rendón-Anaya et al. 2017a).

REFERENCES: Ariani et al. (2016) Molec Breed 36: 87; Ariani et al. (2017) Molec Biol Evol: msx273; Becerra Velásquez & Gepts (1994) Genome 37: 256; Chacón et al. (2007) Plant Syst Evol 266: 175; Debouck et al. (1993) Econ Bot 47: 408; Delgado-Salinas et al. (2006) Syst Bot 31: 779; Delgado-Salinas et al. (1999) Syst Bot 24: 438; Freyre (1996) Econ Bot 50: 195; Kami et al. (1995) Proc Natl Acad Sci USA 92: 1101; Koenig & Gepts (1989) Theor Appl Genet 78: 809; Kwak & Gepts (2009) Theor Appl Genet 118: 979; Rendón-Anaya et al. (2017a) Phytotaxa 313: 259; Rendón-Anaya M et al. (2017b) Genome Biology 18: 60; Tohme et al. (1996) Crop Sci 36: 1375.

ADVANCED INTERSPECIFIC HYBRIDS OF COMMON BEAN & TEPARY BEAN WITHOUT EMBRYO RESCUE

Santos Barrera, Roosevelt Escobar, and Stephen E. Beebe

International Center for Tropical Agriculture (CIAT) (s.y.barrera@cgiar.org)

INTRODUCTION: Tepary beans have an array of traits that confer adaptation to hot and dry environments that is superior to common bean. However, crosses with tepary beans present multiple problems and incompatibility barriers due to their phylogenetic distance from *P. vulgaris*. Therefore, hybrids require numerous pollinations and embryo rescue and consecutive backcrosses to obtain viable plants. Improving the efficiency of hybridization between common bean and tepary bean, will allow accessing more diversity of tepary bean and transferring important traits to common bean. This poster presents the progress in obtaining F_1 of common bean and tepary bean without embryo rescue.

MATERIAL AND METHODS: 27 interspecific crosses were made between thirteen Mesoamerican common bean lines as the female parent and eleven tepary accessions as the male parent, selected from a heat-tolerance screening at the Colombian coast (Table 1). 183 embryos were rescued and cultivated using a modification to the protocol published by Mejia-Jimenez et al. (1994). F_1 plants were backcrossed with pollen from common bean lines (BC_1). The embryos from the BC_1 were rescued using the abovementioned protocol, which yielded seven BC_1 offspring. Each were again backcrossed with pollen from common bean lines, resulting in one hundred and thirty-two BC_2F_1 individuals. No rescuing was required at this stage. The populations advanced until BC_2F_4. One cross between an interspecific tepary-common bean line and *P. parvifolius* resulted in a bridging parent (Fig. 1). We obtained advanced interspecific hybrids without embryo rescue between the bridging parent and different tepary beans (Table 2), in two consecutive generations.

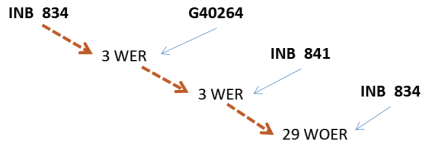


Figure 1. Bridging parent crossing scheme,
WER=with embryo rescue, WOER= without embryo rescue

Table 1. Parent involved in crosses

Common bean lines		Tepary bean germoplasm		
Identification	Justification	Identification	specie	Form
(INB 834xSMR 139)F1	Hybrid	G 40001	*P. acutifolius*	cultivated
ICTA LIGERO	Golden mosaic virus resistant	G 40022	*P. acutifolius*	cultivated
INB 834	Interspecific line (with tepary) Good architecture, early	G 40027	*P. acutifolius*	cultivated
INB 841	Interspecific line (with tepary) heat tolerance, synchronization, good architecture, early	G 40028	*P. acutifolius*	cultivated
SEF 60	Good for heat tolerance	G 40045	*P. acutifolius*	wild
SEF 10	Heat tolerance, good architecture, golden mosaic virus resistant	G 40143	*P. acutifolius*	cultivated
SEN 56	Good for drought	G 40261	*P. acutifolius* var. tenuifolius	cultivated
SEN 97	Good for heat tolerance	G 40264	*P. parvifolius*	wild
SEN 118	Golden mosaic virus resistant	G 40161	*P. acutifolius*	cultivated
SMC 214	Biofortification	G 40279	*P. acutifolius*	cultivated
SMN 57	Biofortification	G 40237A	*P. acutifolius*	cultivated
SMR 132	Biofortification			
SMR 139	Biofortification, Common mosaic virus resistant			
SMR 155	Biofortification, Golden mosaic virus resistant			

Table 2. Tepary lines involved in crosses with bridging parent

Identification	FLCOL	GH	PSC	SSC	SCP	SDBR	SSZ	Source	Form	Country	State	Altitude	specie	Justification	Literature backup
G 40264	5	3B	2	9	M	2	1	CIAT	wild				*P. parvifolius*	Good for high temperature (Colombian Coast)	
G 40068	1	3B	3		0	1	2	CIAT	cultivated	USA	Arizona	544	*P. acutifolius*	High yield and good in drought, good in high temperature	Rao et al. (2013)
G 40111	5	3B	8		0	1	1	CIAT	cultivated	Mexico	Campeche	50	*P. acutifolius*	Strange distribution, low altitude. Good for high temperature (Evaluation 2017	
G 40119	5	3B	8		0	2	1	CIAT	cultivated	Mexico	Oaxaca	1644	*P. acutifolius*	Resistance to the leafhopper Empoasca	Garvin & Norman 1994
G 40036	5	2B	8		0	3	1	CIAT	cultivated	Mexico	Oaxaca	1644	*P. acutifolius*	Resistance to the leafhopper Empoasca	Garvin & Norman 1994
G 40019	5	2B	8		0	3	1	CIAT	cultivated	Mexico	Oaxaca	1644	*P. acutifolius*	Resistance to the leafhopper Empoasca	Garvin & Norman 1994
TEP 22	1	2B	1		0	2	2	Tim Porch	Improved				*P. acutifolius*	Resistant to rust, bacterial blight and seed weevil	Porch et al. (2013)
TEP 23	5	2B	8		0	1	2	Tim Porch	Improved				*P. acutifolius*	Best for drought UCDAVIS 2016	
G 40287	5		2	8	J	1	1	CIAT	wild	Mexico	Sonora		*P. acutifolius*	Good for high temperature >28 °C (Evaluation 2017 Green House CIAT)	
G 40056	5		2	8	J	2	1	CIAT	wild	Mexico	Sonora		*P. acutifolius*	Good for high temperature >28 °C (Evaluation 2017 Green House CIAT)	
G 40161	5		2		0	2	1	CIAT	cultivated	Mexico	Sonora	250	*P. acutifolius*	intermediate for heat tolerance	
G 40279	5		1		0	1	1	CIAT	cultivated	Mexico	Sonora	330	*P. acutifolius*	intermediate for heat tolerance	
G 40237A	1		5		0	1	1	CIAT	cultivated	Mexico	Saltillo		*P. acutifolius*	intermediate for heat tolerance	

RESULTS AND DISCUSSION

Hybridization between common bean and tepary bean it was possible to get only 6 crosses from 24 combinations. However, from all pollinations (318), only 14.7% resulted in small pods. 161 embryos between *P. vulgaris x P. acutifolius* were rescued and only resulted in 12% of F_1 plants. 10 embryos between *P. vulgaris x P. parvifolius* were rescued and only 30% of F_1 plants resulted (Table 3). No crosses allowed obtaining seeds at this stage, as it was only possible to get mature seeds doing two consecutive backcrosses towards common bean (F_1BC_2). The F_1 plants were sterile as evidence of their hybridity, no viable F_2 plants could be obtained. Most of the pollinations were not effective, the flowers abscised at a very early stage, or the pods were without seeds (Fig. 2). The results confirmed that hybridization between common bean and tepary bean is difficult, and embryo rescue is necessary to obtain successful hybrids. Similar results were published by Mok et al. (1978); Thomas & Waines, (1984); Pratt et al. (1985) and Souter et al. (2017). The crosses between the bridging parent and tepary bean permitted us to obtain 22 different combinations. In 70% of pollinations (198) mature pods and mature seeds could

be obtained (Fig. 3), and 80% of interspecific hybrid seeds were viable obtaining 243 F_1 plants (Table 4). As with the hybrids obtained by embryo rescue, a high degree of sterility of hybrids was present, however, when the F_1 was again crossed with another common bean line as a male, we could be obtained mature pods and seeds recovering the fertility in this stage (Fig. 4). While the hybridization between common bean and tepary bean was deficient, the use of the bridging parent improved crossability between the species (Fig. 5).

Table 3. Hybridization between common bean and tepary bean

Female parent x male parent	Number of crosses	Number of pollinations	Number of pods	Number of embryos cultured	Number of embryos per pollinations (%)	Number of F_1's Obtained	Number of F_1's per pollinations (%)	Number of F_1's per embryo (%)
P. vulgaris x *P. acutifolius*	23	278	40	161	57.9	20	7	12
P. vulgaris x *P. acutifolius* var. tenuifolius	1	20	4	12	60.0	0	0	0
P. vulgaris x *P. parvifolius*	1	20	3	10	50.0	3	15	30

Table 4. Hybridization between a bridging parent and tepary bean

Female parent x male parent	Number of crosses	Number of pollinations	Number of pods	Number of mature seeds	Number of F_1's Obtained	Number of F_1's per seeds (%)
Bridging parent x *P. acutifolius*	22	198	139	302	243	80

Figure 2. Flowers abscised at a very early stage and pods without seeds, from crosses between tepary and common bean

Figure 3. Mature pods and seeds from hybridization between bridging parent and tepary bean

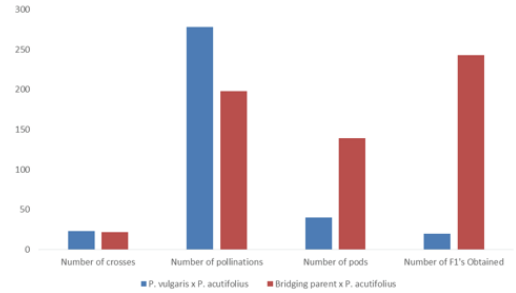


Figure 5. Crossability between common bean and tepary bean; 23 crosses *P. vulgaris* x *P acutifolius*; 22 crosses bridging parent x *P. acutifolius*

Figure 4. Greenhouse-grown plants: fertile bridging parent (left); an interspecific hybrid (bridging parent X G 40019) obtained by WOER showing sterility (center); and an interspecific fertile hybrid (F_1 X 'SEF 10') obtained by WOER (right).

CONCLUSIONS: Several researchers have performed crosses of common bean x tepary bean. However, most breeders have been doing embryo rescue to yield viable interspecific hybrids. Therefore, this result which shows successful hybridization without embryo rescue through a bridging parent opens up the possibility that breeders develop new interspecific hybrids, transferring important economic traits of tepary bean to common bean.

REFERENCES

Mok, D. W. S., Mok, M. C., & Rabakoarihanta, A. (1978). Theoretical and Applied Genetics, 52(5), 209–215.
Mejia-Jimenez, A., Muñoz, C., Jacobsen, H. J., Roca, W., & Singh, S. P. (1994). Applied Genetics, 88, 324–331.
Pratt, R. C., Bressan, R. A., & Hasegawa, P. M. (1985). Euphytica, 34(2), 329–334.
Souter, J. R., Gurusamy, V., Porch, T. G., & Bett, K. E. (2017). Crop Science, 57(3), 1160–1171.
Thomas, C. V., & Waines, J. G. (1984). J. Hered., 75(2), 93–98.

SOURCES OF RESISTANCE TO *Colletotrichum lindemuthianum* IN COMMON BEAN LANDRACES FROM BRAZIL

Souza VB[1*], Gonçalves-Vidigal MC[1], Vidigal Filho PS[1], Gilio TAS[1], Valentini G[1], Mindo NNA[2], Calvi AC[1]

[1]Departamento de Agronomia, Universidade Estadual de Maringá, PR, Brasil. [2]Associação Nacional de Extensão Rural, Nampula, Mozambique. [*]E-mail: vanetbatista@yahoo.com.br.

INTRODUCTION

The anthracnose caused by *Colletotrichum lindemuthianum* is one of the main diseases that occur in the common bean crop. The use of resistant cultivars is characterized as one of the most efficient and economic alternatives to control this disease (Mahuku et al. 2002). Andean and Mesoamerican bean genotypes from Paraná State evaluated by Vidigal Filho et al. (2007) presented genetically highly variable in response to Mesoamerican and Andean races of *C. lindemuthianum*. Given the importance of identifying new anthracnose resistance sources for greater disease control efficiency, this study had as objective to characterize a set of common bean landraces from the Germplasm Bank of Núcleo de Pesquisa Aplicada a Agricultura (Nupagri) for their resistance to different races of *C. lindemuthianum.*

MATERIAL AND METHODS

A total of 85 common bean accessions from Germplasm Bank of Núcleo de Pesquisa Aplicada a Agricultura (Nupagri), Universidade Estadual de Maringá were evaluated with races 65, 73 and 3481 of *C. lindemuthianum*. Fourteen-days-old common bean seedlings were inoculated with a concentration of 1.2×10^6 conidia mL^{-1} of distilled water. The plants were incubated and maintained in a mist chamber with temperature of 20 ± 2^oC and relative humidity of >95%. Plants with disease reaction score between 1 and 3 were considered resistant, whereas plants with scores from 4 to 9 were considered susceptible (Pastor-Corrales et al. 1995). A pathogenicity index (PI) for each *C. lindemuthianum* race was computed by dividing the number of bean landraces with a susceptible reaction by 85 (Balardin et al. 1997; Vidigal Filho et al. 2007).

RESULTS AND DISCUSSION

A total of 31 landraces were resistant to race 65 (21 Andean and 10 Mesoamerican). For race 73, a total of 40 landraces were resistant, of these 25 were Andean and 15 were Mesoamerican. Likewise, a total of 32 landraces were resistant for race 3481, being 13 Andean and 19 Mesoamerican. Ten landraces were resistant to all races, seven of them are Andean. The pathogenicity index (PI) of races 65, 73 and 3481 were 63%, 52% and 62%, respectively. Race 65 was the most pathogenic, while race 3481 was the least pathogenic (Table 1). These results are in agreement with a study conducted by Vidigal Filho et al. (2007), who detected high pathogenicity of the race 65. The results show that the most resistant common bean landraces were Jalo de Listras Pretas (*Co-13*), Jalo Pintado II, Carnaval Mix SC, Amendoim Cavalo (*Co-AC*), Bolinha Amendoim, Branco Argentino, Bolinha 1 PR, Rosinha Opaco, Guarumbé and Roxo Mineiro, showing resistance reaction to all races evaluated. These landraces would be valuable in future common bean breeding programmes as new sources of resistance to *C. lindemuthianum*.

Table 1. Reaction of Andean and Mesoamerican common bean landraces and pathogenicity index of 65, 73 and 3481 of *Colletotrichum lindemuthianum* races.

Landraces	Gene Pool	Races			Landraces	Gene Pool	Races		
		65	73	3481			65	73	3481
Preto	A[a]	S[b]	S	S	Jalo EEP558 (Embrapa)	A	S	R	S
Preto Andino	A	S	S	R	Feijão Enxofre	A	S	S	S
Manteigão Rajado	A	R	R	S	Rosinha PR	MA	S	R	R
Rajado	A	R	R	S	Preto Brilhoso	MA	S	S	R
Pitanga	A	R	R	S	Rosinha PR II	MA	S	S	S
Beija-flor	A	R	R	S	Carioquinha	MA	S	S	S
Roxinho Paraná	A	R	S	S	Preto I	MA	S	S	R
Jalo Listra Pretas	A	R	R	R	Preto IV	MA	S	S	S
Jalo Pintado II	A	R	R	R	Rosinha Paraná	MA	S	R	R
Bolinha	A	S	S	S	Rosinha Opaco	MA	R	R	R
Rosado	A	R	S	S	Rosinha A	MA	S	S	R
Manteiguinha de Cipó	A	R	R	S	Rosinha B	MA	S	R	R
Jalo sem Cipó	A	S	S	R	Rosinha C	MA	S	R	S
Bodoquena	A	S	R	S	Roxinho A	MA	S	S	S
Chita Bonita	A	R	R	S	Mulatinho Vagem Roxa B	MA	S	S	R
Manteiga sem Cipó	A	S	R	S	Carioca Vagem Rosada	MA	S	S	S
Manteigão	A	R	S	R	Rosinha	MA	S	S	S
Carnaval 1 SC	A	S	R	S	Uberabinha Preto	MA	S	S	S
Carnaval Mix SC	A	R	R	R	Carioca sem Cipó	MA	R	R	S
Preto Brilhoso Achatado	A	R	S	S	Rosinha sem cipó	MA	S	R	R
FC 2016	A	S	S	S	Roxinho Mineiro	MA	S	S	S
FC 2001	A	S	S	S	Preto Guamirim	MA	S	S	S
FC 2045	A	S	S	S	Rosinha Guaicucos	MA	R	S	S
FC 2063	A	S	S	S	Cara Suja	MA	S	S	R
FC 2010	A	S	R	S	Preto SC	MA	R	S	R
Preto Redondo CN 694 FC 1212	A	S	S	S	Preto Precoce Cunha Porã	MA	S	S	S
Preto Chatinho	A	S	S	S	Crioulo Brilhoso	MA	S	R	R
Azulão Ab. Luz	A	R	S	S	Crioulo Brilhoso Ponte Serrada	MA	S	R	R
Amendoim Cavalo	A	R	R	R	FC 117	MA	S	S	S
Vermelho Tozzo	A	S	R	R	Porto Real	MA	S	S	S
Jalo Listras Vermelhas	A	S	R	S	Safira	MA	R	S	S
Rosa	A	S	S	S	Guarumbé	MA	R	R	R
Jalo Precoce	A	R	R	S	Princesa	MA	R	S	R
Bolinha Vermelho	A	S	S	S	Roxinho SC	MA	S	S	S
Bolinha amendoim	A	R	R	R	Carioca Novo	MA	S	S	R
Jalo B	A	S	R	R	Roxo Mineiro	MA	R	R	R
Branco Argentino	A	R	R	R	Preto Argentino	MA	S	S	S
Carnaval 1 PR	A	R	R	S	Roxinho PR	MA	R	S	R
Bolinha 1 PR	A	R	R	R	Bolinha 2 PR	MA	S	R	R
Jalo A	A	S	R	R	Feijão Rosinha	MA	S	S	S
Jalo BR	A	R	R	S	Feijão Moro	MA	S	R	S
Jalo	A	S	R	S	Pathogenicity index (%)		63	52	62

[a] A – Andean; M – Mesoamerican; [b] R – Resistant; S – Susceptible

REFERENCES:

Balardin et al. (1997) Phytopathology 87:1184-1191
Mahuku et al. (2002) Plant Dis 86:1383-1387
Pastor-Corrales et al. (1995). Plant Dis 79:63-67
Vidigal Filho et al. (2007) J. Phytopathology 155, 108–113.

EVALUATION OF COMMON BEAN LINES FOR HEAT TOLERANCE AND WEB BLIGHT RESISTANCE

Rosas[1], J.C., H.D. Martínez Figueroa[2], T.G. Porch[3] H.D., C. Estévez de Jensen[4], A. González[4], J.S. Beaver[4]

[1] Escuela Agrícola Panamericana/Zamorano, Tegucigalpa, Honduras; [2] Instituto de Ciencia y Tecnología Agrícolas, Guatemala; [3] Tropical Agricultural Research Station, USDA-ARS, Mayagüez, PR; [4] Dep. Agro-Env. Sci., University of Puerto Rico, Mayagüez, PR

Web blight (WB) caused by *Thanatephorus cucumeris* Frank (Donk) can produce significant reductions in the yield and quality of beans produced in the humid, lowland tropics. Nighttime temperatures > 20 °C can cause sensitive bean genotypes to have poor pod set and reduced seed yield potential. During 2015 and 2016, a total of 644 lines from different bean breeding programs were evaluated for reaction to WB and adaptation to higher nighttime temperatures (23-25 °C) in replicated field trials planted at the Isabela Substation during the summer months (July and August). During the summer of 2017, the most promising lines from the previous WB trials along with 450 F_7 lines from Zamorano from the 3rd cycle of selection for WB resistance were evaluated in a field trial planted at Isabela, Puerto Rico (PR) using an augmented design with four check lines and two replications. The lines derived from recurrent selection were also evaluated for reaction to WB in an un-replicated trial planted at Zamorano University in Honduras. The trials in PR were inoculated at 28-30 days after planting with a mycelial suspension of an isolate of *Rhizoctonia solani* (Rs) AG-1-IB. The nursery at Zamorano was inoculated with a mixture of three Rs isolates at 15 days after planting; followed by five weekly inoculations.

Several breeding lines were identified that had mean WB scores ≤ 4.0 at both locations using the CIAT 1-9 disease rating scale (Tables 1 and 2). These lines also had a low incidence of infection (≤ 10%) in Honduras and ≤ 10% of damaged seed in Puerto Rico. These lines produced mean seed yields > 1,500 kg/ha in Puerto Rico during growing seasons that had nighttime temperatures ranging from 23 to 25 °C. MHN 322-49 had the highest levels of resistance to WB but did not yield well in Puerto Rico in 2017 due to sensitivity to high temperatures. 'Morales', the susceptible check in Puerto Rico, had approximately 30% damaged seed in trials planted in PR. 'Amadeus 77' had high seed yield potential and a low percentage of damaged seed despite having presented in Puerto Rico moderately susceptible reactions in the foliage. It is advisable to conduct evaluations for both leaf reaction and seed damage to identify lines with resistance to WB. The cross 'ALS 0532-6/PR0650-31' produced the greatest number of promising breeding lines from the third cycle of selection for web blight resistance. PR0650-31 was derived from the cross 'BAT 93/PI417662//VAX 6'. PI417662 is a wild common bean germplasm that was collected in Jalisco, Mexico (Beaver et al. 2012. J. Plant Reg. 6:81-84). It was part of a core germplasm collection of *Phaseolus vulgaris* L. that was screened in Puerto Rico for resistance to WB. Eighteen of the most promising breeding lines were included as entries in a Regional Web Blight Trial (ERMUS 2017) that was distributed to bean research programs in Central America. Web blight resistant cultivars are most needed in warm and humid regions such as the Petén in Guatemala and during the 'apante' plantings in Nicaragua and Costa Rica.

Table 1. Performance of promising lines screened for web blight reaction in trials planted at Isabela, Puerto Rico during the summers of 2015 and 2016.

Line	Disease severity score[1] (1-9)	Disease severity score[1] (1-9)	Seed yield (kg/ha)	Damaged seed (%)
	2016	2015	2016	2016
PR1684- 96	2.6	4.0	1600	4.0
TARS MST 1	4.0	3.3	2017	5.0
PR1217-1	3.2	3.5	1595	5.8
PR1147-1	4.2	3.3	1838	5.0
MHN 322-49	2.8	2.8	1528	9.6
PR0401-259 (ck)	4.2	4.2	1566	3.0
Morales (ck)	6.4	6.6	680	29.4
Amadeus 77 (ck)	4.8	6.6	1817	10.8
LSD (0.05)	0.9		365	8.5

[1] Rated on the CIAT 1-9 scale where 1 = no symptoms and 9 = very severe symptoms.

Table 2. Performance of promising lines screened for web blight reaction in trials during the summer of 2017 at Isabela, Puerto Rico and Zamorano University, Honduras.

	Isabela, Puerto Rico				Zamorano, Honduras	
Line	Disease severity score[1] at 28 DAI[2]	Disease severity score[1] at 34 DAI	Seed yield (kg/ha)	Percent damaged seed	Disease severity score[1]	% of plants with symptoms
MHC 3-3-26	2.5	3.5	2545	6.3	3	10
MHC 3-8-20	3.0	3.5	1716	6.5	3	5
MHC 3-13-29	3.5	3.5	1821	5.5	3	5
MHC 3-22-30	4.0	5.0	2242	2.3	3	10
MHC 3-32-8	3.0	5.0	2321	6.4	3	10
MHC 3-32-9	3.5	5.0	2814	9.3	4	10
MHC 3-32-10	3.0	5.0	2683	7.5	3	10
MHC 3-32-11	3.0	4.0	2683	7.0	3	10
MHC 3-32-16	3.5	4.0	2715	7.4	3	5
MHC 3-32-24	3.5	4.5	2328	9.9	3	10
MHN 322-49	2.8	3.5	760	9.4		
PR0401-259 (ck)	3.6	4.8	1966	4.0		
Morales (ck)	3.8	5.2	559	30.9		
Amadeus 77 (ck)	3.7	5.1	2210	5.7		
Tío Canela 75 (ck)					8	70
LSD (0.05)	0.3	0.4	206	12.5		

[1] Rated on the CIAT 1-9 scale where 1 = no symptoms and 9 = very severe symptoms.
[2] DAI = Days after inoculation

GENERATING A REFERENCE GENOME FOR TEPARY BEAN *(PHASEOLUS ACUTIFOLIUS)*: A HIGHLY HEAT TOLERANT SPECIES

C. Robin Buell[1], Kirstin Bett[2], Phil McClean[3], Samira Mafi Moghaddam[1], Tim Porch[4], Jeremy Schmutz[5], Thomas D. Sharkey[6], Sarathi M. Weraduwage[6]

[1]Department of Plant Biology & Plant Resilience Institute, Michigan State University, E Lansing MI, USA; [2]University of Saskatchewan, Department of Plant Sciences, Saskatoon, Canada; [3]Department of Plant Sciences, North Dakota State University, Fargo ND, USA; [4]USDA-ARS, Tropical Agriculture Research Station, Mayagüez, PR, USA; [5]Hudson Alpha, Huntsville AL, USA; [6]Department of Biochemistry & Molecular Biology, & Plant Resilience Institute, DOE-Plant Research Laboratory, Michigan State University, E Lansing MI, USA

BACKGROUND: Tepary bean (*Phaseolus acutifolius* A. Gray), native to Mexico, the Southwest U.S., and parts of Central America, is highly tolerant to heat and drought. Tepary bean has been domesticated and used as a source for introgression into common bean, thereby conferring biotic and abiotic stress tolerance. Access to genomic resources for tepary bean would be informative not only to common bean breeders interested in accessing tepary traits but also to a wide range of biologists interested in the mechanisms of stress tolerance. Currently, transcriptomic and genetic diversity datasets are available for some tepary bean accessions, while access to a robust genome sequence, annotation, and diversity datasets for tepary bean would enable comparative analyses between common and tepary bean and would allow for identification of loci that confer high ambient temperature tolerance. We have formed a consortium to generate a tepary bean reference genome sequence along with annotation, and an expression atlas to facilitate genome-enabled studies.

PROGRESS TO DATE: The accession G40001, a parent of a biparental recombinant inbred line (RIL) population, was selected to serve as the reference genotype. Flow cytometry of G40001 revealed a haploid genome size of 757 Mb. High molecular weight DNA of G40001 has been isolated and PacBio sequencing is underway at HudsonAlpha. To generate a robust expression atlas, RNA from a set of 12 developmental stages from G40001 has been isolated and mRNA-sequencing (RNA-seq) datasets generated (Table 1). The analysis of the RNA-seq libraries is in progress.

For comparative analyses, a second genotype, W6 15578, a wild tepary bean accession, was sequenced and assembled into scaffolds by NRGene yielding a 662Mb assembly in 635 scaffolds. Pseudomolecules were constructed at USASK using a genetic linkage map (Gujaria-Verma et al. 2016); 35 large scaffolds anchored 445Mb of the genome with most pseudomolecules being covered by two to three scaffolds.

Two datasets are being generated to further explore the tepary bean genome. First, comparative heat stress gene expression data from common bean and tepary bean leaves and flower buds are being generated that will facilitate the understanding of the molecular and genetic differences in heat stress tolerance between these two *Phaseolus* species differential in their heat response. Second, in order to understand the extent of diversity in tepary bean at a genome level, a diversity panel of 54 accessions have been constructed and DNA isolation for use in resequencing and a deeper interrogation of genetic diversity within tepary bean is in progress.

We anticipate having an annotated genome assembly for *P. acutifolius* in early 2018 that will serve as a resource for breeding improved tepary cultivars, identifying introgressions of *P. acutifolius* in common bean, and in an improved understanding of the genetics and physiology of heat tolerance in *Phaseolus* species.

Table 1. List of tissues in the tepary bean gene atlas.

Growth stage	Tissue type	Sample ID	Sample description	Time point
V0	Germinating seed	GS	Embryo and hypocotyl	24 h after imbibition
V2	Root	RT-10	Whole roots	10 days after planting the germinated seeds in sand
V2	Stem	ST-10	Whole stem above cotyledons to primary leaves	10 days after planting the germinated seeds in sand
V2	Primary leaves	LF-10	Whole leaf	10 days after planting the germinated seeds in sand
R5-6	Stem	ST	All stem internodes above the cotyledon	30 days after planting the germinated seeds in Suremix
R5-6	Leaf	LF	Leaf punches	30 days after planting the germinated seeds in Suremix
R5-6	Flower	FW	Whole open flowers	30 days after planting the germinated seeds in Suremix
R7	Young pods seedless	YP	Young pods 1-3 cm	32-36 days after planting
R7-8	Seed at heart stage	HS	Seeds between 3-4 mm across	37-41 days after planting
R7-8	Pod shell at seed heart stage	HP	Pod shells at R7-8 stage	37-41 days after planting
R8	Seed at stage 1	S1S	Seeds between 6-7 mm across	33-48 days after planting
R8	Pod shell at stage 1 seed development	S1P	Pod shells at R8 stage	33-48 days after planting

LITERATURE CITED

Gene-based SNP discovery in tepary bean (*Phaseolus acutifolius*) and common bean (*P. vulgaris*) for diversity analysis and comparative mapping. Gujaria-Verma, N., Ramsay, L., Sharpe, A.G., Sanderson, L.-A., Debouck, D.G., Tar'an, B., and Bett, K.E. et al., BMC Genomics 17 (1): 23.

THE LEGUME FEDERATION RESOURCES FOR COMMON BEAN

Cannon EKS[1]*, Berendzen J[2], Campbell JD[1], Cleary A[2], Dash S[2], Hokin S[2], Huang W[1], Krishnakumar V[4], Weeks NT[3], Wilkey AP[1], Chan A[4], Lyons E[5], Town C[4], Fernández-Baca D[1], Farmer AD[2], Cannon SB[3]

[1] Iowa State University, Ames, IA. [2] National Center for Genome Resources, Santa Fe, NM. [3] USDA-ARS-CICGRU and Iowa State University, Ames, IA. [4] J. Craig Venter Institute, Rockville, MD. [5] University of Arizona, Tucson, AZ.

There are many online resources available for common bean and other warm-season legumes. The Legume Federation is an NSF funded project to produce a federation of online legume resources, to enable data and software sharing and better connectivity between these resources, with the ultimate goal of improving access to the data and tools required by legume research communities. To enable comparative legume research, the Legume Federation (https://legumefederation.org) has developed various online tools, including legume-centric gene families and a resource for browsing and visualizing gene trees: PhyloTree. Another tool, the Genomic Context Viewer, is for browsing and visualizing synteny between common bean and other legumes. This tool shows genomic synteny at both micro (gene level) and macro (chromosome) scales.

The Legume Federation partners and their online resources are shown in Figure 1.

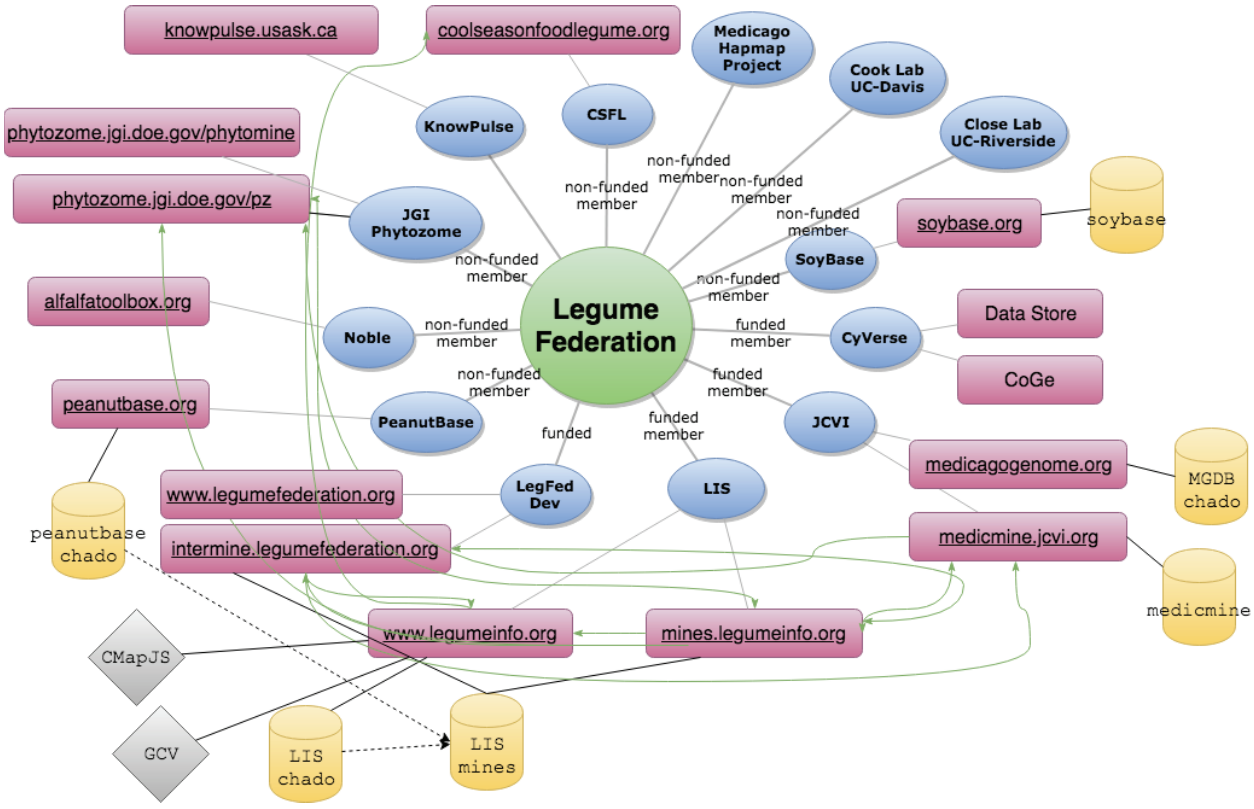


Figure 1. The Legume Federation partners and online resources.

Tools and datasets for common bean provided by the Legume Federation projects include:

- A Geographic Information System (GIS) viewer for visualizing germplasm collections globally against high-resolution maps for beans, cowpea, and other legumes.
- An interactive genetic map viewer, CMap-js, which enables browsing physical and genetic maps individual or in comparison with other maps. See Figure 2.
- An InterMine instance for common bean.
- Genome browsers for both *Phaseolus* genome assemblies, from multiple online resources.
- Sequence search tools for *Phaseolus* genomes and gene models, including sequence match visualizations.
- Gene family viewers for legume species.
- Synteny viewer for exploring genome-wide and gene-focused synteny across the legumes.

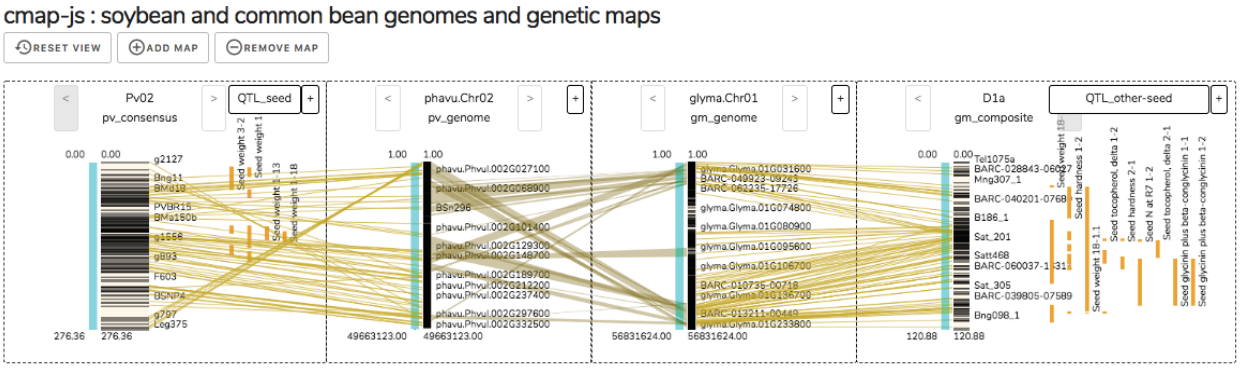


Figure 2. *Phaseolus vulgaris* genetic map for linkage group 2 compared to *Glycine max* genetic map via physical maps for *P. vulgaris* and *G. max*.

In addition to federating online resources, the Legume Federation aims to aid the collection of curated data at its member resources, and to encourage researchers to submit their own data to their community databases. Data collection templates are available for downloading at http://LegumeFederation.org. These templates also serve as guides to authors as to what metadata should be provided with datasets.

The entry point to the Legume Federation and its partners is http://LegumeFederation. Comments as to how the Federation can help meet the common bean research community needs are gratefully welcomed.

REFERENCES

Dash S, Campbell JD, Cannon EKS, Cleary AM, Huang W, Kalberer SR, Karingula V, Rice AG, Singh J, Umale PE, Weeks NT, Wilkey AP, Farmer AD, Cannon SB. Legume information system (LegumeInfo.org): a key component of a set of federated data resources. Nucleic Acids Research, Volume 44, Issue D1, 4 January 2016, Pages D1181–D1188.

CURRENT SITUATION OF THE BEAN GERMPLASM COLLECTIONS IN SPAIN

De Ron AM[1], De la Rosa L[2], and Marcos T[2]

[1] MBG-CSIC, Pontevedra, Spain. [2] CRF-INIA, Alcalá de Henares, Spain

The common bean (*Phaseolus vulgaris* L.) is the most important grain legume for direct human consumption on a global scale. Beans are produced and consumed mainly as dry grain, but also the use of its fresh pods is important in some places. Current bean germplasm collections show a wide variation of phenotypes although, as in many countries local traditional varieties are being replaced by elite cultivars, therefore the genetic erosion is gradually affecting this species. There are several local varieties or "heirloom" varieties, which are characteristic of an area or region, and they can be designated with different names. This germplasm has derived from ancient types by conscious or unconscious selection by farmers and are currently adapted to the agroecological conditions under which they have been grown for centuries.

THE COMMON BEAN IN SPAIN

Production has been dramatically reduced in recent years, due to the abandonment of a crop that may be unprofitable due to unstable yields and the presence of pests and diseases, as well as the massive importation of some varietal types. An example of the reduction of the cultivation area in the Northwest of Spain (Galicia region) in the period 1904-2014 is displayed in Table 1.

Table 1. Common bean cultivation area in Galicia region (1904-2014).

Year	Acreage (ha)	Year	Acreage (ha)
1904	173011	1965	115730
1915	145294	1975	99290
1925	133917	1985	56574
1935	82498	1995	36118
1945	120423	2005	4053
1955	28200	2014	2088

However, a relevant aspect of the local varieties of bean in Spain has been their contribution as a genetic base to the development of different improved varietal types, included those of the current six Protected Geographical Indications (PGIs). This makes Spanish beans more competitive in national and international markets and implies the promotion of the high quality commercial beans, which enhance their market value.

COMMON BEAN COLLECTIONS IN SPAIN

In order to avoid the genetic erosion, conservation activities have been implemented in Spain during the last decades. Since 1993, the Spanish Program for Conservation and Utilization of Plant Genetic Resources for Agriculture and Food, leaded by National Institute for Agricultural and Food Research and Technology (INIA), working as a collaborative network with genebanks distributed along the country, is focused on the conservation, regeneration and characterization of plant genetic resources. It is particularly noteworthy the national collection of common bean placed at the National Plant Genetic Resources Center (CRF-INIA) with 4418 accessions maintained

according the FAO standard procedures (FAO 2014). In the Misión Biológica de Galicia of the National Spanish Research Council (MBG-CSIC, Pontevedra, Spain) since 1987 there is also a collection including 2014 accessions, some of them shared with the national collection at the CRF-INIA. This germplasm collection is the basis for different bean genetic studies and breeding programs (Gil and De Ron 1992, Monteagudo et al. 2006, Yuste-Lisbona et al 2014, González et al. 2016). The MBG-CSIC, and other research institutes, have contributed to the cooperative program in activities associated with the national common bean germplasm collection since its origin. There is a great variability in the types of common bean landraces cultivated by small farmers in Spain, most of them located in the Northwest part of the country. In the collecting missions conducted by the CRF-INIA and the MBG-CSIC, many different varieties have been gathered, like the 167 new accessions collected during the 2011-2014 period in Galicia region (García et al. 2016, De la Rosa et al. 2016).

Due to the small seed quantity donated by farmers, the low germination level of collected accessions or the loss of viability during the conservation time, together with the use of this germplasm, the regeneration and multiplication are usual activities associated to the conservation process (De Ron et al. 2016). So, in the last years (2013-2016), 981 common bean accessions selected from those provided from areas with similar environmental conditions, have been sent by CRF-INIA to different institutions for their multiplication/regeneration. In the case of the MBG-CSIC, 429 accessions (included 108 from the 167 collected in Galicia between 2011 and 2014), were grown in its experimental farm. In the regeneration processes 228 accessions yielded >500 g (approx. 1000 seeds), that should be enough to incorporate to the active collection of the gene bank at the CRF-INIA. The production was less than 500 g in 99 accessions, insufficient for the gene bank requirements and finally 102 were unproductive. There results should be interpreted taking into account the original seed conditions at the moment of gathering and the results of the seed viability tests performed at CRF-INIA. With regard to “on farm” conservation by farmers, the data are scarce, but it is possible to make an approach considering the yield of the 108 accessions sowed directly after collection: 63 yielded more than 500 g, 23 yielded less than 500 g and 22 were unproductive (that is 20 % of the sample). These figures could be an indicator of genetic erosion. As a conclusion, the institutional cooperation is essential when maintaining a wide bean collection coming from very different environmental areas, as the Spanish common bean is.

ACKNOWLEDGEMENTS
The authors thank the projects CSIC 2016SU0004, RFP2013-0001 and RFP2015-00008-C04-03, and Diputación de Pontevedra for farm facilities.

REFERENCES
De la Rosa L, García RM, Marcos T, Fajardo, J. 2016. Mol 16:60-69
De Ron AM, De la Rosa L, Marcos T, et al. 2016. Annu Rept Bean Improv Coop 57: 5-6.
FAO. 2014. Rev. ed. Rome.
García RM, Peluzzo A, Larreina JR, et al. 2016. Actas Hortic 74:15-16
Gil J, De Ron AM. 1992. Plant Breed 109: 313-319
Gonzalez AM, Yuste-Lisbona F, Saburido S, et al. 2016. Frontiers Plant Sci 7:1940
Monteagudo AB, Rodiño AP, Lema M, et al. 2006. HortSci 41: 319-322
Yuste-Lisbona FJ, González AM, Capel C, et al. 2014. Mol Breed 33: 939-952

INVESTIGATING THE POSITIONS OF THE ANTHRACNOSE RESISTANCE GENES LOCATED AT THE BEGINNING OF THE BEAN CHROMOSOME Pv04

Ester Murube, Ana Campa, Juan José Ferreira

Plant Genetic Group, SERIDA (www.serida.org), Asturias, Spain

In this work, we investigate the genetic and physical position of genes controlling resistance to races 6, 38, 39, 357 and 73 in genotype BAT93, and its relationship with the anthracnose resistance genes mapped in the beginning of the chromosome Pv04: *Co-3* in Mex222; *Co-3*^{3} in BAT93; *Co-z* and *Co-y* in JaloEEP558, *Co-3*^{4} in Ouro Negro, *Co-15* in Corinthiano, and *Co-16* in Crioulo 159.

MATERIAL AND METHODS: A total of 145 $F_{2:7}$ recombinant inbreed lines developed from the cross Xana x BAT93 were used in this analysis. This population was genotyped with SNPs markers obtained through Genotyping by Sequencing (Elshire et al., 2011). Additional molecular markers were also analyzed in order to saturate the beginning of the bean chromosome Pv04: *i)* InDel markers (Moghaddam et al., 2013); *ii)* SSR specifically designed from the bean genome; *iii)* markers previously linked to anthracnose resistance genes: Pvctt001 (linked to *Co-3* in Mexico 222; Rodríguez-Suárez et al. 2008), SB12 (*Co-3*^{3} in A493), 254-G15 (*Co-3*^{3} in BAT93; David et al., 2008), g2303 and BARCPVSSR4561 (*Co-3*^{4} in Ouro negro, Gonçalves-Vidigal et al., 2013; Valentini et al., 2017), g2685 (*Co-15* in Corinthiano; Sousa et al., 2015), g2467 (*Co-16* in Crioulo 159; Coimbra-Gonçalves et al., 2016). Goodness-of-fit of observed to expected ratio was tested by chi-square analysis. Linkage analysis was performed with the OneMap package in R v3.4.2 (Margarido et al., 2007) using a LOD value of 3.0 and a maximum RF of 0.30. The physical position of the markers was estimated through alignment of primers or amplicon sequences and the bean genome (V2.1) using the Basic Local Alignment Search Tool (BLAST), both available on www.phytozome.net. Inoculations and disease scoring to five *Colletotrichum lindemuthianum* isolates classified as races 6, 38, 39, 357 and 73 were carried out as described in Ferreira et al., (2013).

RESULTS AND DISCUSSION: Observed segregations for resistance to races 6, 38, 39 and 357 (S x R) fitted to the 1:1 resistant: susceptible ratio, expected for one resistance gene. Co-segregation among the response to the four races was found in 115 recombinant lines; 62 lines susceptible to the four races ($S^{6}S^{38}S^{39}S^{357}$) and 53 lines resistant ($R^{6}R^{38}R^{39}R^{357}$). Lines XB65 and XB66 were resistant to race 39 and susceptible to races 6, 38 and 357 ($S^{6}S^{38}R^{39}S^{357}$) suggesting the involvement of two race-specific genes closely linked, one against races 6, 38 and 357 (named $R^{38\text{-}B}$) and the other against race 39 ($R^{39\text{-}B}$).

A linkage map was developed with 495 non-redundant SNPs distributed in eleven linkage groups (LGs). The total map length was 1546.5 cM with an average distance between markers of 3.12 cM. Linkage analyses between gene $R^{38\text{-}B}$ and SNPs conformed the linkage map revealed a closely linkage with the molecular marker SNP04_027 (RF=0.01; LOD= 30.80). Twelve polymorphic markers were added to the map (Fig 1): 5 InDel, 2 new microsatellite markers, and 5 markers linked to *Co* genes in the Co-3 cluster (BAR004561, Pvctt001, 254-G15F, SB12 and g2303). The gene $R^{38\text{-}B}$ was mapped between the SNP04_027 and 254-G15F markers and, no recombinants were observed with IND04_10570. The closely linkage relationship observed with the four markers BAR004561, Pvctt001, 254-G15F and SB12, tagging the Co-3 cluster, suggests that gene $R^{38\text{-}B}$ is included on it. Concerning physical positions, gene $R^{38\text{-}B}$ is located between 0.55 Mb

(SNP04_027) and 1.62 Mb (254-G15F). Markers BAR004561, SB12, Pvctt001 and g2303 aligned in the physical positions 0.05, 0.28, 0.45 and 3.63 Mb of chromosome Pv04. Interestingly, markers g2685 linked to *Co-15* (5.6 cM) and g2467 to *Co-16* (4.8 cM), that were monomorphic in XB population, aligned at 9.43 and 1.53 Mb of chromosome Pv04.

Segregation for resistance to race 73 (R x R) fitted to 3:1 R:S ratio, expected for two independent genes in a RIL population. Contingency chi-square tests revealed significant associations among the response to race 73, the gene $R^{38\text{-}B}$ ($\chi^2 = 32.91$, $p = 0.00$) and the SNP01_489 ($\chi^2 = 52.89$; $p = 0.00$), a SNP located on chromosome Pv01 in a position corresponding to cluster Co-1 (49.65 Mb). These findings suggest the involvement of the clusters Co-1 and Co-3 in the resistance to race 73. A genetic dissection was performed to locate both resistance genes. In the subpopulation A, including those lines susceptible to race 38 (lacking gene $R^{38\text{-}B}$), the resistance to race 73 showed a monogenic segregation (χ^2=0.01; p=0.89) and it was linked to SNP01_489 in the chromosome Pv01 (RF=0.00; LOD=19.26). In the subpopulation B, which included lines showing the BAT93 genotype for SNP01_489, the resistance to race 73 segregated as a monogenic gene (χ^2=1.58; p=0.21) and, this resistance locus was tightly linked to IND04_10570 in the chromosome Pv04 (RF=0.06; LOD=14.82).

CONCLUSIONS

- Results confirm the presence of an anthracnose resistance cluster (cluster Co-3) at the beginning of the bean chromosome Pv04 including genes conferring resistance to races 6, 38, 39 and 357 in BAT93.

- Results indicate that the resistance to race 73 in the XB population is controlled by two genes, one from Xana located in the position of the Co-1 cluster (LG Pv01) and other from BAT93 in the position of the Co-3 cluster.

- Considering the genetic and physical position of markers tagging the *Co-3* cluster (BAR4561, Pvct0001, 254-G15F, SB12) and *Co-16* (g2467), it is very likely that the *Co-16* gene to be part of it. Concerning to the *Co-15* gene, additional studies should be carried out to verify its location out of the Co-3 cluster.

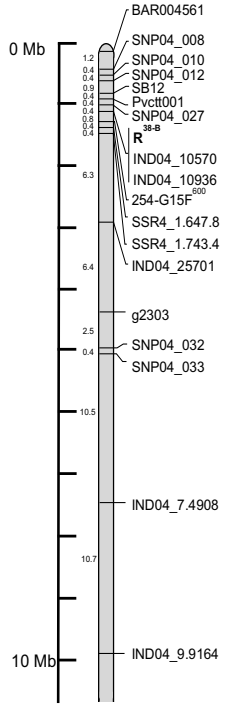


Fig 1. Resulting genetic map for the LG Pv04 with the resistance gene to race 38 ($R^{38\text{-}B}$). Physical and genetic distance, expressed in Mb and cM, is shown on the left.

REFERENCES

Coimbra-Gonçalves et al. (2016) Crop Sci 56:1-12
David et al. (2008) Mol Genet Genomics 280:521-533
Elshire et al. (2011) PloS One 6, e19379
Ferreira et al. (2013) In: Translational Genomics for Crop Breeding
Geffroy et al. (1999) MPMI 12:774-784
Gonçalves-Vidigal et al. (2013) Theor Appl Genet 126:2245
Margarido et al. (2007) Hereditas 144:78-79
Moghaddam et al. (2013) Front in Plant Sci 4:251
Rodríguez-Suárez et al. 2008 Theor. Appl. Genet. 116:807
Sousa et al. (2015) Crop Science 55
Valentini et al. (2017) Theor Appl Genet 130:1705-1722

GENETIC DIVERSITY GATHERED IN A COMMON BEAN PANEL ESTABLISHED FOR GENOME-WIDE ASSOCIATION STUDY

Ana Campa, Ester Murube, Juan Jose Ferreira

Plant Genetic Group, SERIDA (www.serida.org), Asturias, Spain

A diversity Panel of 308 lines was established from the SERIDA bean collection in order to conduct genome-wide association study (GWAS). Results obtained in GWAS strongly depend on factors related to Panel design. The objective of this work was to describe the genetic diversity gathered in this Panel using SNP markers obtained through genotyping by sequencing (GBS).

MATERIAL AND METHODS

The Panel included (i) 213 Spanish local accessions, most of them from the Spanish Core Collection, (ii) 60 elite cultivars for snap bean consumption, traditionally cultivated in Europe, (iii) and 35 well-known genotypes, as anthracnose differential cultivars, parents of recombinant inbreed line populations, or the sequenced bean genotypes BAT93 and G19833. Lines were obtained by self-crossing one plant per accession in greenhouse. Total genomic DNA was isolated from young leaf tissue of each line using the CTAB method. GBS (Elshire et al. 2011) was performed at BGI-Tech using Illumina HiSeq4000 and the *ApeKI* restriction enzyme. SNPs were filtered with the help of software TASSEL V5 (Bradbury et al. 2007).

Population Structure was evaluated using the Structure v.2.3.4 software (Pritchard et al. 2000) with the parameters admixture model and independent allele frequencies. First, the optimum number of subpopulations (*K*) was estimated according to Evanno et al. (2005) using a burn-in period of 1,000 to 5,000 MCMC iterations with 20 replications per subpopulation considered. Then, for the optimum *K* value estimated, accessions were assigned to subpopulations using a burn-in period of 10,000 to 30,000 MCMC iterations and a threshold of 0.9 for *Q* statistics. For study the relationships between accessions a Principal Component Analysis (PCA) was performed using the FactoMineR package of the R project for statistical computing.

RESULTS AND DISCUSSION

Genotyping by sequencing generated 9972 SNP distributed along the 11 bean chromosomes. A total of 3099 SNPs were selected considering physical distance (>500 bp), missing values (<5%) and MAF (>0.01). The number of SNPs per chromosome ranged from 399 on chromosome Pv02 to 179 on chromosome Pv10.

Population structure analysis showed an optimum *K* value of 2 (Fig 1). One subpopulation includes 149 lines closely related to G19833, Andean gene pool, and the other includes 77 lines closely related to BAT93, Mesoamerican gene pool. The remaining 82 lines showed mixtures of both subpopulations, suggesting the existence of a gene flow between Andean and Mesomerican gene pools. Snap bean lines are positioned in both groups, although most of them showed mixtures between the two pools.

The first and second component of the two-dimension plot obtained (Fig 2) represent 40% and 4.9% of the total variation, respectively. Two main groups were identified, represented in the plot by confident ellipses with a threshold value of 0.9. One group includes material closely related to G19833 (Andean) and the other includes materials closely related to BAT93 (Mesoamerican).

Lines that showed a middle position between the two main groups were also observed (e.g. Mexico222, Tu, A493). Results suggest that the Mesoamerican pool is more diverse than the Andean, in which many accessions are grouped together in the plot.

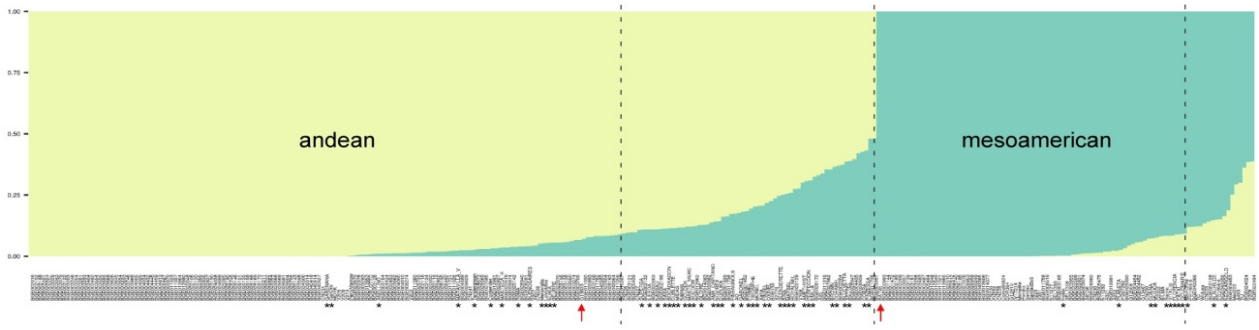


Fig 1. Plot of ancestry estimated for *K*=2. Each bar represents the estimated membership coefficients for each accession in each population (represented by different colors) using a threshold value of 0.9 for *Q* statistic. Arrows indicate the Andean cultivar G19833 and the Mesoamerican BAT93. Asterisks indicate lines derived from commercial snap bean cultivars

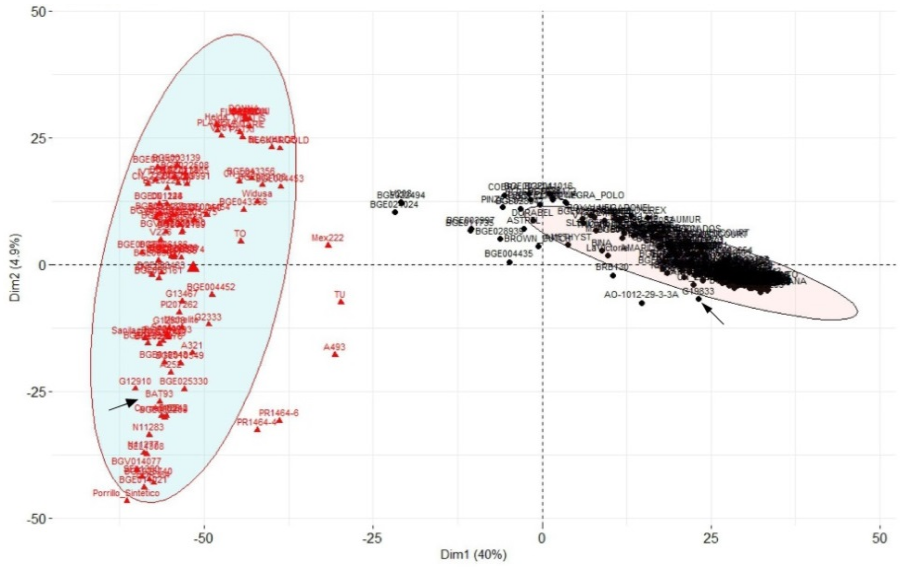


Fig 2. Two-dimension PCA plot obtained for 308 lines and 3099 SNPs. Lines closely related to the Andean or Mesoamerican gene pool are indicated in black or red color, respectively. Arrows indicate the location of cultivars G19833 and BAT93. Confident ellipses are estimated in R with a threshold value of 0.9.

In conclusion, a wide genetic diversity is gathered in this Panel, including accessions of both bean gene pools, Andean and Mesoamerican. Commercial snap cultivars were included in the two gene pools. This Panel can be improved for future GWAS by removing lines with a high similarity levels. The information obtained herein can also help to update the Spanish common bean Core Collection.

REFERENCES

Bradbury et al. 2007. *Bioinformatics* 23:2633-2635
Elshire et al. 2011. PLoS One 6:e19379
Evanno et al. 2005 . Mol Ecol 14:2611-2620
Pritchard et al. 2000. Genetics 155:945-959

EXPLORING THE GENETIC CONTROL OF POD TRAITS IN COMMON BEAN USING GENOME-WIDE ASSOCIATION STUDY

Juan Jose Ferreira[1], Brezo Mateos[1], Alvaro Soler[2], Phil Miklas[2], Ana Campa[1]

[1]Plant Genetic Group, SERIDA (www.serida.org), Asturias, Spain
[2]Grain Legume Genetics and Physiology Research Unit, USDA-ARS-IAREC, Prosser, WA, USA

Depending on pod traits, some bean genotypes can be harvested for consumption as snap beans before the seed development phase. A common bean panel consisting of 308 bean lines was established from the SERIDA collection considering origin (landrace or elite cultivar), form of consumption (dry or snap) and previous genetic knowledge about material. In this work, a genome-wide association study (GWAS) using this panel was conducted to explore the genomic regions involved in genetic control of 15 pod traits.

MATERIAL AND METHODS

The lines were grown in greenhouse (2016-2017) in complete random design and they derived from a single (selfed) plant for each of the 308 accessions.

Phenotyping. Fifteen pod traits, grouped in three classes, were recorded:

i) Pod longitudinal traits: Area_L (cm^2), Width Mid-height_L (cm), Height Mid-width_L (cm), Curved_index, Seed per pod and the qualitative trait Pod length.

ii) Pod cross-section traits: Area_S (cm^2); Width Mid-height_S (cm), Height Mid-width_S (cm), Circular and the qualitative trait Pod section.

iii) Pod color traits. Color was measured in the CIELab color space (L*, a*, b*) and visually (Pod_color).

Potential use as snap bean of each line (Snap) was also recorded considering pod phenotype (fleshiness, presence of hilum, hardness, and color). The four qualitative descriptors (Pod length, Pod section, Pod color, Snap) were recorded in two different crops while the quantitative traits were measured in one crop with the help of the software Tomato Analyzer (Brewer et al. 2006) in 8-10 pods per line.

Genotyping. Total genomic DNA was isolated from young leaf tissue of each line using the CTAB method. Genotyping by sequencing (Elshire et al. 2011) was used to generate 9972 SNP distributed along the 11 bean chromosomes. A total of 3099 SNPs were selected for the analysis after filtering considering physical distance (>500 bp), missing values (<5%) and MAF (>0.01).

Association analysis: The general linear model (GLM) implemented in TASSEL V5 (Bradbury et al. 2007) was used to detect QTL conditioning pod traits. Significance thresholds were determined using Bonferroni correction from the α-level of 0.001. The mixed linear model (MLM), run in TASSEL with a p-value of 0.001 was used to identify common regions with the GLM analysis. The following MLM equation was used: $Y = X\alpha + P\beta + K\mu + e$, where Y is phenotype, X is genotype, P is the PCA matrix and both X and P represent fixed effects, K is the relative kinship matrix value, and e is for residual effects.

RESULTS AND DISCUSSION

Results indicate that many QTL are involved in the genetic control of pod morphological traits. GLM results revealed 36 regions across the eleven chromosomes involved in the genetic control of pod dimension traits, 9 regions located in 7 chromosomes for pod cross section, and 6 regions located in 4 chromosomes for pod color (Fig 1). MLM analysis revealed 22 chromosome regions. Most of these regions are telomeric. Concerning the trait use for consumption, 183 lines were classified as dry and 107 as snap beans. There was overlap detected among the qualitative traits and regions associated to quantitative traits. For example, the trait Snap overlaps with pod longitudinal traits, pod cross sections and pod color (green regions in Fig. 1).

A few studies, using bi-parental populations, observed QTL associated with pod size and color traits on the telomeric regions of the chromosomes Pv01, Pv04 and Pv07 (Yuste-Lisboa et al 2014; González et al 2016). Our results confirm the involvement of those regions, and suggest additional genomic regions affect pod traits on remaining nine chromosomes.

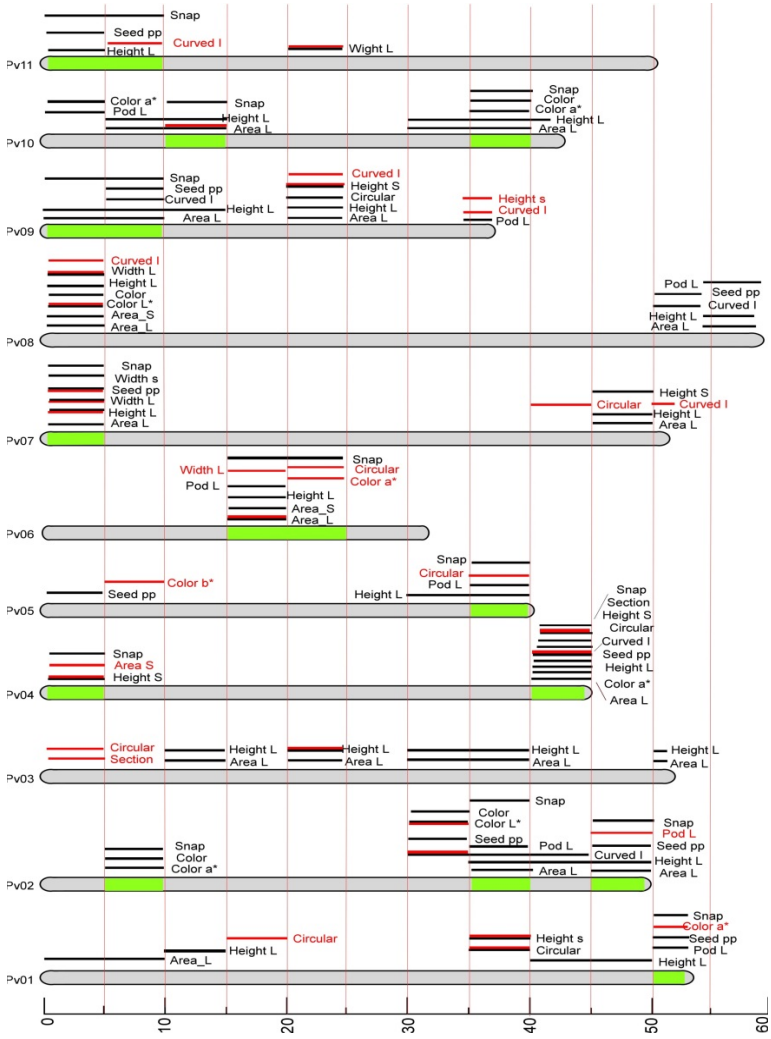


Fig. 1. Chromosome regions (< 5Mb) in which SNP with significant associations were identified using GLM (black bars) or MLM (red bars) analysis. The main regions involved in the use as snap bean are indicated in green, regions with overlapping between the measured traits of pod and the qualitative trait Snap.

REFERENCES

Bradbury et al 2007. *Bioinformatics* 23:2633-2635.

Brewer et al. 2006. Plant Physiol 141 (1), 15-25

Elshire et al. 2011. PLoS ONE 6(5): e19379

González et al. 2016. Front. Plant. Sci. 7:1940

Yuste-Lisboa et al 2014. Mol Breed 33:939-952

CO-SEGREGATION OF RECOMBINANT INBRED LINES OF THE CALIFORNIA DARK RED KIDNEY × YOLANO TO RACES 73 AND 3481 OF *Colletotrichum lindemuthianum*

Calvi AC[1], Gonçalves-Vidigal MC[1*], Valentini G[1], Vidigal Filho PS, Xavier LFS[1], Dartibale GB[1], Souza VB[1], and Gepts P[2]

[1] Departamento de Agronomia, Universidade Estadual de Maringá, Maringá, PR, Brasil;
[2]Department of Plant Sciences, University of California, Davis, CA, USA. *Corresponding
[*]Email: mcgvidigal@uem.br

INTRODUCTION

The common bean (*Phaseolus vulgaris* L.) is the most widely grown grain legume used for direct human consumption (Broughton et al. 2003). Anthracnose, caused by *Colletotrichum lindemuthianum* is the most widespread, recurrent and devastating disease of the common bean in Latin America and Africa (Pastor-Corrales and Tu 1989). Genetic mapping is carried out using segregating populations. Obtaining segregating populations for genetic mapping have been standard practice in the common bean research (Gepts et al. 1993). According to Sanglard et al. (2013), segregating populations have directed to detailed studies and promising to greater efficiency in the genetic breeding of the crop worldwide. Thus, the objective of this research was to evaluate the recombinant inbred lines (RIL's) population from California Dark Red Kidney (CDRK) × Yolano cross using the 73 and 3481 races of *Colletotrichum lindemuthianum.*

MATERIAL AND METHODS

Experiments were conducted under greenhouse conditions and at the Laboratório de Melhoramento do Feijão Comum e de Biologia Molecular do Núcleo de Pesquisa Aplicada à Agricultura (Nupagri) of the Universidade Estadual de Maringá, Paraná, Brazil. The co-segregation tests were carried out in 107 Recombinant Inbred Lines (RIL's), and the parents CDRK and Yolano. Seed samples of the RILs population were kindly provided by Dr. Paul Gepts of the Department of Plant Sciences, University of California, Davis, CA, USA. These seeds were sown in plastic trays containing peat base substrates. In each tray were sown six RIL's to further inoculation with the races 73 and 3481 of *C. lindemuthianum*, both obtained from the mycology collection of Nupagri. The inoculums preparation was performed according to the methodology proposed by Cárdenas et al. (1964). The counting of spores of each race was conducted using the aid of hemacytometer (Chamber of Neubauer-Preciss) and trinocular biological microscope, Motic® brand – mod BA210. After counting, the spore suspension was adjusted to a concentration of 1.2 x 10^6 spores mL^{-1}. The visual evaluation of symptoms in each plant was performed 7 to 10 days after inoculation. Severity scale proposed by Pastor-Corrales et al. (1995) was considered, with score ranging from 1 to 9. Plants with scores from 1 to 3 were considered resistant, whereas those having scores from 4 to 9 were considered susceptible.

RESULTS AND DISCUSSION

A total of 107 RIL's population from CDRK × Yolano were evaluated and it was observed a segregation of 55 resistant and 52 susceptible for races 73 and 3481 ($\chi^2 = 0.084$; $p = 0.7718$), which set the monogenic ratio of 1:1. This results show that the RIL's co-segregated for both inoculated races, indicating that a same gene in CDRK cultivar confer resistance to both races 73 and 3481 of *C. lindemuthianum* (Table 1).

Table 1. Inheritance of anthracnose resistance in the common bean recombinants inbred lines derived from the California Red Dark Kidney × Yolano cross, inoculated with races 73 and 3481 of *C. lindemuthianum*

Races	Reaction		Observed ratio		Expected ratio		χ^2	*P value*
	CDRK	Yolano	R[a]	S[b]	R	S		
73	1	8	55	52	1	1	0.084	0.7718
3481	1	9	55	52	1	1	0.084	0.7718

[a] = Resistant; [b] = Susceptible

The segregation fitted to a 1:1 (R:S) ratio in 107 RIL's for races 73 and 3481, which highlights the potential of these lines in genetic mapping. It was observed that the 107 RIL's showed the same phenotypic behavior. All the lines resistant to race 73 were also resistant to race 3481, and susceptible lines to race 73 were also susceptible to race 3481. This fact highlights the co-segregation of RIL's in relation to resistance to both races (73 and 3481). Genetic analysis revealed that the recombinant inbred lines co-segregated for races 73 and 3481 of *C. lindemuthianum*, showing that the gene present in CDRK confers resistance to both races. Similar results were obtained by Pelembe et al. (2017) for the RILs population from the AND 277 × Rudá cross tested with races 65 and 73 races of *Colletotrichum lindemuthianum*.

CONCLUSION

The reaction of RIL's to races 73 and 3481 of *C. lindemuthianum* presented monogenic inheritance, being conferred by a single dominant gene. The recombinant inbred lines derived from the cross CDRK × Yolano, can be used to map genes that control quantitative and qualitative traits. Furthermore, this population has a great potential for the development of a consensus map of common bean.

ACKNOWLEDGEMENTS: This research was financially supported by the CNPq and Capes. AC Calvi, was supported by a scholarship from Capes. MC Gonçalves-Vidigal and PS Vidigal Filho are grateful for grants from CNPq and Capes.

REFERENCES

Broughton et al. (2003). Plant Soil 252:55–128
Gepts et al. (1993). Ann Rep Bean Improv Coop 36:24-38
Pastor-Corrales and Tu (1989). CIAT 77-104
Pastor-Corrales et al. (1995). Plant Dis 79: 63-67
Pelembe et al. (2017). Ann Rep Bean Improv Coop 60:5-6
Sanglard et al. (2013). Ann Rep Bean Improv Coop 56:23-24.

SEQUENCING THE COMPLEX GENOME OF *UROMYCES APPENDICULATUS*, THE BEAN RUST PATHOGEN

O.P. Hurtado-Gonzales[1], J.R. Diaz-Valderrama[2], M.C. Aime[2], and M.A. Pastor-Corrales[1]

[1]USDA-ARS, Soybean Genomics and Improvement Laboratory, Beltsville, MD, [2]Botany and Plant Pathology Department, Purdue University, West Lafayette, IN

INTRODUCTION: The extensive virulence diversity of *Uromyces appendiculatus* segregates into two distinct groups corresponding to the Mesoamerican and Andean gene pools of common bean (Pastor-Corrales and Aime, 2004). The similarities between the genetic and virulence diversities of common bean and the rust pathogen suggest that these organisms have undergone a parallel co-evolution. The objective of the present study was to use genomics to find new opportunities to improve our understanding of the interaction between common bean and the bean rust pathogen and of the complex diversity of this pathogen. To that end, we have initiated whole genomic sequencing of *U. appendiculatus* using Andean and Mesoamerican races.

MATERIALS AND METHODS: Urediniospores of Andean race 5-0 and Mesoamerican race 31-1, also known as races 39 and 53, were increased under greenhouse conditions. High-quality and high-molecular weight DNA was obtained from urediniospores of these races. Sequencing was done at the Purdue University Genomics Core Facility. Sequencing libraries of three different sizes (500bp, 2Kb, and 4Kb) were prepared. Small libraries (500bp) from each race were paired-end sequenced (2x250) using the HiSeq2500 Illumina sequencer, one lane per race. Large libraries (2Kb and 4Kb mate pairs) from each race were paired-end sequenced (2x100) using the HiSeq2500 Illumina sequencer. Sequencing quality control was attained using the FastQC program. Indexed reads of the 500bp paired-end libraries were subjected to a kmer analysis using the Kmergenie package (v1.6213). De novo assemblies were performed using the program Discovar de novo (Broad Institute, Cambridge, MA). Software BESST (Sahlin et al., 2014) was used for scaffolding of the de novo assemblies using the paired-end reads from the mate-pair libraries. A final assembly was produced with contigs larger than 500bp. Completeness of the genome assemblies was achieved using CEGMA (Parra et al., 2007). Scaffolds from the assembly of each race larger than 10Kb were examined for the presence of SSRs, using the QDD pipeline v3.1 for marker development (Meglécz et al., 2007). Contigs with the SSRs from race 31-1 were blast-compared with contigs with SSRs from race 5-0. Polymorphic SSRs were detected through this comparison using the Geneious software v10.2.3. A total of 83 SSR primers were tested using DNA from urediniospores of races 5-0 and 31-1. PCR products were resolved on 3% agarose gels. SSR markers with clear amplification patterns were selected and used on a collection of 46 diverse races from different countries. PCR products were amplified and resolved on an ABI3070XL capillary machine. Allele scores were recorded and genetic distances calculated using GenAlex 6.503 (Peakall and Smouse 2012). The Unweighted Pair Group Method with Arithmetic Mean (UPGMA) dendrogram was built using the MEGA software v7.0.25 (Kumar et al., 2015).

RESULTS AND DISCUSSION: A total of 73.4 Gb and 69.6 Gb of data were generated for races 5-0 and 31-1, respectively. In addition, 320M and 307M reads for races 5-0 and 31-1 were processed. De novo assemblies resulted in genomes of 587.6 Mb and 546.7 Mb at a k-mer of 90 for races 5-0 and 31-1, respectively. The N50s values were ~78Kb and 50.5Kb and the number of contigs larger than 500bp were 95,581 and 106,558 for races 5-0 and 31-1, respectively. The genome assemblies of the two races resulted in a high number of contigs, suggesting a complex,

and highly repetitive genome, that is larger (>500Mb) than most of the reported genomes of fungal plant pathogens. CEGMA analysis against a conserved set of 248 protein families that occur in eukaryotes determined the assemblies to be 95% and 97% completed for races 5-0 and 31-1 respectively.

A total of 110,000 SSRs were identified and out of 83 SSRs tested, 77 resulted in clear amplification pattern. A subset of 16 SSRs were tested on 46 races of *U. appendiculatus* maintained at Beltsville. These 16 SSRs markers identified 76 alleles, which was equivalent to using 158 SNP markers. Analysis using 16 SSRs markers, separated the 46 races into two different groups of races, one Andean and another Mesoamerican (Figure 1). This separation suggests that the bean rust pathogen has co-evolved with its common bean host. The results from these studies should provide insights into the evolution of the races of *U. appendiculatus* and could be used for surveillance of new races and may provide information leading to the identification of virulence genes in *U. appendiculatus.*

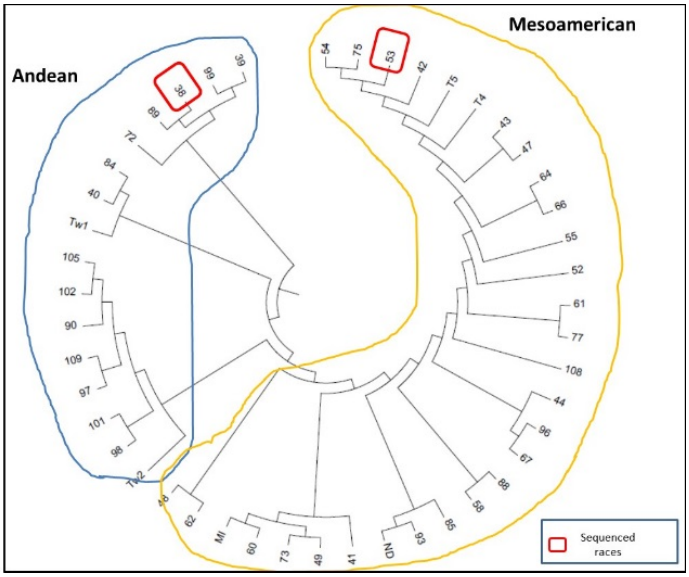


Figure 1. The Unweighted Pair Group Method with Arithmetic Mean (UPGMA) dendrogram done with 16 SSRs separated 46 races of *Uromyces appendiculatus* into two distinct groups, one Andean (left) and one Mesoamerican (right). The sequenced Andean race 5-0 (also known as race 38) and Mesoamerican race 31-1 (also known as race 53) are inside red rectangles

REFERENCES

Koichiro, T. et al. (2013). MEGA6: Molecular Evolutionary Genetics Analysis version 6.0. Molecular Biology and Evolution: 30 2725-2729.

Meglécz, E. et al. (2010). QDD: a user-friendly program to select microsatellite markers and design primers from large sequencing projects. Bioinformatics 26(3):403-404

Parra, G. et al. (2007) CEGMA: a pipeline to accurately annotate core genes in eukaryotic genomes." Bioinformatics, 23: 1061-1067.

Pastor-Corrales, M. A., Aime, M.C. 2004. Differential cultivars and molecular markers segregate isolates of *Uromyces appendiculatus* into two distinct groups that correspond to the gene pools of their common bean hosts. Phytopathology 94 (6): 82.

Peakall, R. and Smouse P.E. (2012). GenAlex 6.5: genetic analysis in Excel. Population genetic software for teaching and research-an update. Bioinformatics 28:2537-2539.

Sahlin, K. et al. (2014). "BESST-efficient scaffolding of large fragmented assemblies." BMC Bioinformatics 15, 281.

PROMISING BLACK-SEEDED COMMON BEAN BREEDING LINES RESISTANT TO BCMV, BCMNV AND BGYMV WITH ADAPTATION TO TERMINAL DROUGHT AND ACID SOILS OF TROPICAL SOUTHEASTERN MEXICO

Ibarra Pérez FJ[1]*, Tosquy Valle OH[1], López Salinas E[1], Rodríguez Rodríguez R[2], Villar Sánchez B[3], Anaya López JL[4], Garrido Ramírez EG[3] and Zetina Lezama R[1]

[1]INIFAP, Campo Experimental Cotaxtla, Veracruz, Mexico (ibarra.francisco@inifap.gob.mx), [2]INIFAP, Campo Experimental Ixtacuaco, Veracruz, Mexico, [3]INIFAP, Campo Experimental Centro de Chiapas, Mexico, [4]INIFAP, Campo Experimental Bajío, Guanajuato, Mexico

INTRODUCTION. Common bean (Phaseolus vulgaris L.) is an important crop in different production regions of Mexico. In Veracruz and Chiapas, located in tropical lowlands of the southeastern region, seed yields are low (<700 kg ha^{-1}) as the crop is affected by biotic and abiotic factors, among them: angular leaf spot [caused by *Pseudocercospora griseola* (Sacc.) Crous & U. Braun], and virus such as and *Bean common mosaic virus* (BCMV), *Bean common mosaic necrotic virus* (BCNMV) and *Bean golden yellow mosaic virus* (BGYMV). Terminal drought and acid soils of low fertility as well, are the abiotic factors that most limit bean production in the residual moisture cropping system. The objective of this work was to assess a group of promising recombinant breeding lines, derived from different parental crosses, to determine their adaptation to bean production environments of tropical lowlands of southeastern Mexico.

MATERIALS AND METHODS. A uniform regional yield trial, composed of 12 breeding lines previously selected for the presence of *I*, *bc*-3 and *bgm*1 resistance genes to BCMV, BCMNV and BGYMV using molecular markers (Anaya *et al*., 2016; Garrido *et al*., 2016), and for their adaptation to terminal drought and acid soils. The yield trial was carried out across ten different environmental conditions of tropical southeastern Mexico during yrs. 2016 and 2017 (Table 1).

Table 1. Location, season, cropping system of ten environmental conditions of tropical lowlands in Veracruz and Chiapas, Mexico, to evaluate 12 black-seeded bean lines during yrs. 2016-2017.

Environments	Season[ǂ]	Crop cycle	Cropping system[†]
Villa F., Chiapas[§]	S	June-Sept	R
Villa F., Chiapas[£]	S	June-Sept	R
Ocozocoautla, Chiapas	F-W	October–January	RM
Rio Grande, Veracruz	F-W	October–January	RM
Medellin, Veracruz	F-W	October–January	RM
Rodriguez C., Veracruz[§]	F-W	October–January	RM
Rodriguez C. Veracruz[£]	F-W	October–January	RM
Tlalixcoyan, Veracruz	F-W	October–January	RM
Medellin, Veracruz	W-S	February–May	IR
Medellin, Veracruz	W-S	February–May	TD

[ǂ] S= summer, F-W= fall-winter, W-S= winter-spring. [†]R= rainfed, RM= residual moisture, I = irrigated, TD= terminal drought. [§],[£] acid soil locations, with and without agricultural dolomite added, respectively.

A randomized complete block design (RCBD) with three replicates was used with three-furrows of five meters long plots. Grain yield (14% seed moisture) was determined as kg ha^{-1}. Combined analyses of variance were carried out for seed yield across ten environments. To estimate stability parameters of each breeding line, Eberhart and Russel (1966) method was followed. The effect of terminal drought was estimated using the drought susceptibility index (DSI) (Fisher and Maurer, 1978) and the relative efficiency index (REI) (Graham, 1984). The last index (REI) was also used to estimate the acid soil effect.

RESULTS AND DISCUSSION. Based on the combined analysis of variance, six bean lines and the two check cultivars obtained seed yields from 1303 to 1504 kg ha^{-1}, significantly higher than the rest of the breeding lines. Within this group, four lines showed stability across environments (bi=1 and $S^2di=0$) but Papaloapan/SEN 46-7-11 and Jamapa Plus/XRAV-187-3-1-2 responded better in good environments and were unpredictable (Table 2). Under terminal drought conditions, a group of five bean lines and Negro Comapa had a drought susceptibility index <1.0, indicating that were the most tolerant to such abiotic factors, of which only Jamapa Plus/XRAV-187-3-1-2 showed the highest relative efficiency under irrigation and terminal drought (REI=2.28). Under acid soil conditions, N Citlali/XRAV-187-3-1-8 and Jamapa Plus/XRAV-187-3-4-4 were the most productive with 911 and 706 kg ha^{-1} and 767 and 576 kg ha^{-1}, with dolomite and without dolomite, respectively. Same breeding lines had the highest relative efficiency, with a REI=2.0 and REI=1.38, suggesting a better adaptation to acid soils (data not shown). In contrast, check cultivars Negro Comapa and Negro Grijalva had low relative efficiency indexes, REI=1.05 and 0.91, respectively which indicates poor adaptation to acid soils (Table 2). It is important to mention that among the bean lines identified as promising in this study, Jamapa Plus/XRAV-187-3-1-8 possesses three genes (*I*, *bc*-3 and *bgm*1) that confer resistance to BCMV, BCMNV and BGYMV, Jamapa Plus/XRAV-187-3-4-4 carries both, *I* and *bc*-3, genes and Jamapa Plus/XRAV-187-3-1-2 and Papaloapan/SEN 46-3-7 only the *I* gene.

Table 2. Stability parameters across ten environmental conditions of 12 tropical black-seeded bean lines evaluated in Veracruz and Chiapas, Mexico during yrs. 2016 and 2017.

Bean lines	Stability parameters				Stress Index		
	bi	S^2di	Mean	GC[&]	DSI	REI[§]	REI[†]
1 Papaloapan/SEN 46-3-7	0.94*	66140	1334*	BU	0.51	0.95	0.90
2 Papaloapan/SEN 46-6-6	0.79	24617	1162	S	1.06	0.55	1.09
3 Papaloapan/SEN 46-7-7	1.02	22703	1203	S	1.04	0.68	0.80
4 Papaloapan/SEN 46-7-11	1.21*	54484	1246	BU	1.48	0.47	0.72
5 N Citlali/XRAV-187-3-1-6	1.04	-12221	1303*	S	0.50	0.92	0.90
6 N Citlali/XRAV-187-3-1-8	0.98	24013	1377*	S	0.91	0.70	2.00
7 N Citlali/XRAV-187-3-14-6	0.97	3920.1	1172	S	1.29	0.39	0.86
8 N Citlali/XRAV-187-3-14-7	1.11	21032	1222	S	1.03	0.71	1.03
9 N Citlali/XRAV-187-3-16-7	0.72	-12281	1145	S	0.87	0.79	0.66
10 Jamapa Plus/XRAV-187-3-1-8	1.20	10416	1438*	S	1.14	1.55	0.90
11 Jamapa Plus/XRAV-187-3-1-2	0.97*	80799	1504*	BU	0.94	2.28	1.06
12 Jamapa Plus/XRAV-187-3-4-4	0.97	19581	1359*	S	1.27	1.07	1.38
Negro Comapa (Check)	1.03	34197	1446*	S	0.86	1.78	1.05
Negro Grijalva (Check)	1.05	30034	1471*	S	1.10	2.17	0.91
Mean			1313		1.00	1.07	1.02

[&] GC= Group class according to bi and S^2di parameters, S= stable cultivar (bi=1 and $S^2di=0$), BU= cultivar responds in better environments but is unpredictable. DSI = drought susceptibility index, [§] REI relative efficiency index under drought, [†] REI under acid soils.

CONCLUSIONS. Jamapa Plus/XRAV-187-3-1-8 and Jamapa Plus/XRAV-187-3-4-4, promising bean breeding lines had high and stable seed yield performance across ten environments; the first one showed high production efficiency to terminal drought while the second one to acid soils. Jamapa Plus/XRAV-187-3-1-2 had high seed yield but unpredictable performance, showed good adaptation to drought (DSI<1.0) and had the highest relative efficiency under drought conditions (REI=2.28).

REFERENCES

Anaya LJL, Ibarra PFJ, López SE, Tosquy VOH and Acosta GJA. 2016. BIC 59:97-98.
Eberhart SA and WA Russell. 1966. Crop Sci. 6:36–46.
Fischer, RA and R. Maurer. 1978. Aust. J. Agric. Res. 29:897–912.
Garrido REG, Ibarra PFJ, López SE and Tosquy VOH. 2017. BIC 60:47-48.
Graham, RD. 1984. Adv. Plant Nutr. 1:57–102.

IDENTIFICATION AND CHARACTERIZATION OF *SLOW DARKENING* GENE IN PINTO BEAN (*PHASEOLUS VULGARIS* L.)

Islam NS[2], Marsolais F[1, 2] and Dhaubhadel S[1, 2]

[1]London Research and Development Centre, Agriculture and Agri-Food Canada, London, ON,
[2]Department of Biology, University of Western Ontario, London, ON
E-mail: nislam33@uwo.ca, sangeeta.dhaubhadel@canada.ca

Postharvest darkening of seed coat in pinto bean (*Phaseolus vulgaris* L.) is an undesirable trait that affects its market value. A single gene *SLOW DARKENING* (*Sd*), is responsible for the delaying of proanthocyanidin accumulation in the seed coat of slow darkening (SD) cultivars of pinto beans. Two simple sequence repeat (SSR) markers, Pvsd-1157 and Pvsd-1158, co-segregate with the SD trait mapped on chromosome 7. A basic helix-loop-helix (bHLH) gene, *PvbHLH333,* was identified as a candidate *Sd* by comparing the physical and linkage distances between the markers.

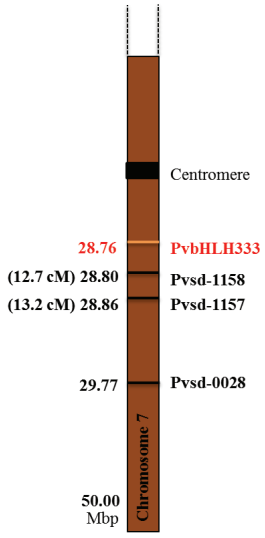


Figure 1: Selection of candidate gene by comparing linkage distance and physical distance of SSR markers. Schematic diagram showing tentative physical location of the candidate gene, *PvbHLH333* and the markers- Pvsd-1157 and Pvsd-1158. Linkage distance from the tightly linked marker Pvsd-1158 to SD trait is 0.4 cM while physical distance is ~37 Kbp. Linkage and physical distance between Pvsd-1157 and Pvsd-1158 is 0.5 cM and ~50 Kbp respectively, indicating the relation of linkage and physical distance is 0.1 cM ≈ 10 Kbp.

Figure 2: Domains in PvbHLH333. From N to C-terminus, MYB Interacting Region (MIR), Poly Glutamate (Poly E), basic helix-loop-helix (bHLH) and ACT-like domain. Molecular weight of the protein is approximately 74 kDa.

PvbHLH333 is annotated as Transparent Testa 8 (TT8). Sequence comparison of PvbHLH333 with characterized TT8 orthologues from other plant species showed high sequence similarity.

To determine localization of PvbHLH333 in the subcellular compartments, PvbHLH333 gene sequences were isolated from two pinto bean cultivars, CDC-Pintium (regular darkening) and 1533-15 (slow darkening), that differ in postharvest seed coat darkening, and a translation fusion with YFP was created. Transient expression of PvbHLH333 in Nicotiana benthamiana leaves

showed that PvbHLH333 from both the pinto bean cultivars localize in the nucleus as expected for a transcription factor.

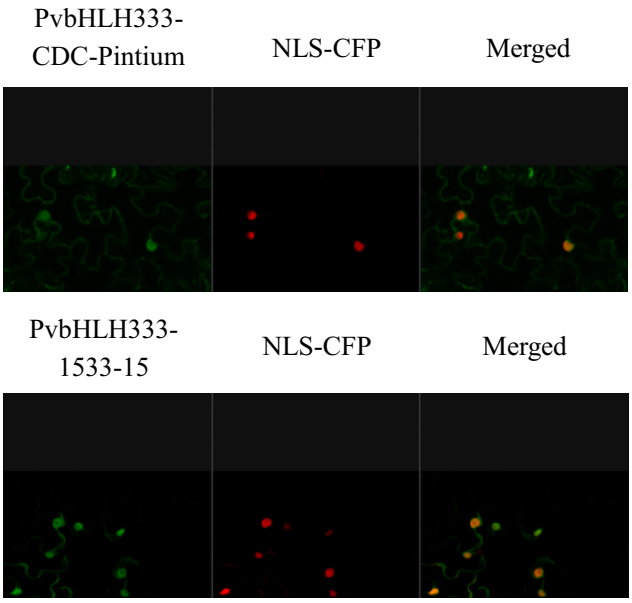


Figure 3: Subcellular localization of the PvbHLH333 from CDC-Pintium and 1533-15. *PvbHLH333*-YFP co-infiltrated with a nuclear localization signal (NLS)-CFP in *N. benthamiana* leaves using *Agrobacterium* mediated transformation. The fluorescence was visualized by confocal microscopy.

To determine if *PvbHLH333* is an orthologue of *Arabidopsis TT8* (*AtTT8*) that regulates the proanthocyanidin biosynthesis in seed coat, a complementation assay was performed. *Attt8* mutants defective in proanthocyanidin accumulation in the seed coat were obtained and the ability of *PvbHLH333* from CDC-Pintium and 1533-15 to restore WT seed coat colour was evaluated.

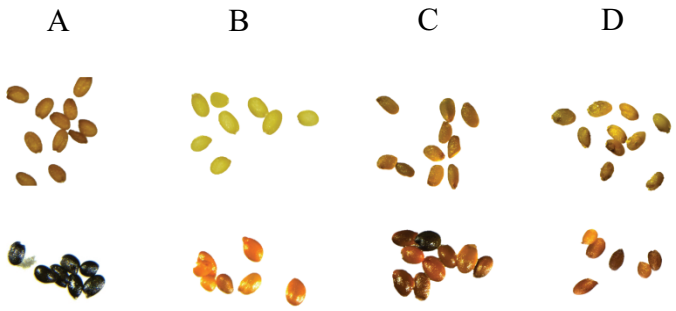


Figure 4: Complementation of Arabidopsis *tt8* seeds with PvbHLH333-CDC-Pintium and PvbHLH333-1533-15 restores partial colour in T1 generation. Seed coat pigmentation (top) and DMACA stained seeds (bottom) of A) wild type, B) *tt8*, C) T1 seeds of *tt8* transformed with PvbHLH333-CDC-Pintium and D) T1 seeds of *tt8* transformed with PvbHLH333-1533-15.

The results revealed that PvbHLH333-CDC-Pintium and PvbHLH333-1533-15 are able to partially rescue WT phenotype, suggesting *PvbHLH333* has a role in proanthocyanidin biosynthesis.

A SATURATED GENETIC LINKAGE MAP OF COMMON BEAN (*PHASEOLUS VULGARIS* L.) DEVELOPED USING GENOTYPING BY SEQUENCING (GBS)

Paulo Izquierdo[1], Scott Shaw[2], Matt Berry[1], Karen Cichy[1,2]

[1] Department of Plant, Soil, and Microbial Sciences, Michigan State University, East Lansing, MI, USA. [2] Sugarbeet and Bean Research Unit, USDA-ARS, East Lansing, MI, USA.

High-density linkage maps are valuable tools to uncover the genetic basis of complex quantitative traits. Our goal was to construct a high-density genetic map to facilitate the identification of markers associated with cooking time in common bean. We used genotypic data of the F6 recombinant inbred line population developed from two Andean lines from Tanzania ADP0027 x ADP0037 to construct a genetic map. The map was constructed from single nucleotide polymorphism (SNP) markers that were genotyped using a Genotyping by Sequencing **(**GBS) protocol reported by Schröder *et al* 2016 with the Taqa1/MseI enzyme combination for library construction. A total of 48,244 markers were identified in 146 RILs and their parents. The SNPs were filtered by alignment quality, minor allele frequency, and percentage of missing data. In total 2,427 markers were assigned to 11 linkage groups Figure 1. The map has a length of 1,137 cM and a mean distance between markers of 0.58 cM. Overall, the map has 175 SNPs in coding (114 Synonymous, and 61 Missense), and 2,252 in non-coding regions Figure 2.

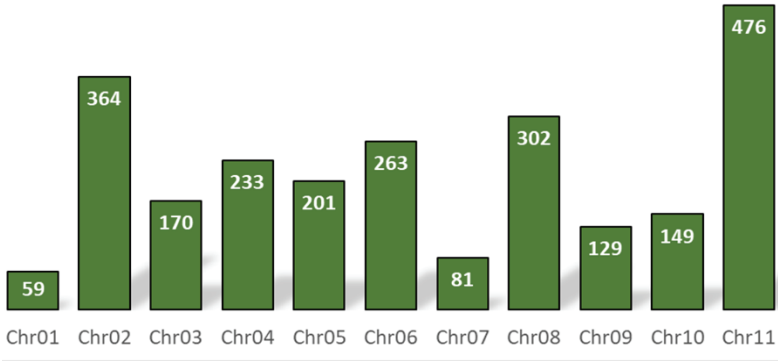


Figure 1. Number of SNPs by chromosome

GBS enabled us to develop a saturated linkage map for common bean with good genome coverage that will facilitate investigation of QTL analysis in ADP0027 x ADP0037 population. However, it is important to highlight that we used a variant of the original GBS protocol that has been reported on in 2011 by Elshire et al. We used the enzyme combination TaqaI/MseI to create the library. We used that combination to increase the genome coverage, but because these enzymes are not sensitive to methylation, we got a high number of SNPs in repetitive sequences across the chromosomes, which is a limitation to do a reliable SNP identification. Although we have enough information to create a saturated map in ADP0027 x ADP0037 population, we suggest that for diversity panels and GWAS purposed it would be better to use methylation-sensitive enzymes to increase the number of SNPs in coding regions and reduce it in repetitive regions.

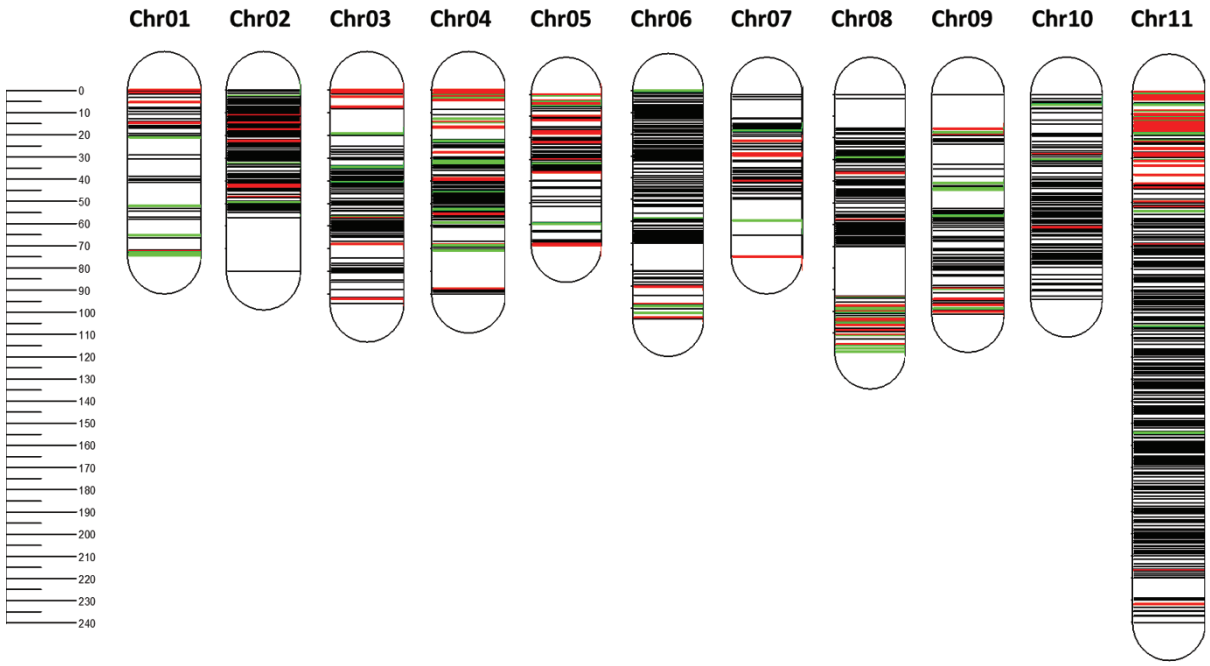


Figure 2. Linkage map based on RILs of the Andean population Incomparable (ADP0027) x W6 16488 (ADP0037). Map has a length of 1,137 cM, and it comprises 2,427 SNPs with a mean SNP distance of 0.58 cM. SNPs in coding regions and introns are colored in red (175) and green, respectively (136).

REFERENCES

Elshire RJ, et al (2011) A robust, simple genotyping-by-sequencing (GBS) approach for high diversity species. PLoS One 6: e19379

Schmutz, *et al* (2014). A reference genome for common bean and genome-wide analysis of dual domestications. *Nature Genetics*.

Schröder, e*t al* (2016). Optimization of genotyping by sequencing (GBS) data in common bean (*Phaseolus vulgaris* L.). *Molecular Breeding*.

PREDICTION OF GENETIC POTENTIAL OF COMMON BEAN SEGREGANT POPULATIONS FOR SLOW SEED-COAT DARKENING

Silva FC[1], Sousa LL[1], Melo PGS[1], Melo LC[2], Pereira HS[2]

[1]Universidade Federal de Goiás, [2]Embrapa Arroz e Feijão, GO- Brazil
eng.fernanda09@gmail.com

INTRODUCTION

In Brazil, Carioca common beans accounts for 70% of the consumer market (Pereira et al., 2017). For this type of grain, the darkening of the grains during storage has deserved attention because it is responsible for the low acceptance of the grains by the consumer. Some studies have demonstrated the possibility of selecting Carioca common bean with slow seed-coat darkening (Silva et al., 2008; Araújo et al., 2012). Thus, the objective of this study was to predict the genetic potential of common bean segregant populations for slow seed-coat darkening.

MATERIALS AND METHODS

Biparental crosses were carried out in a partial diallel scheme between 20 elite common bean lines with carioca seeds, divided into two groups: group I – two genotypes for slow seed-coat darkening (BRS Requinte and BRSMG Madrepérola); group II - ten genotypes for regular seed-coat darkening (BRS Estilo, Pérola, BRS Cometa, BRS Pontal, BRSMG Majestoso, IAC Alvorada, IPR Saracura, IPR Siriri, BRS Sublime and BRS Notável).

Populations and parents were evaluated in the $F_{3:4}$ generation, 2010/winter season, in Santo Antônio de Goiás, state of Goiás and in the $F_{4:5}$ generation, in the 2010/rainy season in Ponta Grossa, state of Paraná in an experimental randomized blocks design, with three replications, in which the plot was 7.2 m^2 .

Seeds of 40 plants were collected individually in each replicate, totaling 120 plants per treatment. The seeds were stored for 155 days in room temperature and humidity. Seeds-coat darkening evaluation were carried out in each plant, which were ranked from 1 (very light colored grains) to 5 (very dark colored grains). Estimates of genetic and phenotypic parameters were obtained for each population in each environment, using the methodology of Melo et al. (1997). Data were submitted to analysis of variance and, later, the methodology of Jinks and Pooni (1976) was used for predicting the genetic potential of the populations. This methodology allows estimating the probability of extraction of lines that exceed a standard. In this case, the average of the population with the slow seed-coat darkening in each environment was used as a standard, subtracting 10%.

RESULTS AND DISCUSSION

Significant differences ($p < 0.01$) were found between treatments in both generations, indicating the presence of genetic variability. The heritability estimates showed great variation, with values of high magnitude (0.00-0.95). The BRSMG Madrepérola populations presented consistent heritability values in both locations and generations (Table 1), demonstrating that genotype selection based on its respective phenotypes may be successful. The probabilities of populations exceeding the standard for the slow seed-coat darkening ranged in the $F_{3:4}$ generation from 0 to 37.1% and $F_{4:5}$ from 0 to 41.7%.

The highest probabilities for the achievement of promising lines were verified in the populations from the crossing with the cultivar BRSMG Madrepérola. High probability values

associated with high genetic variances and low means were found in populations 12, 13, 14, 18, 19 and 20. Based on these results, these populations present a possibility of success in the extraction of lines with slow seed-coat darkening.

Table 1. Means for seed-coat darkening, genetic variance (σ^2_G), heritability (h^2), Z value and P (Probability to obtain lines that exceed the best population by 10%), Santo Antônio de Goiás-GO and Ponta Grossa-PR in 2010/155 days storage.

Populations	$F_{3:4}$ Santo Antônio de Goiás					$F_{4:5}$ Ponta Grossa				
	Mean	σ^2_G	h^2	Z	P (%)	Mean	σ^2_G	h^2	Z	P (%)
1 (1 x 3)*	4.11	0.40	0.52	-1.72	4.27	4.82	0.00	0.00	-5.26	0.00
2 (1 x 4)	3.74	0.31	0.50	-1.46	7.21	4.67	0.26	0.63	-2.57	0.51
3 (1 x 5)	4.54	0.01	0.03	-2.92	0.18	4.37	0.33	0.71	-1.98	2.39
4 (1 x 6)	3.99	0.31	0.47	-1.71	4.27	4.46	0.35	0.72	-2.04	2.07
5 (1 x 7)	4.25	0.11	0.22	-2.36	0.91	4.85	0.02	0.09	-4.37	0.00
6 (1 x 8)	4.39	0.00	0.00	-3.26	0.06	4.73	0.05	0.24	-3.81	0.01
7 (1 x 9)	4.31	0.03	0.07	-2.65	0.40	4.77	0.09	0.36	-3.54	0.02
8 (1 x 10)	3.99	0.20	0.37	-1.89	2.94	3.77	0.01	0.04	-4.46	0.00
9 (1 x 11)	3.99	0.27	0.44	-1.77	3.84	4.27	0.30	0.75	-1.20	11.51
10 (1 x 12)	3.95	0.39	0.53	-1.58	5.71	3.83	0.28	0.69	-1.95	2.56
11 (2 x 3)	3.37	1.13	0.82	-0.66	25.46	3.62	1.11	0.92	-0.74	22.96
12 (2 x 4)	2.96	0.66	0.77	-0.42	33.72	3.46	1.10	0.92	-0.55	29.12
13 (2 x 5)	3.62	1.07	0.79	-0.87	19.22	3.35	1.68	0.95	-0.34	36.69
14 (2 x 6)	3.00	0.56	0.74	-0.49	31.21	3.62	0.85	0.92	-0.35	36.32
15 (2 x 7)	3.72	1.01	0.77	-0.97	16.60	3.46	1.41	0.94	-0.50	30.85
16 (2 x 8)	3.74	1.08	0.78	-0.96	16.85	3.35	1.32	0.93	-0.63	26.43
17 (2 x 9)	3.75	1.02	0.77	-0.99	16.11	3.62	1.37	0.93	-0.69	24.51
18 (2 x 10)	2.84	0.54	0.75	-0.33	37.07	3.77	0.71	0.90	-0.46	32.28
19 (2 x 11)	3.19	0.85	0.79	-0.59	27.76	3.85	1.04	0.93	-0.26	39.74
20 (2 x 12)	3.23	1.00	0.81	-0.58	28.10	3.42	1.07	0.94	-0.21	41.68

*Parents: 1- BRS Requinte; 2- BRSMG Madrepérola; 3- Pérola; 4- BRS Estilo; 5- BRS Cometa; 6- BRS Sublime; 7- BRS Notável; 8- BRS Pontal; 9- BRS Majestoso; 10- IAC Alvorada; 11-IPR Saracura;12-IPR Siriri.

REFERENCES

Araújo LCA et al. (2012). Ciênc agrotec, 36: 156-162.
Jinks JL and Pooni HS (1976). Heredity, 36:243-266.
Melo LC et al. (1997). Brazilian Journal of Genetics, 20: 283-292.
Pereira HS et al. (2017). Rev Ceres, 64: 159-166.
Silva GS et al. (2008). Crop Breed Appl Biotech, 8: 299-304.

COMPARATIVE ANNUAL GRAIN LEGUME ROOT ARCHITECTURE

Jim Burridge and Jonathan Lynch

Penn State University

Grain legume production is fundamental to many smallholder and subsistence farmers and to local and regional economies. Suboptimal water and phosphorus availability are primary limitations to production, which typically have contrasting availabilities in the soil profile when both are limiting. Root architecture is important for water and phosphorus acquisition but tradeoffs between soil exploration strategies may reduce the benefits of breeding for single stress environments. Panels of common bean (*Phaseolus vulgaris)*, tepary bean *(Phaseolus acutifolius)*, cowpea (*Vigna unguiculata)*, soybean (*Glycine max)*, chickpea (*Cicer arientinum)*, and groundnut (*Arachis hypogaea)* were evaluated for a variety of root architectural characteristics (Fig.1). A smaller collection including lima bean (*Phaseolus lunatus)*, faba bean (*Vicia faba)* and minor *Phaseolus* species were also evaluated. We found that legume root systems can be placed on a root system architecture (RSA) spectrum according to the dominance of embryonic or post-embryonic roots (Fig.2). This classification corresponds to putative water availability in their domestication environment. Root system architecture can also be grouped into categories corresponding to epigeal or hypogeal germination. Hypogeal germinators have a root system composed of epicotyl roots, the primary root and lateral roots. Epigeal germinators have distinguishable hypocotyl and basal roots as well as a primary root and primary root laterals. Epigeal and hypogeal root system categories present different adaptive mechanisms, which may be complemented by a particular life strategy. For instance, the chickpea root system may be best suited for an elongated phenology but parsimonious water use strategy. The tepary root system may be best suited for accelerated phenology with dynamic but potentially elevated water use. All strategies likely involve balancing tradeoffs and opportunity costs. We identified inverse relationships among investments in different root classes in most species and between indicators of deep and shallow exploration in all species. Bean and tepary showed particularly strong tradeoffs in investment patterns, while chickpea and groundnut show less pronounced tradeoffs. These tradeoffs may be formed by interactions between resource availability, resource acquisition strategy and life strategy in a given domestication environment. Using an economic analysis to understand the advantages and disadvantages of various root architectures may help to maintain and expand the range of these important food security grain legumes. We highlight instances of dimorphic root architectures that may co-optimize resource acquisition in environments with contrasting edaphic resource availability profiles.

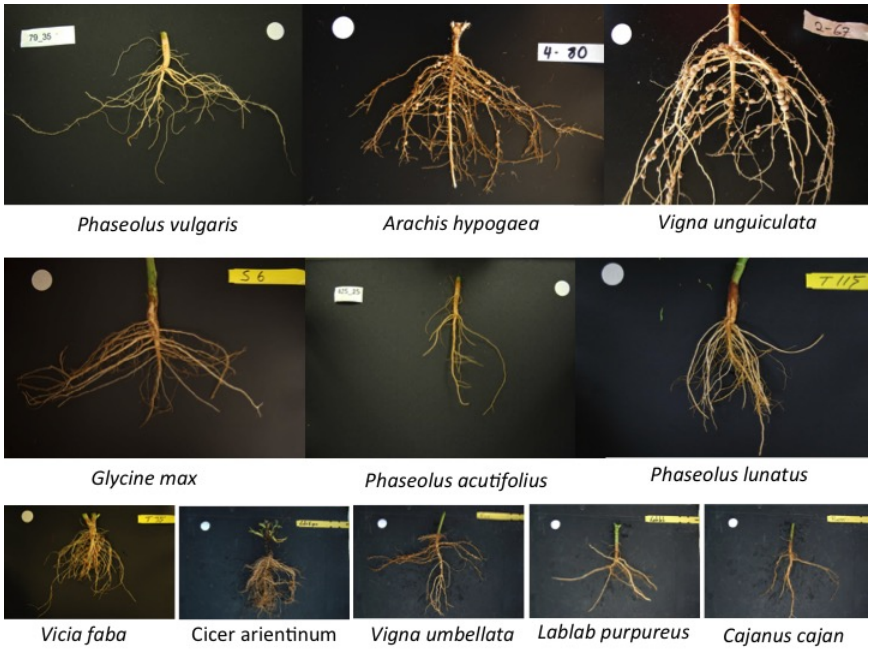


Figure 1: Representative images showing species level variation of crown root architecture. Roots were grown in the field, excavated manually and washed with water before photographing. Circular scale has diameter of 25mm in all images.

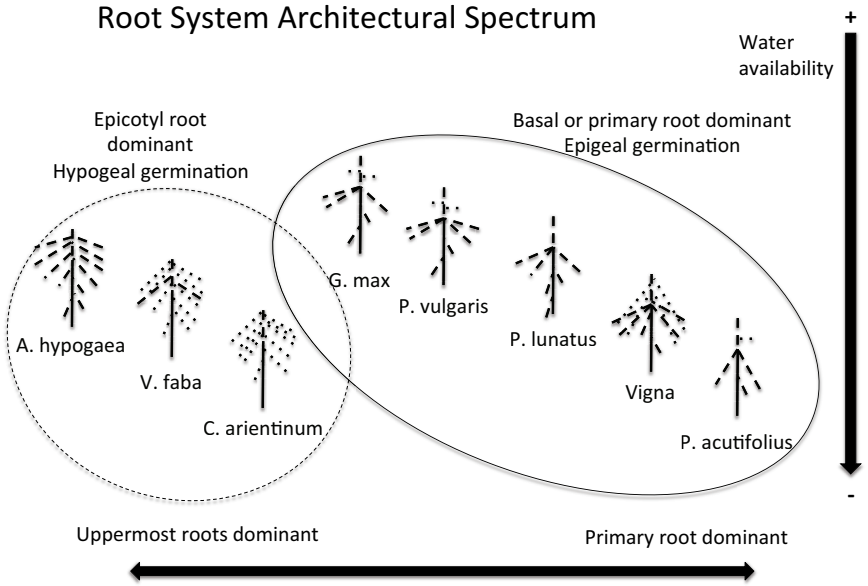


Figure 2. Simplified drawings of crown root architecture of various legumes arranged on the root system architectural spectrum (x-axis) according to dominant root class and estimated water availability in domestication environment (y-axis). Historical weather data acquired from World Bank 1901-1930 averages for SE Jalisco, Mexico (*P. lunatus*), Northern Argentina (*A. hypogea*), Guerrero, Mexico (*P. vulgaris*), North-Central Nigeria (*Vigna unguiculata*), SE Turkey (*C. arientinum*), East China (*G. max*), Tucson, Arizona (*P. acutifolius*), Mt. Carmel, Israel (*V. faba*). http://sdwebx.worldbank.org/climateportal/index.cfm?page=country_historical_climate&ThisCCode=MEX

GENOMICS OF GENOTYPE BY ENVIRONMENT INTERACTIONS IN THE COOPERATIVE DRY BEAN NURSERIES

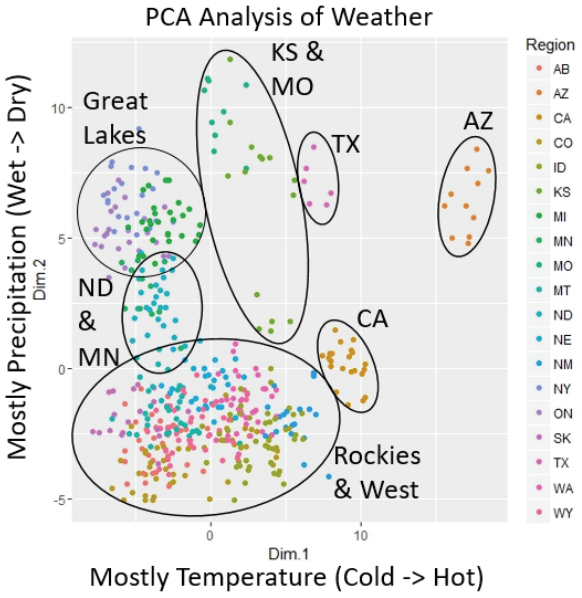
Alice MacQueen[1], Phil McClean[2], Tom Juenger[1]

[1]The University of Texas at Austin, Integrative Biology (alice.macqueen@utexas.edu)
[2]North Dakota State University, Department of Plant Sciences

Common bean (*Phaseolus vulgaris*) yields have been improved by multiple long-term breeding efforts across the world. One example of such an effort is the Cooperative Dry Bean Nursery (CDBN), an ongoing 60+ year collaboration across the United States and Canada. CDBN collaborators have collected phenotypic data for a suite of agronomic traits for over 500 common bean varieties grown in 74 locations. Despite yield improvements, large genotype-by-environment interactions (GxE) persist in common bean. Though genomics-assisted breeding tools and methodological frameworks to study GxE are rapidly improving (Heffner *et al.,* 2009; Perez & de los Campos 2014), accurate phenotyping in relevant field conditions remains a major limitation of these analyses. Major phenotyping efforts such as the CDBN, when combined with genomic data, offer unparalleled opportunities to determine how major genetic factors affect genotype by environment interactions.

In collaboration with current common bean sequencing efforts (Moghaddam *et al.*, 2016), we sequenced 314 varieties from the CDBN and established a genome-wide association mapping population. We obtained monthly weather data associated with each CDBN trial year at each site, and performed a principal components (PC) analysis on all weather variables. The first two PCs explained 39.1% and 22.6% of the variance, and loaded strongly with temperature and precipitation variables, respectively. These PCs separate the CDBN trial years and locations into seven geographic regions (Figure 1). To reduce the location complexity in this dataset, we determined trait averages for each variety within these seven geographic regions for yield, days to flowering, days to maturity, and plant height. We also determined the standard errors about the means within each geographic region as a measure of within-region plasticity for each phenotype.

Figure 2. *Principle component analysis of monthly weather data for 34 years and 35 CDBN locations. Weather variables include temperature, precipitation, cloud cover, vapor pressure, and others.*



We used TASSEL to conduct genome wide association studies (GWAS) on the CDBN panel for both phenotypic means and standard errors about the means within each geographic region. We first determined genomic regions that were differentially associated with phenotypes in the seven geographic regions within the CDBN, or genomic regions with GxE. For the top 100 SNPs associated with each mean phenotype, only 14% of 250kb windows contained SNPs associated with the mean yield for more than one geographic region (Figure 2). Low overlap was also observed for yield standard errors (15% overlap) and

other phenotypes (data not shown). This lack of overlap between SNPs implied that there is substantial GxE in this dataset, or many genomic regions that affect phenotypes only at specific geographic locations. We also determined whether genomic regions uniquely affected phenotypic means, phenotypic plasticity, or both traits. Interestingly, 51.9% of 250kb windows contained SNPs associated with both yield means and yield standard errors (Figure 3). This implies that many genomic regions in common bean might affect both yield levels and yield robustness. This data will be released by 2019 as a new resource on the genomics of GxE within the historical CDBN dataset.

Figure 3. *Genomic regions with top GWAS hits typically affected only one geographic region. Plot shows the distribution of 250kb genomic windows with at least one SNP in the top 100 SNPs for mean yields for 7 geographic regions.*

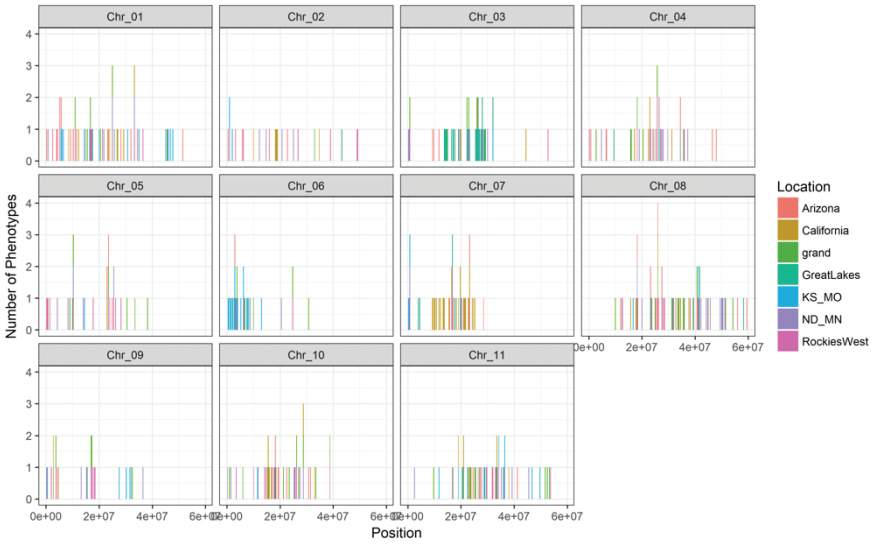
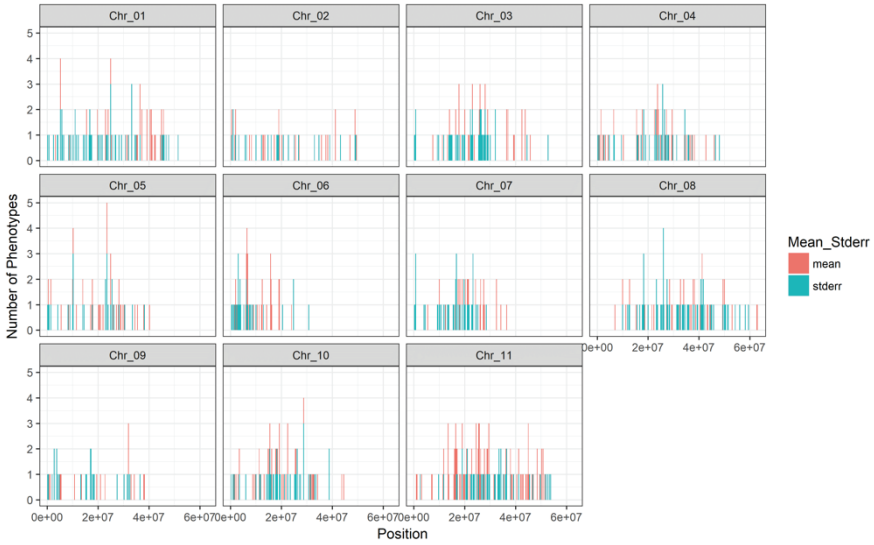


Figure 4*. Genomic regions with top GWAS hits typically affected both yield means and standard errors about the mean. Plot shows the distribution of 250kb genomic windows with at least one SNP in the top 100 SNPs for both yield means and yield standard errors in the seven geographic regions.*



ACKNOWLEDGEMENTS

We would like to thank all researchers who provided germplasm for this study, including Mark Brick, Jim Kelly, Phil McClean, Phil Miklas, James Myers, Juan Osorno, and Tom Smith. AM was supported by a postdoctoral fellowship grant from the NSF PGRP.

REFERENCES

Heffner EL *et al*. (2011) Genomic selection accuracy using multifamily prediction models in a wheat breeding program. *Plant Genet*. **4**:65-75.

Moghaddam SM *et al.* (2016) Genome-Wide Assocation Study Identifies Candidate Loci Underlying Agronomic Traits in a Middle American Diversity Panel of Common Bean (*Phaseolus vulgaris* L.). *Plant Gen.* **9**(3): 1-21.

Perez P & de los Campos G (2014) Genome-Wide Regression and Prediction with the BGLR Statistical Package. *Genetics* **198**:483-495.

POPULATION STRUCTURE IN COMMON BEAN ACCESSIONS FROM PERNAMBUCO STATE, BRAZIL

Martiniano-Souza MC[2*], Gonçalves-Vidigal MC[1], Valentini G[1], Costa AF[2], Vidigal Filho PS[1], Elias JCF[1], Ariani A[3], and Gepts P[3]

[1]Departamento de Agronomia, Universidade Estadual de Maringá, PR, Brazil
[2]Instituto Agronômico de Pernambuco, PE, Brazil, Departamento de Pesquisa *e-mail: mariamartiniano@hotmail.com
[3]University of California, Department of Plant Sciences, Davis, California, USA

INTRODUCTION

Common bean is one of the most important legume for direct human consumption (Schmutz et al. 2014). It is a source of protein, fiber, iron, carbohydrates, vitamins and minerals for millions of people in developed and developing countries and is one of the basic foods for the populations of South America and eastern and southern Africa (Lin et al. 2008). Brazil, with a population of 207.7 million people, is one of the largest producers and consumers of common bean in the world (Coimbra-Gonçalves et al. 2016). The diversity of common bean is structured in two major eco-geographically distinct gene pools. The large-seeded Andean cultivars were originally distributed throughout the South American countries of Peru, Chile, Bolivia and Argentina, and the small to medium-seeded Mesoamerican cultivars comprise those distributed in Central America, from northern Mexico to Colombia (Gepts and Bliss 1988; Gepts et al. 1998). The objective of this work was to evaluate the population structure of common bean accessions from Pernambuco State to assess their relationships with other common bean varieties, especially those originating from similarly dry regions.

MATERIAL AND METHODS

This study, conducted jointly at the Núcleo de Pesquisa Aplicada à Agricultura (NUPAGRI) of the Universidade Estadual de Maringá (UEM), Paraná, Brazil and the University of California, Davis, analyzed 86 accessions of common bean currently maintained in the Instituto Agronômico de Pernambuco (BAG-Feijão-IPA). Of these, 56 landraces were collected in common bean producing areas from Pernambuco State. Genotyping-by-sequencing (GBS) of the accessions was performed as represented in Figure 1. The analysis was conducted by assigning 10,000 as burn-in and performing 100,000 interactions using the Markov chain Monte Carlo (MCMC) method for K-values from 2 to 15 in STRUCTURE (Pritchard et al. 2000). A similarity matrix was developed in the Tassel 5.0 program (Bradbury et al. 2007). To help interpret the population structure, a neighbor joining tree was produced using the Mega 6.0 program (Tamura et al. 2007).

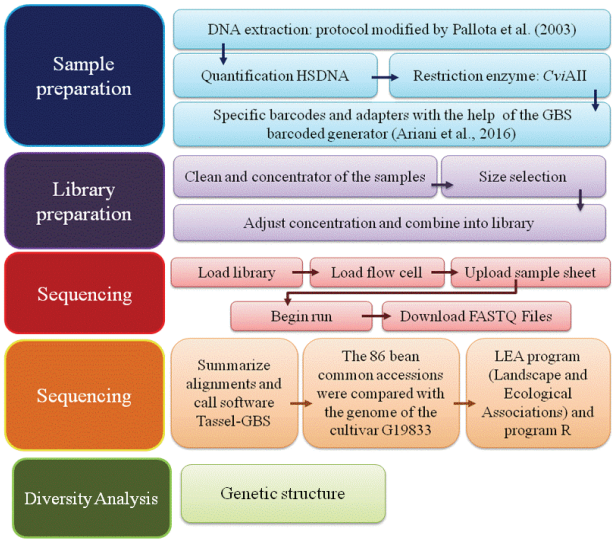


***Figura 1**. Design of the genotyping-by-sequencing analysis.*

RESULTS AND DISCUSSION

A total of 30,529 high-quality SNPs was identified, which were distributed on 11 chromosomes. The number of SNPs per chromosome ranged from 1,731 to 3,853. Population structure analysis among the Pernambuco accessions revealed that the optimum number of subgroups was K = 3 and, at this level, the Mesoamerican gene pool was subdivided into two subgroups, which remained separate from the Andean subgroup. The association of the subgroups with commercial classes of Brazilian beans Mulatinho, Carioca, and Preto Mesoamerican types was observed. The results clearly showed that Mesoamerican accessions exhibited a higher genetic diversity in comparison to the ones of Andean origin (Figures 2 and 3).

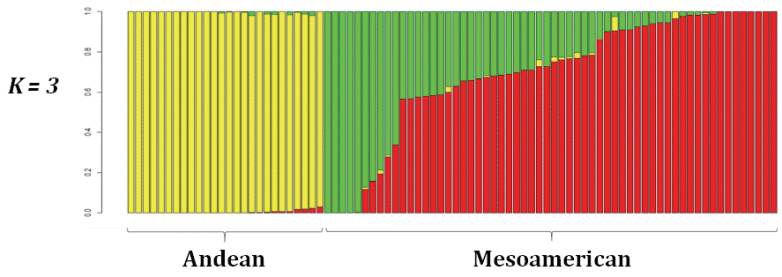


***Figura** 2. Analysis of population structure of 86 common bean accessions based on their genotyping by sequencing profiles. Each color represents one subgroup by Structure in a typical run at the K value of K=3.*

CONCLUSION

GBS revealed the presence of Andean and Mesoamerican genotypes in Pernambuco State, with high genetic diversity. Therefore, the aforementioned accessions are an important potential source of genes for germplasm conservation, and consequently, for future development of cultivars from this region of Brazil.

REFERENCES

Bradbury et al. (2007). Bioinformatics, 23:2633-2635

Coimbra-Gonçalves et al. (2016). Crop Sci. 56:2904–2915

Gepts et al. (1998). HortSci 33: 1124-1130

Gepts and Bliss (1988). Econ Bot 42:86-104

Lin et al. (2008). Food Chemistry 107:399–410

Pallota et al. (2003). In Proc. 10th Int. Wheat Genet. Symp., Paestum, Italy (pp. 1-6)

Pritchard et al. (2000). Genetics 155:945–959

Schmutz et al. (2014). Nature Genet 46:707-713

Tamura et al. (2007). Molecular Biology and Evolution, 24:1596-1599.

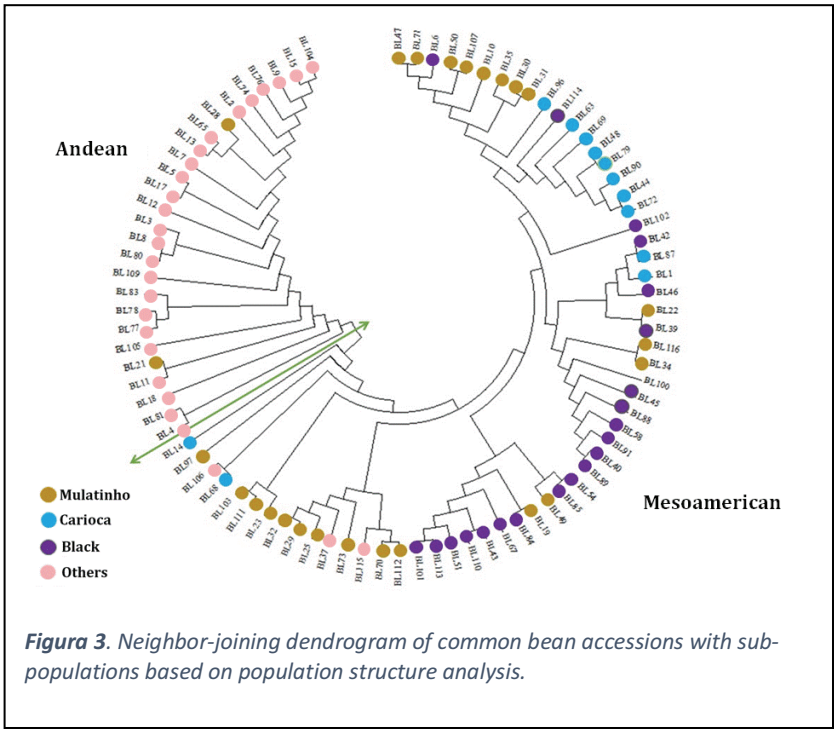


***Figura 3**. Neighbor-joining dendrogram of common bean accessions with sub-populations based on population structure analysis.*

PHENOTYPIC AND GENOMIC VARIATION ASSOCIATED WITH THE CONVERSION OF TYPE III TO TYPE II GROWTH HABIT DURANGO GENOTYPES

McClean PE[1,2], Soltani A[1], Mamidi S[3], Mafi Moghaddam S[1], Osorno JM[1], and Miklas P[4]

[1]North Dakota State University, Department of Plant Sciences, Fargo, ND, USA. (phillip.mcclean@ndsu.edu); [2]North Dakota State University, Genomics and Bioinformatics Program, Fargo, ND, USA; [3]HudsonAlpha Institute for Biotechnology, Huntsville, AL, USA; [4]USDA Vegetable and Forage Crops Research Lab, Prosser, WA, USA.

INTRODUCTION. The growth habit of pinto, great northern, and other race Durango common beans (*Phaseolus vulgaris* L.) was restructured over the last 25 years (Kelly 2001). Breeders converted germplasm with prostrate, indeterminate Type III growth habit into varieties with an upright, indeterminate Type II growth habit. The Type III architecture created a humid environment optimum for disease and required two passes during harvest. The new upright Type II architecture is currently the industry standard and requires only a single pass to combine the crop. Additional breeding within the Type II germplasm resulted in shorter and more upright varieties. The original Type II ideotype is called IIb, while the newer ideotype is called IIa. All type II growth habits aids in reducing humidity in the canopy which in turn reduces disease incidences of diseases such as white mold. We are interested in cataloging the phenotypic differences associated with transition from the Type III to Type II Durango beans, and identifying genomic regions selected during the Type III to Type II transition.

RESEARCH METHODS. In this research, the phenotypic effects of the Type III to Type II conversion were assessed in a Durango Diversity Panel (DDP; n=182) consisting of historic and newly released pinto, great northern, pink, and medium red bean genotypes. The panel was grown in replicated field trials over two years at Prosper, and/or Hatton, ND, USA, and Othello, WA, USA. Data was collected for multiple traits (Table 1) associated with plant architecture, and an analysis of variance was performed on the least square means. All members of the DDP were resequenced to a depth of 8x and mapped against version 2.1 of the common bean reference genome, G19833 (https://phytozome.jgi.doe.gov/pz/portal.html#!info?alias=Org_Pvulgaris). SNPs were then discovered among the genotypes. The number of SNPs was counted for 100kb non-overlapping windows for the type III and type II great northern and pinto genotypes.

RESULTS. Table 1 shows that breeding for Type II architecture resulted in significantly reduced plant length, increased plant height, improved canopy porosity and lodging, reduced internode length, and increased stem diameter. Further improvement in the type II ideotype from IIb to IIa significantly reduced plant length, increased plant height, and improved canopy porosity and lodging. Scans for number of SNPs across 100kb non-overlapping windows within the pinto and great northern populations identified a shared ~600kb Pv07 (34.6Mb – 35.2Mb) region in which the number of SNPs within the Type II genotypes was greatly reduced (Fig. 1). In this interval Type III pintos averaged 236 SNPs per window, while the average for Type II windows in pintos was 4 SNPs. For the great northern genotypes, the Type III population averaged 209 SNPs while the Type II averaged 2 SNPs.

Table 1. Phenotypic means and F-test significance results for plant architectural traits for Type IIa, Type IIb, and Type III members of the Durango diversity panel.

	Mean			F-test		
Trait	**Type IIa**	**Type IIb**	**Type III**	**Genotype**	**Location**	**GxE**
Plant length (cm)	57.7 a	66.7 b	69.6 c	***	**	***
Canopy height (cm)	42.8 a	39.2 b	33.3 c	***	*	***
Canopy porosity (rating)	2.9 a	3.2 b	3.6 c	***	*	ns
Lodging (rating)	3.2 a	4.7 b	6.9 c	***	ns	***
Internode length (mm)	25.2 a	25.8 a	26.9 b	***	**	**
Stem diameter (mm)	5.7 a	5.5 a	5.0 b	***	**	ns
Branch distance (mm)	20.8 a	20.8 a	20.4 a	ns	**	*

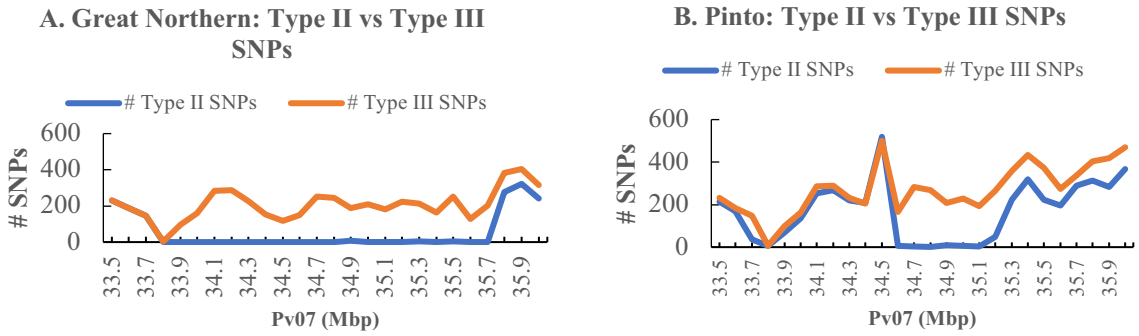


Figure 1. The Type II introgression in Durango varieties. The number of SNPs for Type III and Type II great northern (A) and pinto (B) genotypes are shown for a candidate Pv07 region.

DISSCUSION. Recent Durango breeding efforts produced taller varieties with shorter plant length. The shorter length was correlated with shorter internode length, a trait first established in Type IIb beans. The field analysis confirmed that these plants have improved canopy porosity and lodging. The development of Type IIa genotypes suggest residual variation exists to further improve the ideotype. Localized reduced SNP variability placed the introgression associated with Type into a region on Pv07 that was previously shown to be associated with Type II architecture (Moghaddam et al. 2016). This region contains 58 potential candidate genes.

REFERENCES

Kelly JD. 2001. Remaking bean architecture for efficient production. *Advances in Agronomy*. 71: 109-143.

Moghaddam SM, Mamidi S, Osorno JM, Lee R, Brick M, *et al*. 2016. Genome-wide association study identifies candidate loci underlying agronomic traits in a Middle American diversity panel of common bean. *The Plant Genome* 9. doi: 10.3835/plantgenome2016.02.0012

ACKNOWLEDGEMENT. This material is based upon work that is supported by the National Institute of Food and Agriculture, U.S. Department of Agriculture, under award number 2013-67014-21367.

GENERAL AND SPECIFIC COMBINING ABILITY ANALYSIS FOR EVALUATION OF COMMON BEAN UNDER DROUGHT STRESS

Arruda, I.M.[1]; Gonçalves, L.S.A.[1]; Moda-Cirino, V.[2]

[1]State University of Londrina, Agronomy Department, Londrina – Paraná State – Brazil –
[2]Plant Breeding and Genetic Area, Agronomic Institute of Paraná State, Londrina – Paraná – Brazil. vamoci@iapar.br

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is considered the main legume for human consumption and it is an important source of proteins, carbohydrates and minerals. However, the yield and spatial distribution of this crop are severely affected by biotic and abiotic stresses and among the abiotic factors water deficit is one of the most limiting because it causes reduction in grain yield. The effects of hydric deficit on common beans have been widely studied and depend on the frequency, duration and intensity of stress and the developmental stage of the crop. The selection of superior genotypes for the development of new cultivars is one of the major challenges of breeding programmes, thus, this study aimed to select parents to be used in a breeding program for drought tolerance.

MATERIAL AND METHODS

The general and specific combining ability (GCA and SCA) of the parents were evaluated under conditions with and without water deficit, in partial diallel scheme. The experiment was carried out in a greenhouse and hybridizations were performed between group I, comprised of three Mesoamerican origin bean genotypes drought tolerant (IAPAR 81, BAT 477 and SEA 5) and group II, composed of nine cultivars, widely used by Brazilian farmers. In this last group, six parents were from the Mesoamerican origin, divided into carioca type (BRS Estilo, IPR Campos Gerais and IAC Alvorada), and black type (IPR Uirapuru, IPR Nhambu and BRS Esteio) and three were from the Andean origin (IPR Garça, DRK 18 and BRS Radiante). The experimental design was a randomized block, with four replications. The plants were grown in pots with substrate under 80% of pot capacity (PC) up to R5, appearance of the first floral bud, when water supply was restricted to 30% PC for 20 days for the pots under stress treatment. The plants were evaluated at physiological maturity (R9) for plant height, number of nodes, number of pods per plant, number of seeds per pod, number of seeds per plant, yield per plant and total dry biomass. The treatment effect was decomposed into general and specific combining ability capacities and all statistical analyses were performed on the program GENES (Cruz, 2016).

RESULTS AND DISCUSSION

The values of GCA and SCA indicated the presence of additive and non-additive effects on those traits genetic control and that the hybrid combinations obtained showed significant differences (Table 1). Under water deficit conditions, GCA estimated for group I showed that BAT 477 presented the best results, with positive values for all the characteristics (Table 2). For group II, IAC Alvorada, IPR Uirapuru and BRS Esteio, Mesoamerican cultivars, showed increased for all agromorphological traits too (Table 2). The significance of GCA is of great importance, especially in autogamous species, implying that the higher values for these traits can be fixed throughout the successive generations of self-fertilization to obtain lines superior to the ones currently available.

Table 1. Analysis of variance for agromorphological characteristics of the common bean genotypes evaluated in the presence and absence of water deficit.

Presence of water deficit								
	DF	PH	NN	PP	SP	SPL	yield	TDB
Genotypes	38	2849.45**	12.93**	58.38**	1.87**	1751.33**	112.64**	207.92*
GCA I	2	16351.33**	35.02*	147.64**	1.35	4049.69*	57.50	198.86
GCA II	8	635.41	28.12**	77.65**	5.54**	3583.99**	207.28**	278.37*
SCA	27	2609.69**	6.36**	43.23**	0.88	1006.44**	90.64**	191.50*
Error	114	482.44	4.14	12.41	0.69	436.54	25.46	113.43
Mean		90.67	9.53	10.08	4.24	44.68	11.24	27.16
VC (%)		24.22	21.33	34.96	19.62	46.75	44.83	39.21
Absence of water deficit								
	DF	PH	NN	PP	SP	SPL	yield	TDB
Genotypes	38	4915.11**	19.19**	72.94**	2.24**	2971.68**	164.32**	655.17**
GCA I	2	4091.31*	0.79	114.63*	2.24	5616.74*	230.49*	693.70
GCA II	8	5374.53*	44.92**	67.87	3.97**	3326.34*	218.96**	706.28*
SCA	27	4713.51**	12.02**	71.08**	1.81**	2633.85**	144.73**	650.05*
Error	114	912.48	4.99	33.80	0.74	1250.99	69.27	264.21
Mean		119.25	11.65	15.12	4.95	77.49	19.56	41.47
VC (%)		25.33	19.17	38.44	17.3	45.65	42.55	39.22

PH: Plant height (cm), NN: number of nods, PP: number of pods per plant, SP: number of seed per pod, SPL: number of seeds per plant, yield: yield per plant (g plant^{-1}), TDB: total dry biomass (mg). **,*: significant by F test at 1 and 5 % of probability, respectively.

Table 2. Estimates of the effect from general combining ability evaluated in common bean genotypes cultivated in the presence of water deficit.

Group	Genotypes	PH	NN	PP	SP	SPL	yield	TDB
I	IAPAR 81	2.11	0.10	0.49	-	5.08		
	BAT 477	16.58	0.76	1.38	-	5.11		
	SEA 5	-18.69	-0.86	-1.87	-	-10.19		
II	BRS Estilo		0.74	-0.72	0.00	-3.96	-2.46	-1.85
	IPR Campos Gerais		0.61	-0.92	0.14	-2.72	-2.90	-3.02
	IAC Alvorada		0.58	1.81	0.29	10.26	0.99	3.44
	IPR Uirapuru		0.47	0.93	0.39	7.33	1.73	1.42
	IPR Nhambu		0.46	-1.46	0.00	-6.64	-3.75	-2.26
	BRS Esteio		0.95	3.16	0.58	22.01	4.69	6.05
	IPR Garça		-1.78	-1.92	-0.75	-14.75	-0.99	-2.90
	DRK 18		-0.66	0.09	0.00	-1.90	1.61	-1.58
	BRS Radiante		-1.36	-0.95	-0.65	-9.61	1.08	0.70

[1/]PH: Plant height (cm), NN: number of nods, PP: number of pods per plant, SP: number of seed per pod, SPL: number of seeds per plant, yield: yield per plant (g plant^{-1}), TDB: total dry biomass (mg).

The choice of parents for formation of segregating populations is crucial for the success of breeding programs. In this sense, the results of this study indicated that BAT 477 genotype should be considered an important parent in the development of cultivars for drought tolerance.

REFERENCES

Cruz, C. D. Genes Software – extended and integrated with the R, Matlab and Selegen. Acta Scientiarum Agronomy 38, 547-552, 2016.

TOTAL PHENOLYS, CONDENSED TANNINS, FLAVONOIDS, PHYTATES AND ANTIOXIDANT ACTIVITY IN *Phaseolus vulgaris* L. OF ANDEAN ORIGIN

Kajiwara, V.[1]; Ribeiro, L.B.[1]; Scholz, M.B.S.[1]; Moda-Cirino, V.[1]

[1]Agronomic Institute of Paraná State - IAPAR, Celso Garcia Cid Road, Km 375, 86047-902, Londrina, PR, Brazil. vamoci@iapar.br

INTRODUCTION

Bean is a crop of great economic and social importance and contributes to human food supplying essential nutrients. Its consumption has been associated with health benefits and the treatment of diseases, mainly due to the presence of antioxidant substances. The antioxidant activities (AA) of beans are associated with several phenolic compounds, phytate and the coloration of the grain tegument, since colored grains in general have higher antioxidant effects than white grains (Ombra et al., 2016). The common bean has a great variability of physical and nutritional characteristics. *Phaseolus vulgaris* L. breeding programs search for genotypes to be sources of this genetic variability in order to increase the nutritional value of new cultivars. The objective of this study was to identify the variability of compounds with antioxidant activity in beans genotypes of different Andean origins.

MATERIALS AND METHODS

Fifteen genotypes of beans belonging to the Andean origin center (Table 1), were cultivated at the experimental field of the Agronomic Institute of Paraná, Guarapuava, PR, Brazil, from October 2016 to January 2017, corresponding to the rainy season. The beans were finely ground, and antioxidants compounds were extracted with the ethanol / water / formic acid mixture (70: 29.5: 0.5) kept in a dark place and at room temperature for 16 hours. After that, the samples were centrifuged at 3,500 rpm for 12 minutes at 10 ° C. The total phenols, condensed tannins, flavonoids and AA were determined by the methodology proposed by Heimler, et al., (2005) and phytates were evaluated by Oomah et al., (2008) methodology. All determinations were performed in duplicate.

RESULTS AND DISCUSSION

Among the evaluated genotypes there was a great variation in phytate concentration (10.21 to 16.58 mg g^{-1}) regardless of the grain colors. White beans genotypes showed low concentrations of total phenols, condensed tannins and flavonoids, and consequently low AA. The pinto bean genotypes LPSIA0907 and LPSIA0938 showed high concentrations of phenols, condensed tannins, flavonoids and higher AA (5.09 and 5.86 mg / mg DPPH). It was observed positive correlation between the phenolic compounds and AA, suggesting the participation of these compounds in AA (Table 2). Beans with the tegument red and cranberry presented intermediate AA. Antioxidant activity evaluated, as DPPH, ranged from very low levels in white beans to high levels for pinto genotypes (5.09 mg extract / mg DPPH). These values are due to the higher values of phenolic compounds and phytate in colored tegument beans compared to white tegument beans. Among pinto colored genotypes, the LPSIA0907 showed the highest AA. Studies carried out with beans of the groups black, light brown with stripes, red, yellow, white among others (Ranilla et al., 2007) showed that these compounds were also responsible for the antioxidant capacity of black, red and pinto beans. Ombra et al., (2016) obtained similar results in study with beans of the different colorations, and the beans with white staining exhibited a lower amount of phenolic compounds and less AA than colored grains. There is genetic variability among the different

genotypes of *P. vulgaris* to be used as a source of nutritional value improvement, however, and it is recommended that these crosses involve colored genotypes to increase the AA.

Table 1. Mean values of total phenols (mg /100g), condensed tannins (mg /g), flavonoids (mg/g), phytate (mg/g) and antioxidant activity expressed as EC50 (mg/mg DPPH) in *Phaseolus vulgaris* genotypes.

Genotypes	Tegument color	Total phenols (mg/100g)	Condensed tannins (mg/g)	Flavonoids (mg/g)	Phytate (mg/g)	Antioxidant activity (mg/mg DPPH)
BRS Ártico	White	0,54 ± 0,01	0,009 ± 0,05	0,49 ± 0,04	16,04 ± 0,32	48,18
IPR Garça	White	0,54 ± 0,00	0,015 ± 0,11	0,58 ± 0,02	11,25 ±0,53	39,10
LP0506	White	0,52 ± 0,02	0,016 ± 0,21	0,57 ± 0,03	13,28 ± 1,49	16,78
LP0507	White	0,48 ± 0,01	0,011 ± 0,08	0,53 ± 0,02	13,02 ± 0,42	51,68
LP0517	White	0,52 ± 0,02	0,012 ± 0,09	0,53 ± 0,02	14,90 ± 0,35	63,46
LP0601	White	0,52 ± 0,02	0,011 ± 0,06	0,55 ± 0,01	11,24 ± 1,04	26,36
BRS Realce	Cranberry	3,13 ± 0,01	0,553 ± 1,37	2,13 ± 0,04	13,28 ± 1,33	6,58
BRS Radiante	Cranberry	3,43 ± 0,01	0,588 ± 1,00	1,85 ± 0,11	13,68 ± 1,46	6,73
Corujinha	Pinto	2,19 ± 0,04	0,260 ± 0,64	0,99 ±0,02	16,58 ± 0,33	10,64
LPSIA0907	Pinto	3,99 ± 0,01	0,724 ± 2,74	2,07 ± 0,01	11,16 ± 0,67	5,09
LPSIA0938	Pinto	3,83 ± 0,02	0,699 ± 2,25	1,96 ± 0,07	13,33 ± 0,31	5,86
BRS Embaixador	Red	3,14 ± 0,02	0,731 ± 2,37	1,57 ± 0,07	14,13 ± 1,20	6,89
G6416	Red	2,12 ± 0,01	0,229 ± 0.70	1,42 ± 0,04	15,63 ± 2,51	9,97
Kid 44	Red	2,61 ± 0,03	0,417 ± 1,00	1,38 ± 0,05	13,03 ± 0,54	11,48
LP1504	Red	2,58 ± 0,03	0,462 ± 0,64	1,46 ± 0,06	10,21 ± 0,75	7,46

Table 2. Pearson **c**orrelation Matrix

Variables	Total phenols	Condensed tannins	Flavonoids	Phytate	Antioxidant activity
Total phenols	**1**				
Condensed tannins	**0,977**	**1**			
Flavonoids	**0,970**	**0,943**	**1**		
Phytate	-0,103	-0,164	-0,153	**1**	
Antioxidant activity	**-0,816**	**-0,779**	**-0,799**	0,167	**1**

Values in bold are different from 0 with a significance level alpha=0

REFERENCE

HEIMLER, D.; VIGNOLINI, P.; DINI, M.G.; ROMANI, A. Rapid tests to assess the antioxidant activity of Phaseolus vulgaris L. dry beans. J. Agric. Food Chem**.** 2005, 53,3053-3056.

OOMAH, B.D.; BLANCHARD, C.; BALASUBRAMANIAN, P. Phytic Acid, phytase, minerals, and antioxidant activity in Canadian dry bean (*Phaseolus vulgaris* L.) cultivars. J. Agric. Food Chem. 2008, 56, 11312–11319.

OMBRA, M.N.; D'ACIERNO, A.; NAZZARO, F.; RICCARDI, R.; SPIGNO, P.; ZACCARDELLI, M.; PANE, C.; MAIONE, M.; FRATIANNI, F. Phenolic composition, antioxidant and anti-proliferative activities of the extracts of twelve common bean (*Phaseolus vulgaris* L.) endemic ecotypes of Southern Italy, before and after cooking. Oxid. Med. Cell. Longev. 2016

RANILLA, L. G.; GENOVESE, M. I.; LAJOLO, F. M. Polyphenols and antioxidant capacity of seed coat and cotyledon from Brazilian and Peruvian bean cultivars (*Phaseolus vulgaris* L.). J. Agric. Food Chem. 2007, 55, 90-98.

GENOME WIDE ASSOCIATION STUDY (GWAS) FOR WHITE MOLD RESISTANCE IN SNAP BEAN

Haidar Arkwazee[1], John Hart[3], Tim Porch[3], Phil Griffiths[2], Joel Davis[1] and James R. Myers[1]

[1]Department of Horticulture, Oregon State University, Corvallis, OR; [2]School of Integrative Plant Science, Plant Breeding and Genetics Section, Cornell University, Geneva, NY; [3]USDA-ARS-TARS, Mayagüez, PR.

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.; 2n=2x=22) is one of the most widely produced grain legumes globally. White mold, caused by *Sclerotinia sclerotiorum* (Lib.) de Bary, is considered one of the most important diseases that can cause up to 100% yield loss under certain conditions in bean fields. A genome wide association study (GWAS) was conducted to detect markers significantly associated with white mold resistance in a panel of snap bean cultivars. GWAS is one of the most powerful genomics tools recently developed to detect the genes responsible for disease resistances and other important traits in plants, humans and animals. GWAS exploits linkage disequilibrium to characterize association between genomic markers and a trait (Soto-Cerda and Cloutier, 2012). Several statistical models for GWAS have been developed including General Linear Model (GLM), and Mixed Linear Model (MLM) (Lipka *et al*., 2012). Fixed and random model Circulating Probability Unification (FarmCPU) appears particularly effective in controlling the false positive error rate that can be a problem with GLM, and reduces the occurrence of false negatives caused by excessive stringency of the MLM (Liu et al., 2016).The objectives of this study were: 1) to understand the genetic background for the resistance to white mold in snap bean, 2) detect new QTL associated with partial resistance to white mold and 3) verify previously reported QTL.

MATERIAL AND METHODS

Two populations of snap bean were used in this study. The BeanCAP (Coordinated Agriculture Project) Snap Bean Diversity Panel (SBDP) included 150 cultivars and breeding lines. The Snap Bean Association Panel (SnAP) consisted of 382 cultivars and breeding lines, and the SBDP is contained within the SnAP. The seedling straw test (Arkwazee and Myers, 2017) was used to evaluate the SnAP for white mold resistance. Field tests were conducted to evaluate the SBDP for white mold reaction at OSU Vegetable Research Farm in 2012 and 2013, where incidence (%), severity (1-9) and sclerotia number were measured. The population was genotyped using genotyping-by-sequencing (GBS) (Elshire *et al*., 2011) with 40,023 SNPs obtained after filtering for 20% missing data and 2% minor allele frequency. The FarmCPU R package was used to conduct GWAS.

RESULTS

One-hundred forty-six significant SNPs associated with white mold reaction were detected on all common bean chromosomes. Twenty significant SNPs were detected by the seedling straw test, while 126 significant SNPs were detected in one or both years for incidence, severity and sclerotia number. The 146 SNPs were grouped into 39 regions distributed across all chromosomes (Table 1). Thirteen chromosomal regions contained SNPs from one year in the field, 10 regions had SNPs from both years, 10 regions contained SNPs from one year and the straw test, 4 regions had SNPs from both years in the field and straw test, and two regions were identified by SNPs associated with the straw test only. Eight regions corresponded to meta-QTL previously identified by Vasconcellos et al., (2017), five were in regions identified by researchers in other papers, and 25 were newly identified in this study.

Table 1. Thirty-nine regions significantly associated with white mold resistance identified by GWAS of the Bean CAP Snap Bean Diversity Panel and the Snap Bean Association Panel distributed across the 11 common bean chromosomes.

Chr.*	No. SNPs	Mb** Start	End
Pv01	4	2.37	4.85
Pv01	5	43.71	49.92
Pv02	13	3.14	4.61
Pv02	3	27.31	30.39
Pv02	5	35.21	36.95
Pv02	3	43.33	43.80
Pv03	3	0.16	
Pv03	7	2.29	3.87
Pv03	3	5.36	8.03
Pv03	2	17.12	18.03
Pv03	3	33.55	44.48
Pv03	1	51.29	
Pv04	1	0.46	
Pv04	1	10.90	
Pv04	1	39.33	
Pv05	4	0.10	0.39
Pv05	4	4.34	9.58
Pv05	2	22.88	25.47
Pv05	14	37.63	39.74
Pv06	2	2.32	6.26
Pv06	3	18.12	22.34
Pv06	10	25.98	31.34
Pv07	3	0.36	6.45
Pv07	1	34.78	
Pv07	4	47.71	51.57
Pv08	4	0.33	6.970
Pv08	4	11.16	14.70
Pv08	2	32.48	37.24
Pv08	3	56.28	58.52
Pv09	1	0.80	
Pv09	5	8.19	17.73
Pv09	6	26.99	28.62
Pv09	5	35.88	35.95
Pv10	2	3.46	9.90
Pv10	2	14.73	15.11
Pv10	1	25.83	
Pv10	2	40.09	41.66
Pv11	4	2.21	9.20
Pv11	3	31.00	44.77

*Chromosome; Chromosome numbers in bold represent regions where white mold resistance QTL have not been previously identified., **Mega base

REFERENCES

Arkwazee, H. and Myers, J.R. 2017. Seedling straw test: A rapid and resource-efficient method for evaluating white mold resistance. Ann. Rep. Bean Improv. Coop. 60:39-40.

Elshire, R.J., Glaubitz, J.C., Sun, Q., Poland, J.A., Kawamoto, K., Buckler, E.S. and Mitchell, S.E., 2011. A robust, simple genotyping-by-sequencing (GBS) approach for high diversity species. PloS one, 6:e19379.

Lipka, A.E., Tian, F., Wang, Q., Peiffer, J., Li, M., Bradbury, P.J., Gore, M.A., Buckler, E.S. and Zhang, Z. 2012. GAPIT: genome association and prediction integrated tool. Bioinformatics. 28:2397–2399.

Liu, X., Huang, M., Fan, B., Buckler, E.S. and Zhang, Z. 2016. Iterative usage of fixed and random effect models for powerful and efficient genome wide association studies. PLOS Genetics 12:1-24.

Soto-Cerda, B.J. and Cloutier, S., 2012. Association mapping in plant genomes. In: Genetic diversity in plants. InTech.

Vasconcellos, R.C.C., Oraguzie, O.B., Soler, A., Arkwazee, H., Myers, J.R., Ferreira, J.J., Song, Q., McClean, P. and Miklas, P.N. 2017. Meta-QTL for resistance to white mold in common bean. PLoS ONE 12: e0171685.

INTERACTION OF FUSARIUM SOLANI SPECIES COMPLEX AND SOYBEAN CYST NEMATODE ON FUSARIUM ROOT ROT IN DRY BEAN

Shalu Jain [1], Periasamy Chitrampalam [1, 2], Juan M. Osorno [3], Julie S. Pasche [1] and Berlin D. Nelson Jr[1]

[1]Department of Plant Pathology, North Dakota State University, Fargo, ND, [2]Eurofins BioDiagnostics, Longmont, Colorado, and [3]Department of Plant Sciences, North Dakota State University, Fargo, ND.

INTRODUCTION

The North Dakota-northern Minnesota region is the largest producer of dry beans in the United States. Soybean cyst nematode (SCN), first identified in North Dakota in 2003, is now beginning to infest areas where dry bean production is concentrated. The major dry bean classes grown in the area include pinto, navy, black, and kidney beans. SCN can reproduce on dry bean and cause a significant yield loss in some bean classes (Poromarto et al., 2010). In soybean, it was demonstrated that SCN can interact synergistically with *Fusarium virguliforme*, the cause of sudden death syndrome, increasing SDS foliar symptoms. *Fusarium* root rot is common in dry beans in the region; therefore, similar interactions with SCN could cause increased crop losses. The objective of this study was to determine if the presence of SCN could increase root rot severity caused by *Fusarium solani* species complex 11 (FSSC11), one of the fungal root rot pathogens of dry bean in the region.

MATERIALS AND METHODS

Two kidney bean varieties, Montcalm, susceptible to Fusarium root rot and Rosie, moderately resistant, were used in this research. Previous experiments showed that both varieties were susceptible to SCN HG type 0, a prevalent HG type in the region. The plants were grown in the green house in Cone-tainers filled with pasteurized La Prairie silt loam soil and placed in a water bath to maintain a temperature of 27° C at the rooting zone to favor SCN activity on the roots (Poromarto and Nelson, 2009). Each variety was a separate experiment, but conducted concurrently, and plants were inoculated with three levels of SCN (0, 1,000 and 10,000 eggs per 100 cc of soil) and three levels of FSSC 11 (0, 1,000 and 100,000 spores per gram of soil) inoculum. A split plot design was used where Fusarium treatments were whole plots and SCN treatments were subplots with eight replications. The severity of root rot on 1-7 scale (Bilgi et al. 2008) was evaluated six weeks after inoculation. Plant height and dry root weight were also measured. The experiment was performed three times and each experiment was analyzed separately. Plant height and root weight were subjected to analysis of variance using SAS. F-protected least significant difference (P= 0.05) was used as a posteriori multiple comparison test for numeric data. Disease severity was subjected to nonparametric analysis using the LD_CI.sas and F1_LD_F1.sas macros according to the methods outlined by Shah and Madden (1994) for a split plot design. Contrasts were generated for biologically important comparisons of disease severity caused by Fusarium treatments with and without SCN.

Table 1. Effect of soybean cyst nematode (SCN) on root rot severity caused by *Fusarium solani* FSSC11 on two kidney bean varieties.

Treatments	Average root rot severity on two kidney bean varieties					
	Montcalm			Rosie		
	Exp1	Exp2	Exp3	Exp1	Exp2	Exp3
A No SCN + Fusarium low level	3.3	4.9	3.8	1.9	4.1	2.3
B SCN low level + Fusarium low level	3.8	4.5	3.9	3.0	4.8	3.8
C SCN high level + Fusarium low level	3.4	5.5	5.3	3.4	5.6	5.2
D No SCN + Fusarium high level	2.6	6.0	3.0	2.5	4.0	4.0
E SCN low level + Fusarium high level	3.9	5.3	4.1	4.0	4.6	3.8
F SCN high level + Fusarium high level	4.9	6.1	5.8	4.0	5.6	3.8

Contrasts	Montcalm			Rosie		
	Exp1	Exp2	Exp3	Exp1	Exp2	Exp3
A vs. B	NS	NS	NS	NS	NS	**
A vs. C	NS	NS	*	**	***	***
D vs. E	**	**	NS	*	*	NS
D vs F	***	NS	***	**	**	NS
B vs. C	NS	NS	NS	NS	*	*
E vs. F	**	**	*	NS	NS	NS

EXP = experiment; *, **, and *** = significant increase in root rot severity at P = 0.05, 0.01 and 0.001, respectively. NS = not significant

RESULTS AND DISCUSSION

In Montcalm, a significant increase (α=0.05) in disease severity was observed in two of three experiments at the higher fungal inoculum rate; in one experiment at low SCN and two experiments at high SCN (Table 1). In only experiment 3 there was a significant increase in disease with the low level of fungal inoculum at high SCN. In addition, there was greater disease at high SCN compared to the low SCN at the higher fungal inoculum in all experiments. In Rosie, a significant increase in disease severity was observed only in experiment 3 at the low SCN level and low fungal inoculum, but at the high SCN level and low fungal inoculum, a significant increase in disease severity occurred in all three experiments. With high fungal inoculum, in two out of three experiments there was a significant increase in disease at both the low and high SCN levels. In addition, in two out of three experiments at the low fungal inoculum and low SCN, there was more disease at the higher SCN level. There were no significant differences in plant height or root weight among treatments with SCN and those with SCN plus fungal inoculum. The results of these experiments indicate that the SCN-*Fusarium* interaction can have a significant impact on disease severity not only for a root rot susceptible variety, but also for a moderately resistant variety. Furthermore, the results with Rosie suggest that SCN could reduce or possibly negate genetic resistance to root rot pathogens in dry bean. These results need to be further tested under field conditions to determine the complete effect of this interaction on growth and yield.

REFERENCES

BILGI, V. N., BRADLEY, C. A., KHOT, S. D., GRAFTON, K. F., AND RASMUSSEN, J. B. 2008. Plant Dis. 92:1197-1200.
POROMARTO, S. H, AND NELSON, B. D. 2009. Plant Dis. 93: 507-511.
POROMARTO, S. H., NELSON, B. D., GOSWAMI, R. 2010. Plant Dis. 94: 1299-1304.
XING, L. AND WESTPHAL, A. 2013. J. Pl. Dis. Prot. 120: 209-217.

VARIATION AMONG EDIBLE PODDED SNAP BEAN ACCESSIONS FOR POD AND SEED SUGAR CONTENT

Gartner, W[1], Nienhuis J[1], Bethke, P[1, 2], Kisha T[3]

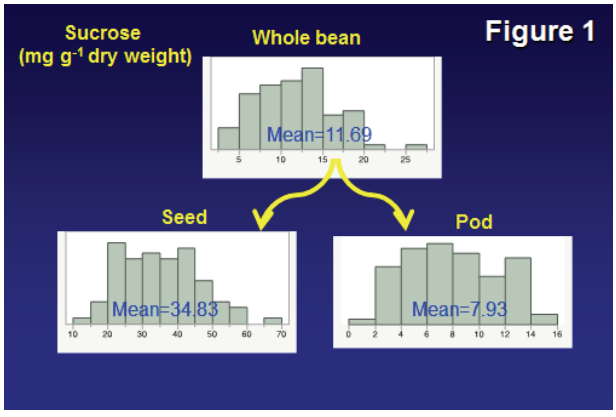
[1]University of Wisconsin, Department of Horticulture (nienhuis@wisc.edu)
[2] USDA-ARS, Madison, WI; [3]USDA-ARS, Pullman, WA

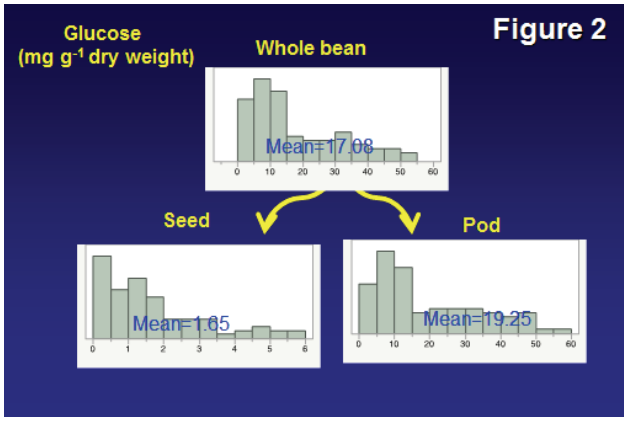
Sugar content of immature snap bean (*Phaseolus vulgaris*) pods and the effects of sugars on other flavor compounds are important to consumers and affect their food and vegetable choices. The objective of this study was to identify variation within *Phaseolus vulgaris* in relation to sugars that affect flavor and sweetness.

We developed and evaluated over two years (2014 and 2015) a diverse sub-core of 94 Plant Introductions (PI) characterized as snap beans, Romano-types, and other beans eaten as edible immature pods, as well as 20 dry bean PIs. Additional genotypes included a kidney bean (Montcalm, Andean gene pool) as well as 8 snap bean cultivars representing various market classes consumed as edible green pods. Pods were sampled when they reached sieve size 4 (a common commercial fresh market size) measured 90 degrees off the suture.

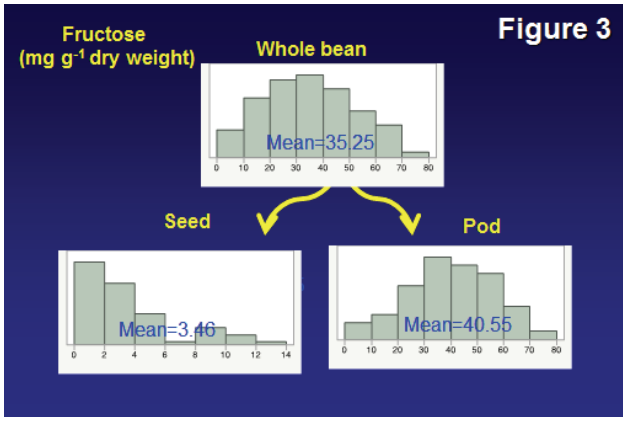
Large positive Spearman rank correlations were observed between years for whole pod glucose (0.89), fructose (0.73) and sucrose (0.62) concentrations, indicating that sugar concentrations were relatively consistent over years. A large positive correlation (r=0.85**) was observed over years between the simple sugars glucose and fructose. In contrast, a large negative correlation was observed between the disaccharide sucrose with both monosaccharides, glucose (r=-0.67) and fructose (r=-0.68). Relationships between mono and disaccharides are consistent with the hypothesis that sucrose is hydrolyzed by acid invertase.

Glucose and fructose are simple sugars, and represent water soluble immediate sources of energy for growth and development in pods. In contrast, sucrose is a water soluble transport sugar that may be synthesized in pods or imported from other parts of the shoot. The population of 87 edible podded and check accessions was partitioned into pod tissue and seed tissue data sets. We observed that glucose and fructose accumulated mainly in the pod tissue (Fig. 2 and 3); in contrast, sucrose accumulated primarily in seeds (Fig. 1).





Figures 1, 2 and 3 show the histogram distributions for sucrose, glucose, and fructose concentrations across among 87 accessions of edible podded beans.



In the pod tissues, fructose concentrations ranged from near 0 to over 70 mg g^{-1} dry weight (Fig. 3). Glucose concentrations ranged from near 0 to over 40 mg g^{-1} dry weight (Fig. 2). Pod tissues contain twice as much fructose (mean 40.55 mg g^{-1} dry weight) as glucose (mean 19.25 mg g^{-1} dry weight). In pod tissue, sucrose concentrations were lower than the other two sugars, ranging from 0 to over 14 mg g^{-1} dry weight and a mean of 7.93 mg g^{-1} dry weight (Fig. 1).

In the seed tissues, fructose concentrations ranged between 0 to 14 mg g^{-1} dry weight, while glucose concentrations ranged between 0 to 6 mg g^{-1} dry weight, respectively (Figs 2 and 3). In a pattern similar to pod tissues, the seed tissues also contain twice as much fructose (mean 3.46 mg g^{-1} dry weight) compared to glucose (mean 1.65 mg g^{-1} dry weight). Sucrose concentrations were much higher in the seed than the other two sugars, with a mean of 34.83 mg g^{-1} dry weight and a range between 10 to 70 mg g^{-1} dry weight (Fig. 1).

Fructose is the sweetest sugar of the three, followed by sucrose and glucose respectively (Fructose is 117% as sweet as sucrose, while glucose is only 74% as sweet as sucrose [Joesten et al. 2007]). Fructose had the highest concentration of whole bean sugars; double the concentration of glucose and three times the concentration of sucrose. Fructose is likely the most important contributing factor to the overall sweetness and flavor profile of edible podded snap beans. Sucrose is mainly accumulated in the seeds which are typically undesirable and small sized in edible podded snap beans and thus contributes little to the overall sweetness profile. We included a wide array of mostly edible podded beans, some with relatively large seeds; thus, cultivars with smaller seeds would be expected to have a low sucrose concentration. Although glucose was more abundant in whole beans than sucrose, the lower sweetness rating (0.74) for glucose indicates that sucrose and glucose contributed approximately equally to bean sweetness.

REFERENCES

Joesten, M., M.E. Castellion, and J.L. Hogg. 2007. The world of chemistry: Essentials. 4th ed. Thomson Brooks/Cole, Belmont, CA

SNPS IDENTIFIED FOR COMMON BACTERIAL BLIGHT RESISTANCE IN DRY BEAN

Simons KJ[1], Lamppa RS[1], McClean PE[2], Osorno JM[2], and Pasche JS[1]

[1]Department of Plant Pathology, North Dakota State University, Fargo, ND, USA
[2]Department of Plant Sciences, North Dakota State University, Fargo, ND, USA

INTRODUCTION: Common bacterial blight (CBB) on dry edible bean (*Phaseolus vulgaris*), caused by *Xanthomonas axonopodis* pv. *phaseoli* (*Xap*), is an important disease worldwide. Lesions can occur on leaves, stems, pods and seeds, causing reductions in seed quality and yield losses up to 50% due to loss of photosynthetic area. Host resistance is controlled by minor and major QTL identified across all eleven chromosomes of *P. vulgaris* (reviewed in Miklas *et al.* 2006). Using resistant varieties is a key management practice. Markers would ease the selection process during development of resistant varieties.

MATERIALS AND METHODS:
Phenotyping: Breeding lines belonging to both the Andean and Middle American gene pools from the NDSU dry bean breeding were evaluated for CBB resistance. One unifoliate and two trifoliate leaflets were inoculated per plant at two points on either side of the mid-vein with a single *Xap* isolate in a walk-in growth chamber. Each leaflet was rated at 14 days post inoculation on a 1 to 9 scale (van Schoonhoven and Pastor-Corrales 1987) and frequency distributions of the mean disease rating were generated (Fig. 1).

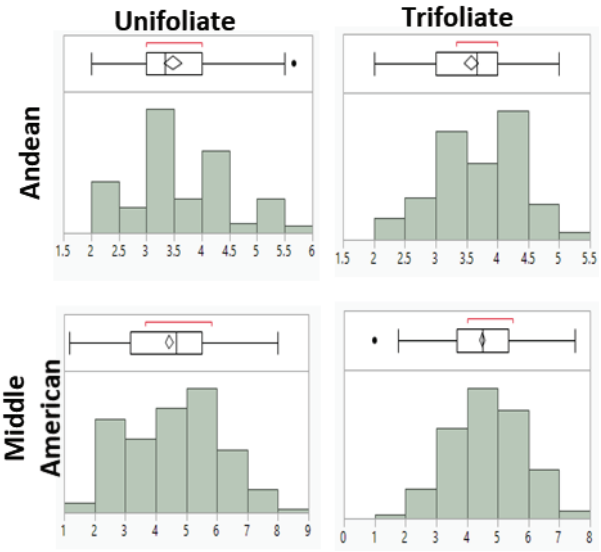


Figure 1. Distribution of mean common bacterial blight (CBB) reactions for Andean and Middle American breeding lines on unifoliate and trifoliate leaflets.

Genotyping: DNA from each line was extracted using the Omega biotek Mag-bind Plant DNA Plus kit. Barcoded sequencing libraries were generated, pooled and sequenced on an Illumina Hi-Seq in rapid run mode. Sequences were quality trimmed, aligned to the *P. vulgaris* reference genome v2.0 using BWA, and variants called within the Andean and Middle-American gene pools using GATK (Maghaddam *et al.* 2016). JMP Genomics 8 was used to identify SNPs associated with CBB resistance testing four models.

RESULTS: Of the 821 lines examined, 131 exhibited resistance at the unifoliate and trifoliate stage. Eighty additional lines were resistant at the unifoliate stage, and 78 at the trifoliate stage. Lines clustered together primarily by race and not by market class (data not shown). The best model for association mapping included both individual relatedness (kinship) and population structure variables. A previously unidentified region of resistance consisting of SNPs spanning a 1.6 kb region was identified in the breeding lines in the Andean gene pool at both the unifoliate and trifoliate stages (Fig. 2). Most of the previously described CBB QTL were identified with

genetic marker(s) lacking full sequence information. As such, many of these QTL are not located definitively on the physical sequence. Comparison of the GWAS results including 4 chromosomal regions identified within the Andean gene pool and 9 chromosomal regions in the Middle American gene pool suggest these regions may be the physical locations of previously described QTL.

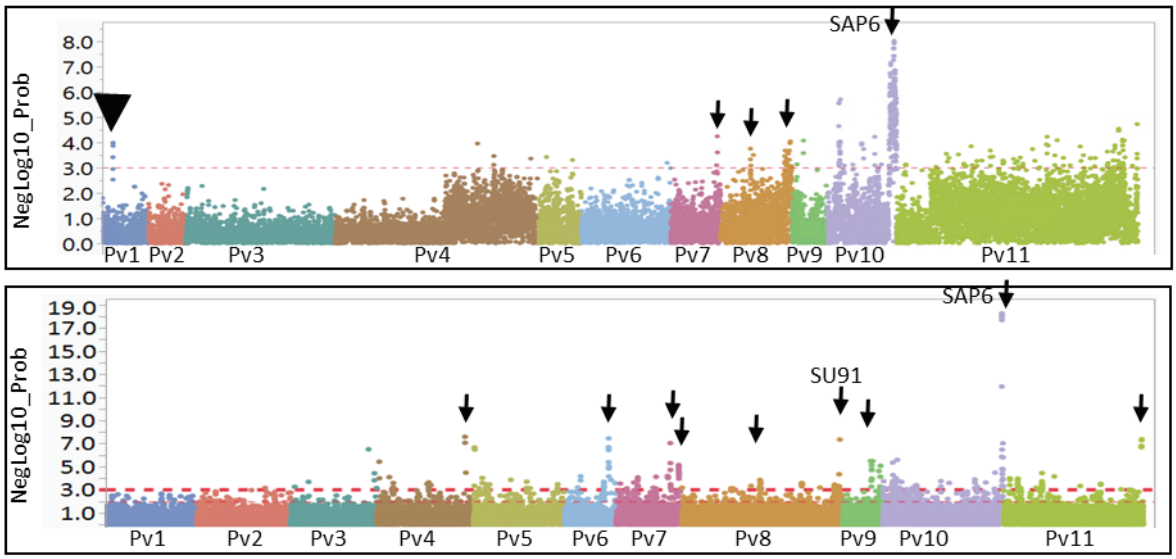


Figure 2. Association mapping in Andean breeding lines (top) and Middle American breeding lines (bottom) using the mean common bacterial blight reaction on the unifoliate leaves. Black arrows depict probable previously described QTL. The solid triangle depicts a potential new QTL. Manhattan plots not shown for the trifoliate association mapping.

CONCLUSIONS: SNPs underlying each associated resistance region can be verified and subsequently used in developing universal markers for marker assisted selection in multiple market classes. The SNP data generated here for breeding lines from both gene pools can be further utilized to identify SNPs associated with agronomic and other disease resistance traits.

ACKNOWLEGEMENTS: This publication was supported by the U.S. Department of Agriculture's (USDA) Agricultural Marketing Service through grant 15-SCBGP-ND-0026. Its contents are solely the responsibility of the authors and do not necessarily represent the official views of the USDA.

REFERENCES

Miklas, PN, Kelly, JD, Beebe, SE, Blair, MW (2006) Common bean breeding for resistance against biotic and abiotic stresses: From classical to MAS breeding. *Euphytica* 147:105-131.

Moghaddam, SM, Mamidi, S, Osorno, JM, Lee, R, Brick, M, *et al*. (2016) Genome-Wide Association Study Identifies Candidate Loci Underlying Agronomic Traits in a Middle American Diversity Panel of Common Bean. *The Plant Genome* 9. doi: 10.3835/plantgenome2016.02.0012.

van Schoonhoven, A, Pastor-Corrales, MA (1987) Standard System for the Evaluation of Bean Germplasm. Cali, Colombia: Centro Internacional de Agricultura Tropical.

CLIMBING BEAN BREEDING FOR HIGH IRON CONTENT AND DISEASE RESISTANCE

Portilla A.E, Mayor-Duran V.M, Buendia H, Blair M.W, Cichy K, Raatz B.

International Center for Agriculture, Cali, Colombia,
email: a.e.portilla@cigar.org, b.raatz@cigar.org

INTRODUCTION: Climbing beans are the farmer preferred growth type in some regions in ES Africa and LAC (dominant in Rwanda and Colombia), due to higher yields and resilience. *Bean common mosaic virus* (BCMNV) is the most common and destructive poty virus known to infect common bean worldwide. Also Anthracnose, caused by the fungus *Colletotrichum lindemuthianum* can cause devastation to farmer's fields, resulting in yield losses as high as 95% in susceptible cultivars (Zuiderveen *et al.,* 2016). The work aims were to identify new sources of anthracnose and BCMNV resistance, high seed iron content, commercial color and good agronomic traits. in other hands in this work validated the usefulness of SNP markers tagging bc-3 and I genes for BCMNV and Co-3 for anthracnose.

MATERIAL AND METHODS: The ENF/CGA lines were codified resulted from agronomic evaluation in field. These lines are coming from double and triple crosses between parents with virus and anthracnose resistance, high seed iron and good agronomic qualities. This panel of 316 samples was tested in Popayan (Colombia) to anthracnose. Disease severity was evaluated with the CIAT standard scale from 1 (no disease symptoms) to 9 (severe disease symptoms). The reaction to viral infection BCMNV was evaluated in the green house with the CIAT scale with 5 categories M= Mosaic (susceptible), N= necrotic reaction, N= No symptoms (Resistant), Local lesions and V= variable. From this panel were selected 80 lines outstanding by commercial qualities and diseases resistance to do a yield trial in two localities, Darien and Popayan. In this essay were included 20 checks with good performance and acceptation by the farmers. An alpha lattice design 10 x 10 with 3 repetitions was used to evaluate the agronomics variables. The data were analyzed with Plant breeding tools (PBTools) Version 1.3. For the validation of SNP genotyping the services was provided by INTERTEK

RESULTS AND DISCUSSION: The most important results show 24 resistant lines for BCMNV, 16 resistant lines for anthracnose, 36 resistant lines for both diseases and 290 were susceptible for both diseases. All the lines have good grain quality.

BCMNV resistance marker and Anthracnose marker validation: BCMNV greenhouse phenotyping and SNP genotyping resulted in significant association (r: 0.88 ***) for *bc-3* and *I* gene. This result suggests the use of this marker like a good molecular breeding tool (Bello *et al*., 2014) (Fig 1). The same results had to anthracnose marker (Co3) were found a significant p-value in a T-test demonstrating an association of the marker with the anthracnose response, and a good marker to select resistant lines (Table 1). These results show the genetic potential of the lines which can be used like resistant genes sources and be released in some regions interested to smallholder farmers.

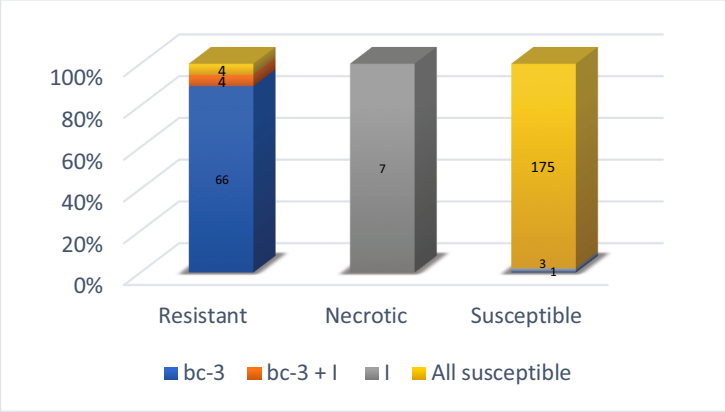


Figure 1. Phenotypic BCMNV Evaluation and bc-3 and I gene genotypes

Table 2. Anthracnose resistance genes evaluation.

Marker	Ant_Pods *p*-value	Ant_leaves *p*-value	Marker effect	Positive Allele
Co-u_ss715648452	0.014	0.022	0.848	G:G
Co-1_ss715646578	0.020	0.019	-0.764	A:A
Co-3_ss715640025	0.023	0.033	-1.346	G:G
Co-4_08_CG_2329860	0.589	0.561	-0.608	G:G

Significance *p* value evaluated by t-test

Interesting traits

The yield results were strongly correlated between locations (r: 0.89 ***). ENF 25, 27 and 31 got results above the average outstanding in all the traits evaluated. In addition, high contents of Fe and Zn were found. Lines with Fe values > 80 ppm with a good yield and commercial seed types can be a good alternative for producers and consumers. In this study a new trait was a canning quality, where the results are promising for the industry markets and final consumers.

Table 3. Highlight lines that combine excellence in at least two traits

Line	Yd*Dar	Yd*Pop	Fesd	Znsed	APP	BCNMV	ANTLF	ANTPF	PSC
ENF163	2143.7	1985.4	***82.4***	**45.0**	***4.24***	M	1	1	red
ENF172	***4424.6***	**4535.3**	72.0	32.7	1.90	V	2	2	red
ENF182	***4382.9***	3849.4	63.7	30.3	***4.57***	M	1	1	red
ENF25	**3819.2**	**3856.8**	**72.38**	**36.3**	**2.48**	O	2	2	red
ENF27	**3984.3**	**4033.0**	**72.81**	**35.6**	**2.81**	O	1	1	red
ENF31	***4381.08***	**3547.6**	***77.98***	**34.4**	**2.62**	O	3	3	red
ENF85	***4468.91***	3789.5	***86.64***	**39.7**	3.00	M	3	3	wh
Aver.	***3593.3***	***3507.59***	***69.24***	***34.85***	***2.5***				

Yd *Average of Dar/Pop*
BCNMV *Bean Common Mosaic Necrosis Virus (M = mosaic, O = resistant, V = variable)*
ANTLF *Anthracnose on leaves in field 1 to 9.*
ANTPF *Anthracnose on pods in field*
APP 1:worst, 5:best
PSC Principal seed color

References: Zuiderveen et al. (2016), *PLoS ONE*, *11*(6); Bello et al (2014), BMC Genomics 15:903. Naderpour et al., (2010), Mol Plant Pathol 11:255–263.

BEAN PASTE QUALITY OF SELECTED WHITE AND YELLOW GENOTYPES AS A NEW FOOD APPLICATION

Rie Sadohara[1]* and Karen A. Cichy[1,2]

[1] Michigan State University, Department of Plant, Soil and Microbial Sciences, 1066 Bogue Street, East Lansing, MI 48824 *Presenter (sadohara.rie@gmail.com)
[2] USDA-ARS, Sugarbeet and Bean Research Unit, 1066 Bogue Street, East Lansing, MI 48824

INTRODUCTION: Bean paste is a Japanese confectionary made of common beans and sugar. Otebo beans are developed specifically for this purpose. Bean paste has a possibility of being used in the US market because of the increasing interest in gluten-free products and health-promoting food. However, the suitability of varieties grown in Michigan for bean paste is unknown. In this study, three Otebo (Fuji, Samurai, Hime as controls), three white (Alpena, Powderhorn, Snowdon) and one yellow (Cebo Cela) genotypes were tested for paste making quality, namely texture and sensory profile including color to assess their potential for a new food application.

MATERIALS AND METHODS: Unsweetened paste was prepared by soaking 110 g of dry seeds for 16 hours, cooking them in 2.5 L of distilled water for their respective 80% cooking time multiplied by 1.52, a factor determined by a preliminary experiment. Cooked seeds were drained, mashed, and passed through a 0.5 mm-sieve to remove seed coat. The filtrate was washed with excessive water three times, and was drained in cheesecloth to yield unsweetened paste.
Sweetened paste was prepared by cooking 50 g of unsweetened paste with 50 mL of distilled water and 30 g of granulated sugar until the paste was 65.0±0.1 g. Stickiness of 41 g of sweetened paste was measured using a TA.XTplus100 equipped with a 25.4-mm cylindrical probe (TA-11), operated by a software Exponent ver 6,1,11,0 (Stable Micro Systems Ltd, Godalming, UK) at a compression rate of 70% and test speed of 1 mm/sec. Negative area recorded while bean paste is pulled by the probe was defined as stickiness.

In sensory evaluation, each genotype was evaluated twice in separate sessions, in which three genotypes were subjected to 5-7 trained assessors. They evaluated the whiteness of sweetened and unsweetened paste, and beaniness, vegetativeness, total flavor intensity, and smoothness of unsweetened paste on a 1-5 scale. Pictures were taken of both pastes using Canon ios T3i at a shutter speed of 1/100 sec, F-number 5.6, and ISO 100 on a black background. L^*, a^*, b^* values were calculated using a software PhotoMeasure [1]. Whiteness index was calculated as $L^* / (a^* + b^*)$ to estimate the strength of whiteness relative to red and yellow hue.

Results And Discussion: Table 1 shows the bean paste qualities of the seven genotypes tested. For stickiness, there was variability within the Otebos with Fuji being the stickiest. Alpena and Powderhorn were as sticky as Samurai and Hime, while Cebo Cela and Snowdon were less sticky. This may be due to their large seed size, which leads to higher water uptake that may make cotyledon cells more swollen and moist. However, a validation would be necessary to confirm this trend.

For color, Cebo Cela, a yellow genotype, was rated as white as Otebo varieties, which are used specifically for bean paste production. On the other hand, Alpena (navy) and Powderhorn (Great Northern) had low scores in whiteness. Whiteness index by image analysis corresponded well with this result, indicating that color measurement can be used as a fast method to predict consumers' perception of paste color.

The sensory evaluation results showed significant differences among genotypes in vegetativeness and flavor intensity ($p<0.0001$, $p=0.001$, respectively). For vegetativeness, Snowdon and Cebo Cela were rated to be more vegetative than the rest. For flavor intensity, Cebo Cela was rated to have the strongest flavor and the rest were the same. The genotype effect on beaniness and smoothness were non-significant, suggesting that they are difficult to evaluate at this number of replications and/or assessors who are not acquainted to consuming bean paste. Alternatively, smoothness could be estimated by median particle size using a particle distribution analyzer [2]. The unsweetened paste of Cebo Cela was rated to be strongly vegetative, which is traditionally not considered desirable, but this strong flavor can be a positive attribute when made into sweetened paste because it reduces overpowering sweetness from sugar. This could be an interesting attribute for further consumer research in countries such as in the US, where beans are consumed as in meals, not dessert. Overall, this study showed that the seven genotypes possess different paste quality attributes, which could be used for developing bean paste products for the US market.

Table 1. Cooking and paste quality of the seven genotypes tested.

		Fuji	Samurai	Hime	Alpena	Powderhorn	Cebo Cela	Snowdon
	Unit	*Otebo*	*Otebo*	*Otebo*	*Navy*	*Great Northern*	*Yellow*	*White Kidney*
Dry seed								
80%CT*	min	140.6a	22.7de	28.2de	61.4bc	73.4b	19.3e	45.6dc
Sweetened paste								
Stickiness	g.sec	2331a	1800ab	1422ab	1997ab	2020ab	1222b	1195b
Whiteness index	ratio	1.6b	2.8ab	2.7ab	2.3b	2.0b	3.2a	2.3ab
Whiteness	point	ND†	4.5ab	4.3ab	3.0c	2.7c	4.9a	4.0b
Unsweetened paste								
Whiteness index	ratio	2.7 b	4.6a	4.5a	3.6ab	3.0b	3.9ab	3.5ab
Whiteness	point	ND	4.9a	4.6ab	3.4c	3.7bc	4.5ab	4.0cb
Beaniness	point	ND	2.3a	2.6a	2.8a	2.5a	3.3a	2.4a
Vegetativeness	point	ND	1.5c	1.6bc	1.7bc	1.3c	3.2a	2.5ab
Total flavor intensity	point	ND	2.4b	2.7b	2.5b	2.9b	4.1a	2.9b
Smoothness	point	ND	3.4a	3.5a	3.2a	3.8a	3.1a	3.0a

Letters in the same row indicate no significant difference (Tukey's HSD, $\alpha=0.05$). * Cooking time. † No data.

REFERENCES

[1] H. Ruiz, H. Diaz, PhotoMeasure, (2017). https://sourceforge.net/projects/photo-measure/.

[2] J. Kato, E. Hosoya, N. Ichikawa, T. Meguro, Relationship between seed size of adzuki beans and Ann particles (bean jam particles), Bulletin of Hokkaido Prefectural Agricultural Experiment Stations. 66 (1994) 15–23.

GENOME-WIDE ASSOCIATION AND FINE-MAPPING OF THE *BCT* ALLELE FOR RESISTANCE TO BEET CURLY TOP VIRUS IN SNAP BEAN

Alvaro Soler[1],John P. Hart[2], Alyson Thornton[3], Deidrah Goldoff[3], Phillip D. Griffiths[4], Timothy G. Porch[2], James R. Myers[5], and Phillip N. Miklas*[1]

[1]Grain Legume Genetics and Physiology Research Unit, Agricultural Research Service, US Department of Agriculture, Prosser, WA, United States; [2]Tropical Agriculture Research Station, Agricultural Research Service, US Department of Agriculture, Mayagüez, Puerto Rico; [3]HM Clause, Sun Prairie, WI, United States; [4]School of Integrative Plant Science, Cornell University, Ithaca, NY; [5]Department of Horticulture, Oregon State University, Corvallis, OR

INTRODUCTION

Beet curly top virus (BCTV) (family *Geminiviridae*) is a curtovirus vectored by the beet leafhopper *Circulifer tenellus* (Baker) that causes serious crop damage in common bean. The most effective control of BCTV in bean is genetic resistance. The SCAR marker SAS08 (Larsen and Miklas, 2004) linked with *Bct* gene has been used in snap bean breeding programs for marker-assisted selection for resistance to BCTV, but new genomic tools provide the opportunity to fine map the locus. Our goal was to develop SNP markers from Genotyping by sequencing (GBS) data on the region associated with the Bct gene.

MATERIALS AND METHODS

Genotyping-by-sequencing of the 376 cultivars of the Snap bean Association Panel (SnAP) provided 23,304 SNPs to further elucidate the genetic basis of *Bct* by GWAS and fine mapping. *Beet curly top virus* reactions were obtained through agro-inoculation of the SnAP with a proprietary infectious clone in the greenhouse.

Population structure was determined using principal component analysis (PCA). An identity-by-state kinship matrix was created using the Efficient Mixed Model Association (EMMA) algorithm implemented in GAPIT R package. The mixed linear model (MLM) implemented in GAPIT with a P-value of 0.05 was used for GWAS analysis.

Missense variants were detected in candidate genes sequenced by Sanger sequencing. For significant SNP was designed two forward allele-specific primers with the 3′ base of each primer matching one of the SNP allele bases, and a reverse common primer, which was analyzed by melting curve.

RESULTS AND DISCUSSION

GWAS revealed a resistance allele (*Bct*) at a single locus on Pv07 **(Fig. 1)**, just 6 kb away from SAS08 SCAR marker. Eight putative candidate genes were identified in a 62.5 kb region. Sanger sequencing of gene Phvul.007G036300 (Exonuclease V - a 5' deoxyribonuclease), revealed a single SNP that was 99.5% predictive of the *Bct* resistance allele in the SnAP as compared to 94% predictive for the SAS08 SCAR marker. This SNP provides a more diagnostic, rapid and breeder-friendly molecular marker for MAS of *Bct* in snap beans **(Fig. 2)**.

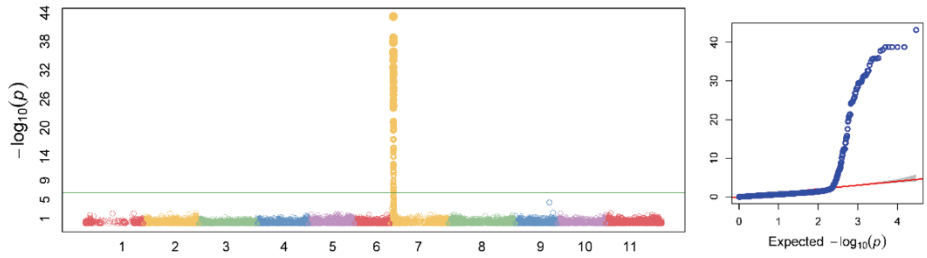


Figure 1. Genome-wide association detects markers on Pv07 associated with resistance to BCTV in Snap Bean Diversity panel. Chromosomal distribution of −log10(P) values for 23,304 SNP associations, and accompanying Q-Q plots for disease score.

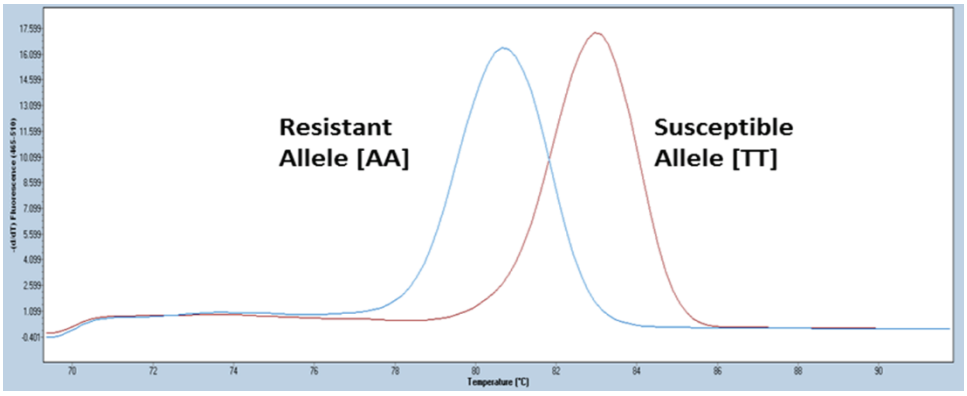


Figure 2. Melting curve analysis of 07_2970381 SNP evaluated by Tm shift assay.

Only 2 of 376 snap beans, Unidor and Golden Gate Wax, were resistant but lacked the corresponding Pv07 2,870,381 SNP allele. These two snap beans are being retested and investigation of Phvul.007G036300 as a candidate gene continues. Meanwhile, SNP 2,970,381 is recommended for MAS of *Bct* gene in snap bean.

REFERENCES

Larsen, R.C., Kurowski, C.J. & Miklas, P.N., 2010. Two Independent QTL are Responsible for Novel Resistance to Beet curly top virus in Common Bean Landrace G122. Phytopathology, 100(10), pp.972–978.

Larsen, R.C. & Miklas, P.N., 2004. Generation and Molecular mapping of a SCAR Marker Linked with the Bct Gene for Resistance to Beet curly top virus in Common Bean. Phytopathology, 94(4), pp.320–325.

Lipka, A.E. et al. (2012) GAPIT: Genome Association and Prediction Integrated Tool. Bioinformatics. doi: 10.1093/ bioinformatics/bts444.

GENERAL AND SPECIFIC COMBINING ABILITIES AS AN EFFICIENT APPROACH TO SELECT PARENTS AND SUPERIOR POPULATIONS FOR RESISTANCE TO WHITE MOLD IN COMMON BEAN

Lenio U. Ferreira[1], Patrícia G. S. Melo[1], Rogério F. Vieira[2], Murillo Lobo-Junior[3], Helton S. Pereira[3], Leonardo C. Melo[3], and Thiago Lívio P. O. Souza[3]*

[1]Universidade Federal de Goiás (UFG), Goiânia, GO 74.690-900, Brazil;
[2]Embrapa/Epamig, Vila Gianetti 47, Campus UFV, Viçosa, MG 36.571-000, Brazil;
[3]Embrapa Arroz e Feijão, Santo Antônio de Goiás, GO 75.375-000, Brazil;
*Corresponding author: thiago.souza@embrapa.br

INTRODUCTION

White mold (WM), caused by the soil borne fungus *Sclerotinia sclerotiorum* (Lib.) de Bary, is one of the most destructive diseases of common bean (*Phaseolus vulgaris* L.) in the world, mainly in the North and South American countries, including the United States, Canada, Argentina, and Brazil. This pathogen greatly undermines the seed yield and quality. In susceptible cultivars, WM can cause losses of 30 to 100%, especially in conducive weather of mean temperature around 10–25°C and high soil moisture, and in absence of disease management (Singh & Schwartz, 2010; Schwartz & Singh, 2013). The objective of this study was to select common bean parents and populations aiming to obtain elite lines resistant to WM by estimating general combining ability (GCA) and specific combining ability (SCA) for WM severity in three field nurseries carried out in Brazil.

MATERIAL AND METHODS

Partial diallel crosses were performed between parents from two groups: GI) three sources of partial resistance to WM identified abroad, and GII) nine Brazilian cultivars and elite lines. Twenty-seven populations were obtained and advanced in bulk up to F6 generation, when they were screened for WM severity in three field nurseries in Brazil (Oratórios-MG, Viçosa-MG, and Goianira-GO). The field nurseries were installed in a complete randomized block design. In Goianira, three replicates were used and each plot was two rows 2.0 m long, with 0.45 m between rows. In Oratórios, four replications were used and each plot was a single 3.0 m long row, with 0.50 m between rows. In Viçosa, five replications were used and each plot was a single 5.0 m long row, with 0.50 m between rows. BRS Requinte was used as a susceptible control and to fill the borders. The nurseries were sprinkler irrigated periodically. The WM severity was evaluated at the growth stage R8–R9, using a rating scale of 1 to 9 (1 = no disease symptoms and 9 = 80–100% diseased plants and/or 60–100% infected tissues) (Miklas et al., 2013). GCA and SCA estimates were obtained by the Griffing model IV, adapted by Geraldi & Miranda Filho (1988) for partial diallel. Statistics analyses were carried out using R software (R Care Team, 2013).

RESULTS AND DISCUSSION

Significant effects of populations (P) and of interaction between populations and environments (P × E) for WM severity were shown by the combined analysis of variance. The overall mean of WM severity in the combined analysis ranged from 2.83 to 5.03, in contrast with the mean severity of 7.21 presented by the susceptible control cultivar BRS Requinte. The results also showed the existence of variability for WM severity among parents from GII but not among the parents from GI, indicating that the selection should focus on cultivars and elite lines. In general, the GI parents contribute with favorable alleles for resistance to WM in the tested populations, but the genetic

effects depend of the environment. K-59, in Oratórios-MG and Viçosa-MG, and K-407, in Goianira-GO, contributed to increase the resistance to WM in the tested populations. Considering the GII parents, BRS Executivo and BRS Esplendor performed better across the three filed nurseries. The populations identified as most promising to be explored to obtain common bean lines resistant to WM were K-59/BRS Executivo, PI204717/BRS Campeiro, PI204717/Jalo Precoce, K-59/BRS Radiante and K-407/BRS Cometa, which presented significant and negative effects of the SCA. The results suggest that common bean breeding programs for WM resistance should use the strategy of selecting parents and populations resistant to WM based on estimates of GCA and SCA for WM severity in multi-field nurseries. Based on these evaluations, segregating populations that associate as many favorable alleles as possible to obtain resistant lines with broadly adaption to different environments may be achieved from crosses between selected parents.

ACKNOWLEDGMENTS

This work was supported by Brazilian Agricultural Research Corporation – Embrapa (Grant No. 02.14.01.003.00). We thank the Agricultural Research Institute of Minas Gerais (Epamig) for all support to carrying out the white mold nurseries in Minas Gerais. T.L.P.O. Souza, P.G.S. Melo, R.F. Vieira, H.S. Pereira and L.C. Melo are supported by Brazilian Council for Scientific and Technological Development (CNPq).

REFERENCES

Geraldi I., and Miranda Filho J.D. (1988) Adapted models for the analysis of combining ability of varieties in partial diallel crosses. Revista Brasileira de Genética 11: 419-430.

Miklas P.N., Porter L.D., Kelly J.D., and Myers J.R. (2013) Characterization of white mold disease avoidance in common bean. European Journal of Plant Pathology 135: 525-543.

R Core Team (2013) R: A language and environment for statistical computing. <http://www.R-project.org/>.

Schwartz H.F., and Singh S.P. (2013) Breeding common bean for resistance to white mold: a review. Crop Science 53: 1832-1844.

Singh, S.P. & Schwartz, H.F. (2010) Breeding common bean for resistance to diseases: a review. Crop Science 50: 2199-2223.

REDUCTION IN ROOT SECONDARY GROWTH AS A STRATEGY FOR PHOSPHORUS ACQUISITION

Christopher F. Strock, Laurie Morrow de la Riva, Jonathan P. Lynch

Dept. of Plant Science, The Pennsylvania State University, University Park, PA USA

Most soils on earth have suboptimal phosphorus (P) availability for plant growth (Lynch 2011; Lynch & Brown 2008; Vance et al. 2003), as it is only available to plants as inorganic P (P_i), and is rarely present in concentrations greater than several μM in soil solution (Bieleski 1973). Diffusion of P in soil is greatly outpaced by plant uptake, resulting in the formation of P depletion zones around roots (Hinsinger et al. 2005). Due to the limited availability and slow movement of P in soil, one of the most effective strategies of increasing P uptake is to increase the volume of soil explored by the root system. Although increasing resource allocation to root growth improves P acquisition, unbalanced root development reduces overall plant growth due to the increased metabolic cost of added root tissue (Lambers et al. 2006; Nielsen et al. 1998, 2001). To improve the balance between soil exploration and consumption of growth limiting resources, a decrease in root secondary growth would reduce the carbon cost of producing and maintaining root length (Lynch 1995). It has been hypothesized that this may be an adaptive strategy to improve the metabolic efficiency of soil foraging under P stress, where roots will favor primary growth (elongation) over secondary growth (radial thickening) to achieve greater exploration of soil domains that have not been depleted of P (De la Riva & Lynch 2010; Lynch & Brown 2008; Lynch 2007, 2011).

In this study, we utilize functional-structural modeling as well as empirical observations of plants grown in controlled environment mesocosms and in the field to explore the effect of reduced secondary growth of roots on P acquisition. Our goals were to test the hypotheses that 1) secondary growth is suppressed by P stress, 2) genetic variation exists for this response, and 3) reduced secondary growth of roots improves P acquisition. We address these hypotheses by first utilizing the functional-structural plant model *SimRoot* to determine the relationship between secondary growth of roots and P acquisition, followed by greenhouse and field studies to validate *in silico* results.

Functional-structural modeling in *SimRoot* indicates that in common bean (*Phaseolus vulgaris),* reduced root secondary growth reduces root metabolic costs, increases root length, improves phosphorus capture, and increases shoot biomass in low phosphorus soil. Observations from the field and greenhouse confirm that under phosphorus stress, resource allocation is shifted from secondary to primary root growth, genetic variation exists for this response, and reduced secondary growth improves phosphorus capture from low phosphorus soil. Under low phosphorus in greenhouse mesocosms, genotypes with reduced secondary growth had 39% smaller root cross sectional area, 60% less root respiration, 27% greater root length, 78% greater shoot phosphorus content, and 68% greater shoot mass than genotypes with advanced secondary growth. In the field under low phosphorus, these genotypes had 43% smaller root cross sectional area, 32% greater root length, 58% greater shoot phosphorus content, and 80% greater shoot mass than genotypes with advanced secondary growth. Secondary growth eliminated arbuscular mycorrhizal associations as cortical tissue was destroyed.

These results support the hypothesis that reduced root secondary growth increases resources available for primary growth, thereby increasing the total volume of soil explored and acquisition of soil resources. Although all *P. vulgaris* genotypes tested favor primary growth of roots over secondary growth under P stress, genotypes differ in the intensity of this response. Genotypes with reduced secondary growth had suppressed anatomical development, reduced metabolic and construction costs per length of root, greater soil exploration, and greater P acquisition than genotypes with advanced secondary growth.

Literature Cited

Bieleski RL (1973) Phosphate pools, phosphate transport, and phosphate availability. Annual Review of Plant Physiology and Plant Molecular Biology 24: 225-252

De la Riva LM, Lynch JP (2010) Root etiolation as a strategy for phosphorus acquisition in common bean (Masters dissertation). The Pennsylvania State University Library (Call Number: Thesis 2010mDeLaR,LM)

Hinsinger P, Gobran GR, Gregory PJ, Wenzel WW (2005) Rhizosphere geometry and heterogeneity arising from root mediated physical and chemical processes. New Phytologist 168: 293–303

Lambers H, Shane MW, Cramer MD, Pearse SJ, Veneklaas EJ (2006) Root structure and functioning for efficient acquisition of phosphorus: matching morphological and physiological traits. Annals of Botany 98: 693–713

Lynch J (1995) Root architecture and plant productivity. Plant Physiology 109: 7-13

Lynch JP (2007) Roots of the second green revolution. Australian Journal of Botany 55: 493-512

Lynch JP (2011) Root phenes for enhanced soil exploration and phosphorus acquisition: tools for future crops. Plant Physiology 156: 1041-1049

Lynch JP, Brown KM (2008) Root strategies for phosphorus acquisition. In: White PJ, Hammond JP, eds. The ecophysiology of plant-phosphorus interactions, Dordrecht:Springer, 83-116

Nielsen KL, Bouma TJ, Lynch JP, Eissenstat DM (1998) Effects of phosphorus availability and vesicular-arbuscular mycorrhizas on the carbon budget of common bean (*Phaseolus vulgaris*). New Phytologist 139: 647-656

Nielsen KL, Eshel A, Lynch JP (2001) The effect of phosphorus availability on the carbon economy of contrasting common bean (Phaseolus vulgaris L.) genotypes. Journal of Experimental Botany 52: 329-339

Vance CP, Uhde-Stone C, Allan DL (2003) Phosphorus acquisition and use: critical adaptations by plants for securing a nonrenewable resource. New Phytologist 157: 423-447

EFFECT OF DROUGHT ON BEAN COOKING TIME AND WATER ABSORPTION USING GERMPLASM SELECTED FOR ROOT ROT RESISTANCE IN MOZAMBIQUE AND ZAMBIA

Carlos A. Urrea[1*], James Steadman[1], Eduardo Valentin Cruzado[1], and Graciela Godoy-Lutz

[1]University of Nebraska-Lincoln, NE

INTRODUCTION

Root rot and crown rot of dry beans have becoming a limiting factor in Mozambique and Zambia. *Fusarium* spp. were identified as the major pathogens associated with root rot crown disease in Zambia and Mozambique. In addition, bean cooking time is a major concern in Africa because longer cooking time requires use of more energy resources. Therefore, we explored the effect of drought on bean cooking time and water absorption using germplasm selected for root rot resistance in Mozambique and Zambia.

MATERIALS AND METHODS

Based on common dry bean trials planted in Zambia and Mozambique in 2015, we assembled a common root rot trial of 22 entries to be planted in both countries. This trial was also planted at Mitchell, NE in 2016 to explore the effect of drought on cooking time for the above entries. This was accomplished by comparing the cooking time of beans grown under drought and non-drought conditions. The lines were grown in replicated trials in adjacent irrigated (non-stressed, NS) and non-irrigated (drought-stressed, DS) plots. Both NS and DS blocks were irrigated until flowering to ensure good plant establishment and normal vegetative growth. Thereafter, the stressed block was not irrigated.

After beans were harvested and stored for four to five months, a Matson Bean cooker was used to evaluate the effect of drought on cooking time. Seed from each plot was processed separately using the following procedures. A 60-seed sample was soaked in distilled water overnight (16 h). Initial seed weight was recorded. Distilled water was added to the cooker and heated to 98°C, then 24 of the pre-soaked seeds were placed in the template in the cooker to align the seeds with the plungers. Final seed weight was recorded and water absorption was calculated. An observer recorded the time when the beans were placed in the cooker and when 80% were cooked (indicated by the plungers dropping).

RESULTS AND DISCUSSION

In general, dry beans grown under DS took 7 minutes longer to cook than NS (Table 1). Under both DS and NS, seed water absorption was negatively correlated with cooking time (-0.91** and -0.88**, respectively). Some sources of root rot resistance grown under DS took longer to cook and had less water absorption than when grown under NS conditions (Table 1).

In terms of yield, G10994 and INIAP 414 had the highest and lowest Geometric Mean, respectively (Table 1). G10994, Larga Comercial, USDK-4, NE34-12-50, W16560, INIAP 414, and PI321094-D had the lowest root rot incidence (Table 1). G10994, Larga Comercial, W16560, INIAP 414, PI321094-D, NE34-12-28, and NE34-12-45 had the longest cooking time under both DS and NS environments, whereas NE34-12-47 and NE34-12-50 had the lowest (Table 1).

Thus, many of the entries showing good resistance to root rot had longer cooking times than entries with poorer resistance. In contrast, both NE34-12-50 and NE34-12-47 showed good resistance to both root rot (0.0 and 1.7%, respectively) and CBB (1.0 and 0.0%, respectively), had fairly high yields (grand mean = 1778 and 1890 kg/ha, respectively), and were among the lowest entries for cooking time (Table 1). Although USDK-4 and Carioca, Kihala also showed good resistance to root rot (0.0 and 1.7%, respectively) and had fairly low cooking times (Table 1), their yield was lower (grand mean = 1530 and 1539 kg/ha, respectively) and their incidence of CBB was higher (3.7 and 5.0%, respectively).

Table 1. Effect of terminal drought stress on cooking time and water absorption using germplasm selected for root rot resistance in Mozambique and Zambia.

					Drought Stress		Normal	
ID	**Geometric Mean**	**Yield Reduction**	**CBB**	**Root Rot**	**Cooking Time**	**Water Absorption**	**Cooking Time**	**Water Absorption**
	kg/ha	**%**	**(1-9)**	**%**	**min**	**%**	**min**	**%**
G 10994	2888	52.7	3.0	0.0	160	23	136	24
Larga Comercial	1294	62.7	2.3	0.0	139	32	119	26
USDK-4	1530	43.0	3.7	0.0	50	111	48	107
NE34-12-50	1778	43.2	1.0	0.0	49	81	39	76
Local 2 (W16560)	1668	68.1	1.0	0.0	165	22	121	15
INIAP 414	299	27.6	5.0	0.0	103	30	98	20
PI321094-D	788	95.7	1.0	0.0	180	17	184	14
Carioca, Kihala	1539	48.5	5.0	1.7	60	71	44	83
NE34-12-47	1890	41.2	0.0	1.7	40	98	40	87
NE34-12-28	1961	72.5	5.3	5.0	116	30	106	37
NE34-12-45	1536	63.3	6.0	5.0	142	38	150	34
Local 1 (OPS-RS1)	1336	54.6	4.0	18.3	75	51	57	54
Uyole 96	1055	43.7	4.7	20.0	56	90	52	84
GRAND MEAN	1167	55.6	4.5	28.3	87	65	80	60
LSD (0.05)			2.3	20.7	46	14	25	18

CHARACTERIZATION OF NESTED ASSOCIATION MAPPING POPULATION IN DRY BEAN

Maryam Vazin, Tom Smith and K. Peter Pauls

Department of Plant Agriculture, Univ. of Guelph, Guelph, Ontario, Canada N1G 2W1

INTRODUCTION: Understanding the genetic bases of different traits agronomic and quality traits are important for efforts to breed improved lines in common bean. The aim of plant breeding is to continually improve advantageous traits in order to make the crops more efficient. The use of Nested association mapping (NAM) populations combines the advantages of linkage mapping and association mapping and is the choice for mapping the effects of rare alleles in germplasm collections (Yu *et al*, 2008). The objective of this study is to phenotype the RILs in the NAM population in field studies for traits agronomic such as yield.

MATERIAL AND METHODS: A Nested Association Mapping (NAM) population (Yu *et al*, 2008) of 600 $F_{4:5}$ recombinant inbred lines (RIL) was developed from 10 bi-parental crosses with ExRico23, including: ExRico23xApex; ExRico23xCompass; ExRico23xCruiser; ExRico23xEnvoy; ExRico23xGryphon; ExRico23xLaser; ExRico23xMist; ExRico23xRex; ExRico23xRexeter; and, ExRico23xT9905. The crosses were made by Tom Smith at the University of Guelph. The F_1s were self-pollinated to produce recombinant inbred lines. Four to 10 F_1 seeds from each cross were grown at Elora in 2014 to produce F_2s. Approximately 140 F_2 seeds from each family were grown in the field in Puerto Rico in winter in 2014/15 and allowed to self. One hundred and forty individual F_3 seeds from each F_2 family were grown in Puerto Rico in 2014/15. The individual F_4 lines were grown at Elora in 2015 and 75 seeds per line were bulked. The $F_{4:5}$ was increased in the field in Puerto Rico during the winter 2016.

600 RILs from the NAM population (approximately 60 /cross) were evaluated for two years (2016, 2017) in two field sites [Guelph Elora Research Station (ERS) and Woodstock Research Station (WRS)] in Ontario, Canada. The field trials were planted as a 6 x (10 x 10) unbalanced square lattice design (Cochran and Cox, 1957) designed with Agrobase®99 software. The experimental units for all field locations were 36 cm x 1.9 m plots with 135 seeds planted at 2.5 cm row spacing.

RESULTS AND CONCLUSION:
The yield data obtained from the RILs in the NAM population and their parents tested at the Elora Research Station (ERS) and Woodstock Research Station (WRS) in 2016 is shown in **Figure 1**. The distribution of the yield trait was continuous and showed some transgressive segregation, compared to the parental lines. The NAM population will be genotyped using Genotyping by Sequencing (GBS). The study of the phenotypic and genotypic characteristics of the NAM population from Ex Rico 23 and 10 founder lines will improve our understanding of the genetic diversity of Ontario Mesoamerican germplasm.

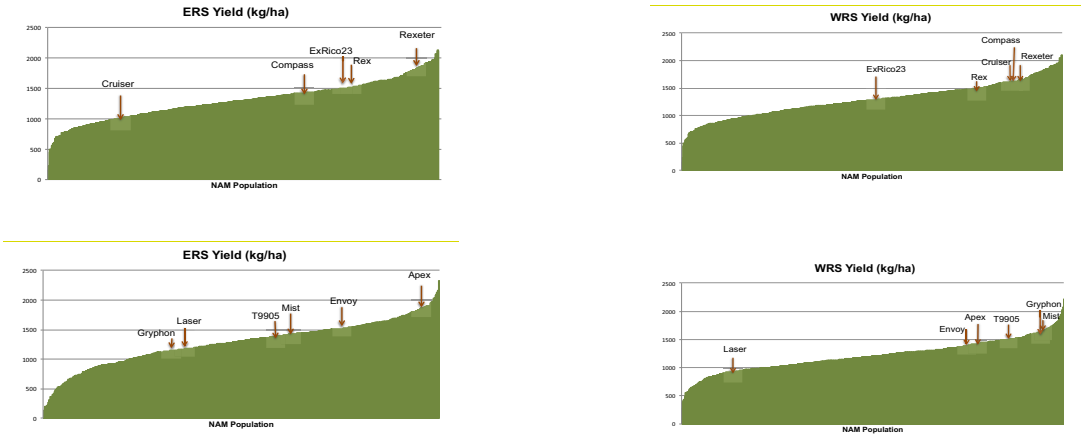


Figure 1. Yield distributions in the two trials for the Nested Association Mapping (NAM) population in 2016 at the Elora Research Station (ERS) on the left side and Woodstock Research Station (WRS) on the right side. Scores of the parental lines are indicated on each graph by arrows.

REFERENCES:

Cochran W G, Cox G M .1957. Random Permutations. In W. G. Cochran, and G. M. Cox. Experimental Designs.

Yu J, Hollan J B, McMullen M D, Buckler E S. 2008. Genetic design and stistical power of nested association mapping in maize.

RACES OF *Colletotrichum lindemuthianum* IN COMMON BEAN FROM PARANÁ STATE, SOUTHERN OF BRAZIL

Xavier LFS[1], Poletine JP[1], Vidigal Filho PS[1], Martiniano-Souza MC[2], Calvi AC[1], Castro SAL[1], Silva JB[1], Dartibale GB[1], and Gonçalves-Vidigal MC[1]*

[1]Departamento de Agronomia, Universidade Estadual de Maringá, PR, Brazil; [2]Instituto Agronômico de Pernambuco, PE, Brazil. *E-mail: mcgvidigal@uem.br

INTRODUCTION

Colletotrichum lindemuthianum possesses wide pathogenic (Balardin et al. 1999) and molecular variability (Coelho et al. 2016), and it is the causal agent of the anthracnose, considered one of the most important common bean fungi disease around the world (Kelly and Vallejo 2004). It can cause total yield loss in susceptible common bean cultivars when submitted to favorable environments (Pastor-Corrales et al. 1995). Studies revealed that more than 250 physiological races have been identified in the world (Nunes et al. 2013), and Paraná state, outstands out as the largest of common bean producer in Brazil, presenting the largest number of physiological identified races of this pathogen (Gonçalves-Vidigal et al. 2008). This study had as objective to characterize isolates of *C. lindemuthianum* from different regions of Paraná State using the 12 Differential Cultivars for *C. lindemuthianum* races and evaluate their genetic diversity through sequencing of the internal transcribed spacer (ITS) regions of ribosomal DNA (rDNA).

MATERIAL AND METHODS

This research was conducted at the Núcleo de Pesquisa Aplicada a Agricultura (Nupagri), Universidade Estadual de Maringá, Paraná state, Brazil. Pods and leaves with anthracnose symptoms were collected from infected common bean plants in the different regions of Paraná. A total of 19 isolates of *C. lindemuthianum* was obtained through monosporic isolation process and evaluated according to the virulence in the set of 12 Differential Common Bean Cultivars (Pastor-Corrales et al. 1995). Genomic DNA was extracted from 12 isolates (Cárdenas et al. 2012) and PCR amplification of ITS1 and ITS2 regions were performed. Subsequent sequencing of these regions and bioinformatics analysis were carried out in order to identify SNPs.

RESULTS AND DISCUSSION

The characterization of *C. lindemuthianum* isolates from Paraná state using the common bean differential cultivars set, allowed the identification of 13 races (Table 1). The races 1, 9, 72 and 73 showed compatibility only with Mesoamerican cultivars, while races 17, 24, 25, 27, 81, 89, 95, 339 and 345 presented compatibility with Mesoamerican and Andean cultivars. This is the first report of the race 24 in the Paraná state and the race 345 in the world, demonstrating the wide pathogenic variability of *C. lindemuthianum* isolates and its potential of evolution. Analysis of the sequences in the ITS1 and ITS2 regions of 12 isolates revealed the presence of 72 SNPs and 55 SNPs, respectively, evidencing high molecular variability among the *C. lindemuthianum* isolates (Figure 1). Similar results were described by Balardin et al. (1999) and Coelho et al. (2016) who detected more variability in the ITS2 region when compared with the ITS1 region in the sequences from the isolates of *C. lindemuthianum*. The amount of SNPs observed demonstrated the wide genetic diversity among *C. lindemuthianum* isolates. Thus, the monitoring of *C. lindemuthianum* races by virulence and molecular analyses in the main areas of common bean production is very important to breeding programs aiming to obtain resistant cultivars against the broader variability of this pathogen.

Table 1. Races of *C. lindemuthianum* characterized using the 12 Differential Common Bean Cultivars

ISOLATES	DIFFERENTIAL CULTIVARS												RACES
	A	B	C	D	E	F	G	H	I	J	K	L	
	1	2	4	8	16	32	64	128	256	512	1024	2048	
1	+	+	-	+	+	-	-	-	-	-	-	-	27
2	+	+	+	+	+	-	+	-	-	-	-	-	95
3	+	-	-	+	-	-	+	-	-	-	-	-	73
4	-	-	-	+	-	-	+	-	-	-	-	-	72
5	+	-	-	+	+	-	+	-	-	-	-	-	89
6	+	-	-	+	+	-	-	-	-	-	-	-	25
7	+	-	-	+	+	-	+	-	+	-	-	-	345
8	+	-	-	+	+	-	-	-	-	-	-	-	25
9	+	-	-	-	-	-	-	-	-	-	-	-	1
10	+	-	-	+	+	-	-	-	-	-	-	-	25
11	+	-	-	+	-	-	-	-	-	-	-	-	9
12	+	-	-	-	+	-	+	-	-	-	-	-	81
13	+	-	-	-	+	-	-	-	-	-	-	-	17
14	+	-	-	-	+	-	-	-	-	-	-	-	17
15	-	-	-	+	+	-	-	-	-	-	-	-	24
16	+	-	-	-	+	-	-	-	-	-	-	-	17
17	+	+	-	-	+	-	+	-	+	-	-	-	339
18	+	+	-	+	+	-	-	-	-	-	-	-	27
19	+	-	-	+	+	-	+	-	-	-	-	-	89

A: Michelite; B: Michigan Dark Red Kidney; C: Perry Marrow; D: Cornell 49-242; E: Widusa; F: Kaboon; G: Mexico 222; H: PI207262; I: TO; J: TU; K: AB136 and L: G2333; +: Susceptible; -: Resistant.

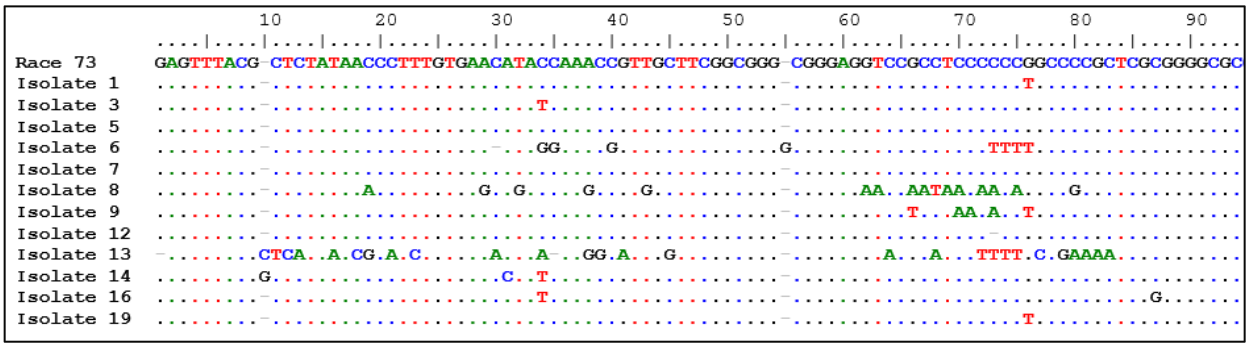


Figure 1. Presence of SNPs in the initial part of the ITS1 region sequencing for 12 isolates of *C. lindemuthianum*.

ACKNOWLEDGEMENTS: This research was financially supported by CNPq and Capes

REFERENCES

Balardin et al. (1999). Mycological Research, 103: 841-848
Cárdenas et al. (2012). Plant Disease, 96: 456
Coelho et al. (2016). Acta Scientiarum - Agronomy, 38: 429-438
Gonçalves-Vidigal et al. (2008). Brazil Archives of Biology and Technology, 51: 883-888
Kelly JD and Vallejo VA (2004). HortScience, 39: 1196-207
Nunes et al. (2013). Biennial Meeting of the Bean Improvement Cooperative
Pastor-Corrales et al. (1995). Plant Disease, 79:63-67.

RELATIONSHIP OF ROOT ARCHITECTURE WITH TOTAL PLANT BIOMASS AND SEED YIELD IN COMMON BEAN (Phaseolus vulgaris L.) UNDER GLASS HOUSE CONDITIONS

Waines JG[1], Chiu T[1], Herrera V[1], Rangel B[1] and Ibarra-Perez FJ[2]

[1]University of California Riverside, Botany and Plant Sciences Department (waines@ucr.edu)
[2]INIFAP, Campo Experimental Cotaxtla, Mexico.

INTRODUCTION. The majority of research studies in the last 150 years have been on above-ground organs, such as stems, leaves, flowers, fruits and seeds. Research on below-ground organs, such as roots has been neglected. It is time to understand the relationship of root characters to shoot traits and seed yield. Similar relationships of root traits to grain yield were undertaken with wheat (Ehdaie, Whitkus and Waines, 2003), and maize (Lynch, 1995). Only now, genes were discovered in common bean that control quantitative trait loci (QTL) for root characters and root diseases (Nakedde et al., 2016). The objective of this study was characterize root architecture of ten bean cultivars including landraces, old and modern cultivars adapted to seed production in USA and Mexico. The hypotheses were: 1) Seed yield is correlated with above-ground biomass, 2) seed yield is correlated with total root biomass, 3) seed yield is correlated with shallow or deep root biomass, 4) seed yield is correlated with total plant biomass.

MATERIALS AND METHODS. Ten black bean lines included in the study represented historical releases dating over more than 70 years, which allowed determination if root traits had a consistent trend over those years (Table 1). The experiment was set up and managed as described in Nakedde et al. (2016) with a modification, 50:50 by weight sand:turface was used instead of only silica sand. The experiment ran from early April to early June 2017.

Table 1. Historical releases in Mexico and USA of indeterminate, early to full maturity season with small-medium seed size black bean cultivars.

Cultivar	Puebla 152	Negro Jamapa	Huasteco 81	Negro Veracruz	Negro INIFAP	Negro Tacana	Negro Papaloapan	Negro Comapa	Verdin	Zenith
Released	1950	1958	1981	1986	1994	1993	2006	2010	2014	2014
G. habit*	IIIb	IIIa	IIb	IIa	IIb	IIa	IIb	IIb	IIa	IIa
Maturity**	Full	Mid	Mid	Mid	Mid	Mid	Mid	Mid	Early	Full

*IIa = upright short vine, IIb = upright long vine, IIIa = prostrate short vine, IIIb = prostrate long vine. **Early-season = 70 d, Mid-season = 90 d and Full-season = 100 – 105 d after planting.

RESULTS AND DISCUSSION. No significant differences among cultivars were found for all measured root characteristics and the above-ground biomass; in contrast, cultivar differences were significant for total plant biomass, seed yield and yield components (Table 2). The mean values for shallow root, deep root, total root and above-ground biomass were 4.6, 2.3, 6.65 and 11.83 g pl^{-1}, respectively. The longest root averaged 89.7 cm among cultivars. The root to shoot ratio fluctuated from 0.51 (Puebla 152) to 0.67 (Negro INIFAP). Seed yield averaged 11.8 g pl^{-1} and Negro Jamapa had the highest mean (17.9 g pl^{-1}) and was different from the rest of the cultivars. Seeds pod^{-1} and seeds $plant^{-1}$ were yield components that showed significant differences among cultivars, Negro Jamapa produced 4.4 seeds pod^{-1} the same as Zenith (4.3) and Negro Veracruz (4.2). Negro Jamapa produced almost four-fold more seeds $plant^{-1}$ than Puebla 152 and 37% more than Verdín. Total plant biomass ranged from 24.2 to 39.9 g pl^{-1} and a group of four cultivars had

the highest mean values: Negro Jamapa (39.9), Negro Papaloapan (33.3), Negro INIFAP (32.8) and Negro Comapa (32.2), which were similar but different from the rest of the cultivars (Table 2). Seed yield (r=0.84*) and total plant biomass (r=0.83*) were positively correlated only with deep root biomass. Shallow root biomass was positively correlated (r=0.86*) with above-ground biomass.

Table 2. Root, shoot, total biomass and seed yield (g plant^{-1}) and yield components of tropical black common bean cultivars grown in tubes under glass house conditions 2017.

Cultivar	SRB	DRB	TRB	RL (cm)	AB	R/S	NSP	NSPL	Seed yield	TPB
Puebla 152	4.93	1.30	6.23	90.00	12.25	0.51	3.85	19.00	5.77	24.25
Jamapa	4.65	3.75	8.40	91.50	13.63	0.61	4.41*	74.75*	17.86*	39.88*
Huasteco 81	4.00	1.90	5.90	79.00	11.13	0.61	3.33	45.00	10.81	27.84
Veracruz	4.30	1.88	6.18	96.25	12.13	0.51	4.17*	58.50	12.34	30.64
INIFAP	4.58	2.38	6.95	97.25	12.03	0.67	2.78	48.25	13.19	32.17*
Tacaná	3.45	2.48	5.93	85.00	10.93	0.54	1.53	57.50	12.62	29.47
Papaloapan	5.43	2.80	8.23	92.50	14.35	0.57	3.57	39.50	10.74	33.32*
Comapa	4.70	2.55	7.25	84.25	12.95	0.61	3.47	51.25	11.98	32.18*
Verdín	4.23	2.10	6.33	100.50	10.07	0.63	3.89	47.00	12.22	28.63
Zenith	3.33	1.83	5.15	80.75	8.90	0.60	4.33*	53.00	10.95	25.00
Mean	4.36	2.30	6.65	89.70	11.83	0.59	3.53	49.38	11.85	30.34
C.V. (%)	28.7	48.0	30.2	12.9	27.4	37.3	25.69	19.7	23.0	21.0
ANOVA	ns	ns	ns	ns	ns	ns	**	**	**	*
SD (0.05)							1.32	14.12	3.95	9.19

SRB= shallow root biomass, DRB= deep root biomass, TRB= total root biomass, RL= root length, AB= above-ground biomass, R/S= root to shoot ratio, NSP= seeds/pod, NSPL= seeds/plant, SY= seed yield, TPB= total plant biomass. *,**= significant at 0.05 and 0.01 of probability, ns= non-significant.

CONCLUSIONS. It was difficult to wash out the sand:turface from adventitious roots, which may account for the lack of significance in the root and shoot data among the different bean cultivars. Significant differences were found for seed yield and yield components. Jamapa, type IIIa, released in 1958 was superior for most seed yield components. The landrace Puebla 152 was not superior to modern cultivars for root traits. Zenith, released in 2014, had the smallest total root biomass and root length, and low seed yield, along with Papaloapan and Huasteco 81. Seed yield and total plant biomass were positively correlated with deep root biomass. The experiment should be repeated.

REFERENCES

Lynch, J. 1995. Root architecture and plant productivity. Plant Physiology 109:7-13.

Ehdaie, B., Whitkus RW, and J.G. Waines. 2003. Root biomass, water-use efficiency and performance of wheat-rye translocations of chromosomes 1 and 2 in spring bread wheat Pavon F76. Crop Sci. 43:710-717.

Nakedde T, Ibarra-Perez FJ, Makankusi C, Waines JG and Kelly JD. 2016. Mapping QTL associated with root architecture and root rot resistance in black beans. Euphytica 212:51-63.

PRE-GERMINATION FLOODING TOLERANCE OF MIDDLE AMERICAN DRY BEAN GENOTYPES

Katelynn Walter[1]*, Ali Soltani[2], Atena Oladzad Abbasabadi[1], Phillip E. McClean[1], Dipayan Sarkar[1], and Juan M. Osorno[1]

[1] Department of Plant Sciences, North Dakota State University, Fargo, ND [2]Department of Plant Biology, Michigan State University, East Lansing, MI

INTRODUCTION

Dry bean (*Phaseolus vulgaris* L.) is a very sensitive crop to flooding stress and is the most sensitive among all crops grown in North Dakota, the leading producer of dry bean in the nation. Excess water was the leading production issue for dry bean growers in North Dakota and Minnesota from 2013-2015 (Knodel et al., 2016). Germination is particularly sensitive to flooding stress and dry bean genotypes with a pigmented seed coat (black, small red, pink market classes) are more tolerant than those with an unpigmented seed coat (navy and great northern market classes) (Soltani et al., 2017). The objectives of this study were to screen unpigmented genotypes from a Middle-American Diversity Panel (MDP) for pre-germination flooding tolerance and to identify genomic regions associated with this tolerance using a genome wide association study (GWAS).

MATERIALS AND METHODS

Unpigmented seed coat genotypes (navy and great northern market classes; n=87) from MDP were screened for pre-germination flooding tolerance in greenhouse conditions. Genotypes were arranged in a randomized complete block design (RCBD) with a split-plot arrangement, with treatments (non-flooded and flooded) as the main plots and genotypes as the subplots. Ten seeds per genotype were planted into 24-well trays with autoclaved play sand and three replications were performed. Based on preliminary experiments, flooded main plots were subjected to 3h of complete submergence and non-flooded main plots were watered to field capacity. Flooded main plots were drained to field capacity and maintained for 14d. Germination rate (GR, %), shoot weight (SW, g), root weight (RW, g), total weight (TW, g), and plant height (PH, cm), were measured 14d after planting. GWAS was performed using the GAPIT package in R (Lipka et al., 2012). Around 200 K SNP markers were used in for this analysis (Soltani et al., 2017). Multiple models (Naïve, EMMA, PC, and EMMA+PC) were tested for each trait analyzed and the best model was selected based on the lowest mean square deviation (MSD).

RESULTS AND DISCUSSION

Mean GR of non-flooded treatment was significantly higher (89%) than that of flooded treatment (14%) and all genotypes in the flooded treatment had a GR less than 50% except for ‘Royalty’ (93%) and ‘Verano’ (63%) (Figure 1). Genotype was highly significant for all traits analyzed and genotype-by-treatment interactions were significant for all traits except for PH which suggests GR and dry weights can help identify pre-germination flooding tolerant genotypes. A significant genomic region on Pv06 was identified which is ~2.7 kb upstream of the *Phvul.006G148300* gene (Figure 2). This gene encodes for a PMP22 protein, which is suggested to be involved in enzymatic antioxidant defense systems (Murphy et al., 2003).

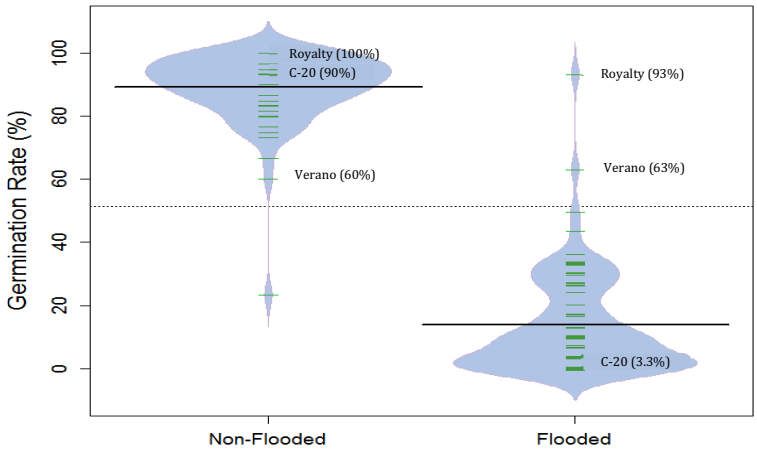


Figure 1. Distribution of germination rates for non-flooded and flooded treatments. Green lines depict individual observations and the blue area shows the distribution.

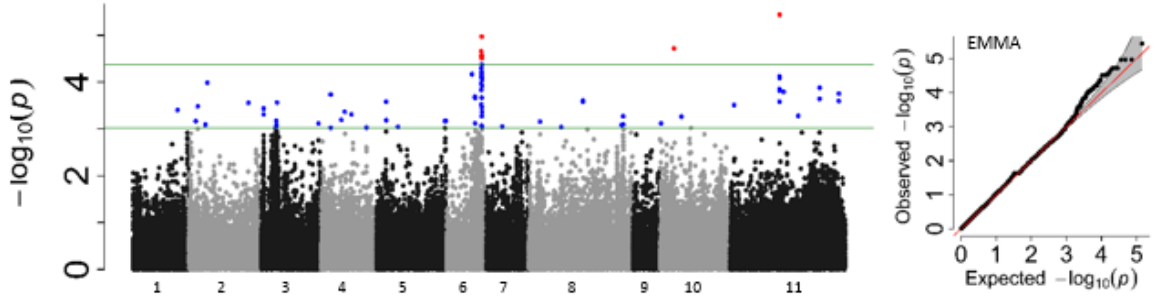


Figure 2. Manhattan and Q-Q plots for germination rate in both flooded and non-flooded treatments for unpigmented seed genotypes.

CONCLUSIONS

Pre-germination flooding tolerant unpigmented genotypes were identified in this experiment. Verano was found to be the most tolerant unpigmented seed coat genotype performing significantly better than other genotypes analyzed. *Phvul.006G148300* is a potential candidate gene for pre-germination flooding tolerance in unpigmented seed coat genotypes and indicates that tolerant navy and great northern genotypes might have increase reactive oxygen species scavenging systems in comparison to their sensitive counterparts.

REFERENCES

Soltani, A., MafiMoghaddam, S., Walter, K., Restrepo-Montoya, D., Mamidi, S., Schroder, S., Lee, R., McClean, P.E. and Osorno, J.M., 2017. Genetic Architecture of Flooding Tolerance in the Dry Bean Middle-American Diversity Panel. *Frontiers in plant science*. 8:1-15.

Knodel, J.J., P.B. Beauzay, G.J. Endres, D.W. Franzen, H.J. Kandel, S.G. Markell, J.M. Osorno, J.S. Pasche, and R.K. Zollinger. 2016. 2016 Dry Bean Grower Survey of Production, Pest Problems and Pesticide Use in Minnesota and North Dakota (E1841).

Murphy, M.A., Phillipson, B.A., Baker, A., Mullen, R.T. 2003. Characterization of the Targeting Signal of the Arabidopsis 22-kD Integral Peroxisomal Membrane Protein. *Plant Physiology*. 133:813-828.

CHARACTERIZATION AND DISTRIBUTION OF A NEW EMERGING RACE OF ANTHRACNOSE IN MICHIGAN

Halima E. Awale, Nolan Bornowski, Evan M. Wright, Greg V. Varner and James D. Kelly

Michigan State University, East Lansing, MI

Anthracnose, caused by *Colletotrichum lindemuthianum*, a specialized hemibiotrophic fungus, is a seed borne disease of common beans (*Phaseolus vulgaris*). The fungus is found in all bean growing areas of the world, but predominantly in subtropical and temperate regions where cool and humid conditions prevail (Padder et al., 2017). The pathogen causes significant yield loss when field conditions are conducive for infection. Due to the high degree of pathogenic variability of the fungus and continual emergence of new races, genetic resistance in the host is not durable (Kelly et al., 1994). In recent years, isolates collected from Michigan bean fields were consistently characterized as Races 7 and 73. Resistant varieties have been developed by deploying the Co-1^2 gene, which confers resistance to all known races present in Michigan. In the 2017 growing season, a severe infection of anthracnose was observed in Zenith black bean fields in Alcona County in Northern Michigan. A survey was conducted to collect samples across the bean growing counties of Michigan. The objectives were: a) survey and sample infected fields throughout Michigan, b) characterize, determine virulence, and geographic distribution of the race infecting Zenith, and c) develop a breeding strategy for pyramiding additional resistance genes into future cultivars.

MATERIALS AND METHODS: In 2017, a disease survey was conducted across nine counties of the Michigan bean growing region. A total of thirty-nine infected pod samples were collected. Each sample was designated as ANT17-xx (Table 1). Each sample was surface sterilized using 1% bleach, followed by 70% ethanol and rinsed with sterile water. The samples were then cultured in both Potato dextrose agar (PDA) and Mathur agar to isolate the *C. lindemuthianum* fungus and placed in an incubator at 24 degrees C. Fungal growth was monitored for sporulation on both media and isolates were sub-cultured onto either Potato Dextrose Agar (PDA) or Mathur agar (MA) plates, whichever produced most vigorous fungal growth. These subcultures were used for subsequent inoculations. Four samples were discarded due to absence of fungal growth and upon closer inspection, pod lesions were likely caused by *Xanthomonas axonopodis*.

PLANTING AND INOCULATION: Six seeds each of the twelve-cultivar anthracnose differential series were planted in the MSU greenhouse for race identification (Kelly et al., 1994). Two weeks after planting, seedlings were inoculated with a spore suspension of $1.2x10^6$ spores/mL and placed in a mist chamber where high humidity was maintained for three days. The plants were evaluated for disease reactions after seven days and scored as resistant (R) or susceptible (S).

RESULTS AND CONCLUSIONS: A total of thirty-five anthracnose samples collected from nine bean growing counties in Michigan were characterized by their reactions on twelve differential cultivars of *Phaseolus vulgaris*. Twenty-seven isolates from the Michigan dry bean growing region were identified as Race 73 that commonly occurs when conditions are conducive for disease development. An isolate from Montcalm County was identified as Race 7, which overcomes the *Co-1* gene. Six isolates from Alcona County and one isolate from Alpena County were characterized as Race 109, previously reported in Manitoba, but not previously reported in Michigan. Race 109 is virulent on the *Co-1^2* gene possessed by Zenith, which previously conferred resistance to all known races found in Michigan (Zuiderveen et al., 2016). Due to the emergence of Race 109, a new breeding strategy will be initiated to pyramid additional resistance genes such as *Co-4^2, Co-5* and *Co-6* genes into future dry bean cultivars.

Table 1. Isolate number, location collected, reaction of common bean differential cultivars, and race designation of 35 races of *Colletotrichum lindemuthianum* from Michigan. Differential cultivars designated as follows: A: Michelite, B: Michigan Dark Red Kidney, C: Perry Marrow, D: Cornell 49242, E: Widusa, F: Kaboon, G: Mexico 222, H: PI 207262, I: TO, J: TU, K: AB 136, L: G2333.

Isolate number	County	A	B	C	D	E	F	G	H	I	J	K	L	Race identified
ANT 17-1	Alcona	S	R	S	S	R	S	S	R	R	R	R	R	109
17-2	Alcona	S	R	S	S	R	S	S	R	R	R	R	R	109
17-3	Alcona	S	R	S	S	R	S	S	R	R	R	R	R	109
17-4	Alcona	S	R	S	S	R	S	S	R	R	R	R	R	109
17-5	Alcona	S	R	S	S	R	S	S	R	R	R	R	R	109
17-6	Alcona	S	R	R	S	R	R	S	R	R	R	R	R	73
17-7	Alcona	S	R	R	S	R	R	S	R	R	R	R	R	73
17-8	Alcona	S	R	R	S	R	R	S	R	R	R	R	R	73
17-9	Alcona	S	R	R	S	R	R	S	R	R	R	R	R	73
17-10	Alcona	S	R	R	S	R	R	S	R	R	R	R	R	73
17-11	Alcona	S	R	S	S	R	S	S	R	R	R	R	R	109
17-12	Alcona	S	R	R	S	R	R	S	R	R	R	R	R	73
17-13	Alcona	S	R	R	S	R	R	S	R	R	R	R	R	73
17-14	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-15	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-16	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-17	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-18	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-19	Alpena	S	R	S	S	R	S	S	R	R	R	R	R	109
17-20	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-24	Alpena	S	R	R	S	R	R	S	R	R	R	R	R	73
17-25	Gratiot	S	R	R	S	R	R	S	R	R	R	R	R	73
17-26	Tuscola	S	R	R	S	R	R	S	R	R	R	R	R	73
17-27	Huron	S	R	R	S	R	R	S	R	R	R	R	R	73
17-29	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-30	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-31	Arenac	S	R	R	S	R	R	S	R	R	R	R	R	73
17-32	Arenac	S	R	R	S	R	R	S	R	R	R	R	R	73
17-33	Arenac	S	R	R	S	R	R	S	R	R	R	R	R	73
17-34	Huron	S	R	R	S	R	R	S	R	R	R	R	R	73
17-35	Huron	S	R	R	S	R	R	S	R	R	R	R	R	73
17-36	Bay	S	R	R	S	R	R	S	R	R	R	R	R	73
17-37	Montcalm	S	S	S	R	R	R	R	R	R	R	R	R	7
17-38	Sanilac	S	R	R	S	R	R	S	R	R	R	R	R	73
17-39	Tuscola	S	R	R	S	R	R	S	R	R	R	R	R	73

Kelly, J.D., Afanador, L., and Cameron, LS. 1994. New races of *Colletotrichum lindemuthianum* in Michigan and implications in dry bean resistance breeding. Plant Dis. 78:892-894.

Padder, B.A., Sharma, PN., Awale, HE., and Kelly, JD. 2017. *Colletotrichum lindemuthianum*, the causal agent of bean anthracnose. J. Plant Pathology 99: 317-330. doi: 10.4454/jpp.v99i2.3867

Zuiderveen et al. 2016 Genome-wide association study of anthracnose resistance in Andean beans. PLOS One 11(6):e0156391.doi:10.137/journal.pone.0156391

DEVELOPMENT AND APPLICATION OF 50K VERSUS 6K BEADCHIP SNP ASSAYS IN SOYBEAN MAY PROVIDE A REFERENCE FOR DEVELOPING SNP ASSAYS IN COMMON BEAN

Qijian Song[1], Gaofeng Jia[1], Charles Quigley[1], Edward Fickus[1], David Hyten[1,2], Randall Nelson[3] and Perry Cregan[1]

[1] USDA-ARS, Soybean Genomics and Improvement Lab., Beltsville, MD 20705, USA
[2] Present address: Pioneer Hi-Bred International Inc, Johnston, IA
[3] USDA-ARS, Soybean/Maize Germplasm, Pathology and Genetics Research Unit and Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

INTRODUCTION: SNP markers are widely used to evaluate genetic diversity, identify genes/QTL or genomic regions controlling disease and environmental stress resistance, seed quality and productivity. There are two major approaches for SNP detection, i.e. genotyping by BeadChips (Song *et al.* 2013) and genotyping by sequencing (GBS) (Huang *et al.* 2009). Compared to GBS, the BeadChip assay has several advantages: low cost and high efficiency; high quality SNP allele calls with few or no missing calls and thus no imputation of the SNP allele is required and no bioinformatics skill or high-performance computing is needed for data analysis. The BeadChip assay is especially useful for genotyping RIL populations in early generations with a high heterozygote rate. In addition, genotyping different families with the same set of SNPs present in the BeadChip will facilitate combined QTL analysis across different RIL populations. Genome-wide association analysis (GWAS) and linkage mapping are commonly applied to associate genes/QTL with markers. GWAS usually requires a large number of markers to capture the haplotype diversity in the genome, while linkage mapping does not due to the limited number of recombination events among the RILs derived from bi-parent. Evaluation of too many markers will not only increase the cost but also generate redundant information (Song *et al.* 2017). Therefore, our objectives were to develop high throughput Illumina Beachip assays with high and low density markers and apply the assays to genetic and genomics research in soybean. The work on soybean may serve as a reference to common bean.

MATERIALS AND METHODS: ***Development of soybean SoySNP50K BeadChip:*** DNA from six cultivated soybean cultivars (Essex, Evans, Archer, Minsoy, Noir 1, Peking) and two wild soybean genotypes (PI468916 and PI479752) were sequenced for SNP discovery using Illumina platform, the short reads were aligned to the soybean whole genome sequence Glyma1.01 (Schmutz *et al.* 2010). SNPs were identified using the software MAQ and Illumina CASAVA software (Illumina Inc, San Diego, CA). Called SNPs were eliminated for Infinium design if the SNPs did't meet the requirements described by Song et al. (2013).Because the recombination rate was five times bigger in the euchromatic regions than the heterochromatic regions in soybean, the number of SNPs to be included in the BeadChips was five times higher in the euchromatic than the heterochromatic regions. A total of 60,800 high quality SNPs were chosen evenly from the two regions of each chromosome using the in-house computer algorithm and program.

Genotyping the USDA-ARS Soybean Germplasm with SoySNP50K BeadChips: The SoySNP50K BeadChips were used to genotype the USDA-ARS Soybean Germplasm Collection consisting of 18,480 *G. max* and 1,160 *G. soja* accessions (Song *et al.* 2015). Protocol for SNP assay and allele call was described by Song et al. (2015).

Designing BARCSoySNP6K containing 6,000 SNPs: The haplotype map including the haplotype block size and positions in the *G. max* genome were determined based on the analysis

of the 18,480 *G. max* accessions profiled with SoySNP50K SNPs. Because the SNPs in the same haplotype blocks are in strong linkage disequlibrium, these SNPs most likely provide redundant information, such as co-segregation with the markers from the same haplotype blocks on the linkage maps constructed from RIL populations. Thus, only one SNP was selected from each haplotype block with size>50kb in the euchromatic region and >100kb in the heterochromatic regions to be included in the BARCSoySNP6K BeadChip. The remaining SNPs from the 6,000 were then evenly selected between haplotype blocks.

RESULTS: ***The SoySNP50K BeadChips:*** A total of 50,701 in euchromatic regions (111 SNPs/Mbp), 10,000 in heterochromatic regions (20 SNPs/Mbp), and 99 SNPs from 71 unanchored scaffolds in the Glyma1.01 build were included in the ***SoySNP50K*** BeadChip. Of the 52,041 SNPs in the SoySNP50K bead pool, a total of 42,509 SNPs were polymorphic among the 19,648 soybean and wild soybean accessions characterized. Approximately 27% of the SNPs resided in genes and 24% were distributed within 2 kb upstream or downstream of genes.

Application of the genotypic dataset of the USDA Soybean Germplasm Collection to genetics and genomic research: The SoySNP50K dataset has been used to identify redundant or highly similar accessions and select a core set of *G. max* and *G. soja*, define haplotype blocks and LD among Wild, Landrace and U.S. Elite genotypes, explore the relationship of molecular genotypes with geographic origins of soybean, determine genomic regions associated with domestication and breeding selection, and to determine genome-wide association of markers with a number of traits, such as protein content and oil content, seed composition; seed size; carbon isotope signature (δ13C); resistance to sudden death syndrome, rot and stem rot, rust, frogeye leaf spot, SCN, and aphids; flowering time and maturity date and Ureide concentration(Hwang *et al.* 2014; Song *et al.* 2015).

The BARCSoySNP6K BeadChip: The BeadChip contained 6,000 SNPs consisting of 1,000 SNPs from heterochromatic and 5,000 from euchromatic regions. Analysis of progeny from two large families genotyped with SoySNP50K vs. the BARCSoySNP6K showed that position of the markers along linkage maps, position of seed size QTL and number of unique bins were consistent based on the two tools, however, the rate of redundant markers were dramatically reduced with the BARCSoySNP6K. We also observed that this tool has the same efficiency and resolution to identify genes controlling traits as that of whole genome sequencing approach in most populations created by breeders with population size less than 350.The BARCSoySNP6K is an excellent tool for the detection of QTL and for assessing genetic diversity in the families derived from bi-parents and is being widely used by the soybean community.

Acknowledgement: Thanks to Rob Parry and Chris Pooley for technical support in assembling the necessary hardware and software.

REFERENCES:

Huang, X., Q. Feng, Q. Qian, Q. Zhao, L. Wang *et al.*, 2009 Genome research **19:** 1068-1076.
Hwang, E.-Y., Q. Song, G. Jia, J. E. Specht, D. L. Hyten *et al.*, 2014 BMC genomics **15:** 1.
Schmutz, J., S. B. Cannon, J. Schlueter, J. Ma, T. Mitros *et al.*, 2010 nature **463:** 178-183.
Song, Q., D. L. Hyten, G. Jia, C. V. Quigley, E. W. Fickus *et al.*, 2013. PloS one **8:** e54985.
Song, Q., D. L. Hyten, G. Jia, C. V. Quigley, E. W. Fickus *et al.*, 2015 G3: 1999-2006.
Song, Q., L. Yan, C. Quigley, B. D. Jordan, E. Fickus *et al.*, 2017 The Plant Genome.
doi:10.3835/plantgenome2016.10.0109

THE RHIZOSPHERE MICROBIOME AS AN AUXILIARY BREEDING COMPONENT IN COMMON BEAN AGAINST *Fusarium oxysporum*

Lucas William Mendes[1,2*], Jos M. Raaijmakers[2,3], Rodrigo Mendes[4], Siu Mui Tsai[1]

[1]Cell and Molecular Biology Laboratory, Center for Nuclear Energy in Agriculture CENA, University of Sao Paulo USP, 13416-000, Piracicaba, SP, Brazil. E-mail: lwmendes@cena.usp.br
[2]Departament of Microbial Ecology, Netherlands Institute of Ecology NIOO-KNAW, 6708 PB, Wageningen, The Netherlands
[3]Institute of Biology, Leiden University, Leiden, the Netherlands
[4]Embrapa Meio Ambiente, 18020-000, Jaguariúna, Brazil.

The rhizosphere microbiome significantly impacts on plant growth, development and resistance against soil-borne pathogens (Berendsen *et al.*, 2012; Mendes *et al.*, 2013). It is now well documented that plants shape their rhizosphere microbiome composition through exudates that stimulate or repress the abundance of specific microbial genera (Lundberg *et al.*, 2012; Bulgarelli *et al.*, 2015). In return, rhizosphere microbes provide a range of ecosystem services for the plant, such as nutrient acquisition (Mendes *et al.*, 2014), abiotic stress tolerance (Philippot *et al.*, 2013), and protection against pathogens either directly via antagonism or indirectly via induction of systemic resistance (Mendes *et al.*, 2011; Berendsen *et al.*, 2012). Here, we investigated the composition and metabolic potential of the rhizobacterial community of different common bean (*Phaseolus vulgaris*) cultivars with variable levels of resistance to the fungal root pathogen *Fusarium oxysporum* (*Fox*). For the different bean cultivars grown in two soils with contrasting physicochemical properties and microbial diversity, rhizobacterial abundance was positively correlated with *Fox*-resistance. Pseudomonadaceae, Bacillaceae, Solibacteraceae and Cytophagaceae were more abundant in the rhizosphere of the *Fox*-resistant cultivar (Figure 1). Network analyses showed a modular topology of the rhizosphere microbiome of the *Fox*-resistant cultivar, suggesting a more complex and highly connected bacterial community than in the rhizosphere of the *Fox*-susceptible cultivar. Metagenome analyses further revealed that specific functional traits such as protein secretion systems and biosynthesis genes of antifungal phenazines and rhamnolipids were more abundant in the rhizobacterial community of the *Fox*-resistant cultivar. Metatranscriptome data revealed that community assembly in the rhizosphere follows niche-based mechanisms, presenting lower diversity and distinct community

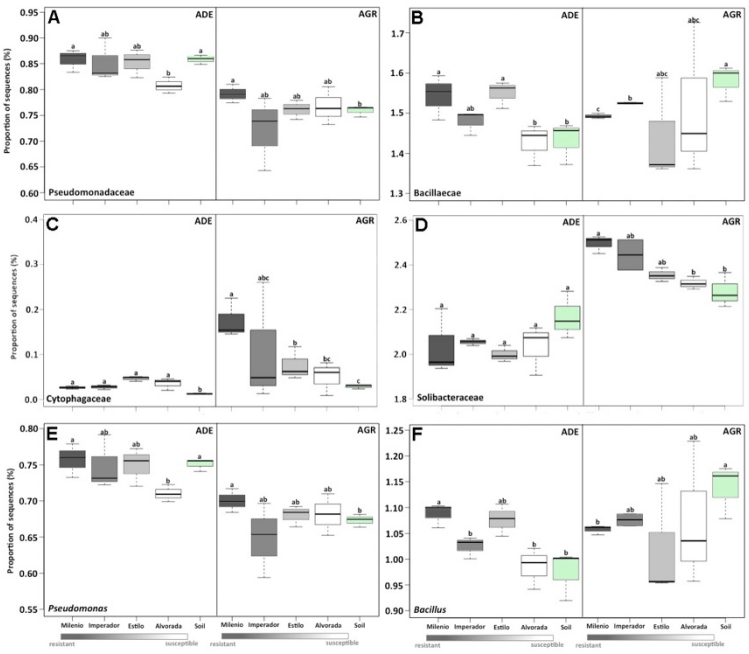


Figure 1. Distribution of the most differential bacterial groups in the rhizosphere of different common bean cultivars grown in Amazon Dark Earth (ADE) and agricultural soil (AGR).

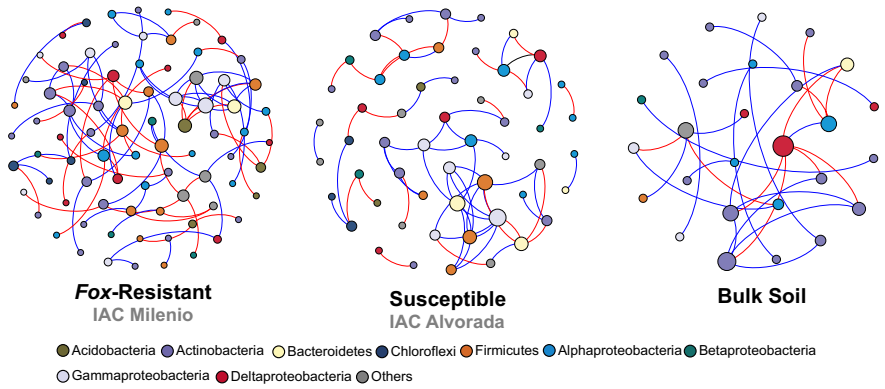


Figure 2. Network co-occurrence analysis of microbial communities in rhizosphere of two common bean cultivars with contrasting resistance to *Fusarium oxysporum*. Each dot represents a microbial phylotype.

structure comparing to the bulk soil. In comparison with the susceptible plant, the microbiome of the *Fox*-resistant cultivar presented high expression of genes affiliated to the family Paenibacillaceae, a group known by its antifungal activity. The *Fox*-resistant cultivar also presented high expression of genes related to metabolism of nutrients and specific functional traits related to pathogen suppression, such as motility and chemotaxis, and phenazine and colicin V. Network analysis showed similar results to the metagenome approach, revealing a more complex community in the *Fox*-resistant cultivar and pointed the genus *Paenibacillus* as a keystone species in the microbiome (Figure 1).

Our findings suggest that breeding for *Fox*-resistance in common bean have co-selected for other unknown plant traits that support a higher abundance of specific beneficial bacterial families in the rhizosphere with functional traits that support a more complex rhizosphere microbiome and reinforce the first line of defense against the pathogen. Also, our study reinforces the importance of understanding the processes of microbiome assembly in the rhizosphere, where the identification of microbial groups and traits related to pathogen suppression could help the future development of plant breeding to select for plant traits that enrich and activate specific microbial genera and genes, and support a more complex and modular network structure of the rhizosphere microbiome.

REFERENCES

Berendsen RL, Pieterse CMJ, Bakker PAHM. 2012. The rhizosphere microbiome and plant health. *Trends in Plant Science* 17: 478–486.

Bulgarelli D, Garrido-Oter R, Münch PC, Weiman A, Dröge J, Pan Y, McHardy AC, Schulze-Lefert P. 2015. Structure and Function of the Bacterial Root Microbiota in Wild and Domesticated Barley. *Cell Host & Microbe* 17: 392–403.

Lundberg DS, Lebeis SL, Paredes SH, Yourstone S, Gehring J, Malfatti S, Tremblay J, Engelbrektson A, Kunin V, Rio TG del, *et al.* 2012. Defining the core Arabidopsis thaliana root microbiome. *Nature* 488: 86–90.

Mendes R, Garbeva P, Raaijmakers JM. 2013. The rhizosphere microbiome: significance of plant beneficial, plant pathogenic, and human pathogenic microorganisms. *FEMS microbiology reviews* 37: 634–663.

Mendes R, Kruijt M, Bruijn I de, Dekkers E, Voort M van der, Schneider JHM, Piceno YM, DeSantis TZ, Andersen GL, Bakker PAHM, *et al.* 2011. Deciphering the Rhizosphere Microbiome for Disease-Suppressive Bacteria. *Science* 332: 1097–1100.

Mendes LW, Kuramae EE, Navarrete AA, van Veen JA, Tsai SM. 2014. Taxonomical and functional microbial community selection in soybean rhizosphere. *The ISME Journal* 8: 1577–1587.

Philippot L, Raaijmakers JM, Lemanceau P, van der Putten WH. 2013. Going back to the roots: the microbial ecology of the rhizosphere. *Nature Reviews Microbiology* 11: 789–799.

ADAPTABILITY AND STABILITY OF ELITE LINEAGES AND COMMON BEAN CULTIVARS OF SPECIAL GRAINS IN DIFFERENT SEASONS OF CULTIVATION IN MINAS GERAIS, BRAZIL

O.G. Brito[1], A.J. de Carvalho[2], V.B. de Souza[3], M.B. de Oliveira[2], T.L.P.O. Souza[4], J.E. de S. Carneiro[5], J.A.A. Moreira[5], M. Martins[7], L.C. Melo[4], L.C. Faria[4], H.S. Pereira[4], J.C. Medeiros[2], M.L. Lacerda[2], A.A. de Souza[2], J.V.S. Guerra[2]

[1]Universidade Federal dos Vales do Jequitinhonha e Mucuri, [2]Universidade Estadual de Montes Claros, [3]Universidade Estadual de Maringá, [4]Embrapa Arroz e Feijão, [5]Universidade Federal de Viçosa, [6]Embrapa Milho e Sorgo, [7]Universidade Federal de Uberlândia.
Corresponding author: vanetbatista@yahoo.com.br

INTRODUCTION: The special beans comprise beans of Andean and Mesoamerican origin and present varied colors, sizes and shapes. In Brazil the beans Mesoamerican are more consumed especially the type Carioca and Black. The Andean beans produced in Brazil meet a growing demand for beans for export, with type of large and shiny grains favorite in the international market. Common bean is cultivated under different edaphoclimatic conditions, such as different seasons and planting systems, and this crop variation directly affects the selection processes of lineages in breeding programs. Cultivars with predictable behavior that best respond to environmental variations can be identified from the adaptability / stability analysis, both in specific and broad conditions (Cruz & Regazzi, 2003). This work aimed to identify elite lineages and cultivars of special grains with better agronomic performance, adaptability and productivity stability at different growing seasons in the state of Minas Gerais, Brazil.

MATERIAL AND METHODS: The experiments were carried out in the cities of Sete Lagoas, Uberlândia, Janaúba and Jaíba, in the seasons of spring-summer (october-november), summer-fall (february-march) and fall-winter (april-june) seasons (VIEIRA et al., 2006) between the years of 2010 and 2013. Twelve lineages and four cultivars were evaluated in nine environments (place / season / year combinations), four in the fall-winter season, three in the summer-fall and two in the spring-summer. The grain productivity data were submitted to the analysis of variance for each growing season and, when significant, the averages of the lineages and cultivars were compared by the Scott-Knott group method at 5% significance. The analysis of adaptability and stability of lineage productivity was performed by the method of Annicchiarico (1992), which is based on the recommendation index of the genotype (ω_i), estimated by the equation $\omega i = \mu i - z_{(1-\alpha)} \sigma_{zi}$, where: μ_i is the average of genotype i (%); σ_{zi} is the standard deviation of the Z_{ij} values associated with the i-th genotype; $z_{(1\ \alpha)}$ is the percentile of the standard normal distribution function. The coefficient of confidence adopted was 75%, that is, $\alpha = 0.25$. Adaptable / stable genotypes present ωi higher than 100%. For the analysis, the GENES program was used (CRUZ, 2013).

RESULTS AND DISCUSSION: Considering the recommendation index of the genotype (ω_i) and the lineages productivity (Table 1), it was observed that in the winter season the best adaptability and stability were CNFRx 15275 (115.39%), Jalo EEP (115.35%) and PT-68 (107.60%) and BRS Radiante (103.38%). In the water season, the best performance lineages were VR-18 (157.03%), CNFRx 15275 (151.06%), BRS Vereda (149.67%), VR-16 (123.71%) and BRS Timbo (114.43%). For the dry season, RC2RAD-155 (11.05%), VR-18 (107.76%), CNFRx 15275 (107.09%), BRS Vereda (120.86%) and BRS Radiante (113.62%) stood out. The lineage CNFRx 15275 showed high productivity, adaptability and stability in all growing seasons.

TABLE 1. Grain productivity and adaptability and stability parameters estimates for elite lineages and common bean cultivars of especial grains classes in the three growing seasons in Minas Gerais, Brazil.

GENOTYPE	Class**	Productivity (kg ha^{-1})			Annicchiarico					
		Spring-Summer	Summer-Fall	Fall-winter	ω_i [2] Spring-Summer	C[3]	ω_i Summer-Fall	C	ω_i Fall-winter	C
CNFR × 15275	Rx	2600 Aa[1]	1682 Ba	1665 Ba	115.39	1	151.06	2	107.09	5
JALO EEP	M.J	2506 Aa	849 Bb	1285 Ba	115.35	2	86.94	7	72.96	14
PT-68	Rs	2288 Aa	903 Bb	1236 Ba	107.60	3	67.13	11	73.15	13
BRS RADIANTE	M.R	2249 Aa	446 Bb	1660 Aa	103.38	4	33.80	16	113.62	2
RC2RAD-155	M.R	2127 Aa	868 Bb	1678 Aa	93.84	9	94.01	6	111.05	3
BRS VEREDA	Rs	2072 Aa	1384 Aa	1823 Aa	95.03	8	149.67	3	120.86	1
VR-16	Vr	2061 Aa	1291 Ba	1293 Ba	98.16	5	123.71	4	80.72	12
VR-14	Vr	2039 Aa	828 Bb	1396 Ba	95.80	7	83.45	8	89.23	9
BRS TIMBO	Rx	2024 Aa	1141 Ba	1168 Ba	96.80	6	114.43	5	68.28	15
CNFJ 15288	M.J	1995 Aa	534 Bb	1588 Aa	93.32	10	55.02	13	86.40	11
VR-18	Vr	1986 Aa	1719 Aa	1581 Aa	83.46	14	157.03	1	107.76	4
RAD/E550-284	M.R	1972 Aa	323 Bb	984 Ba	90.94	11	34.28	15	62.99	16
PT-65	Rs	1928 Aa	501 Bb	1451 Aa	89.87	12	46.41	14	95.89	7
VR-15	Vr	1860 Aa	701 Bb	1337 Aa	85.93	13	66.78	12	88.52	10
VR-17	Vr	1811 Aa	813 Bb	1448 Aa	78.62	16	81.73	9	94.15	8
OURO VER.*	Vr	1786 Aa	705 Bb	1474 Aa	81.34	15	75.35	10	97.54	6
CV (%)[4]		18.98	20.08	17.98						
Average (kg ha^{-1})		2081	918	1441						

* Red Gold; ** Commercial class (Rx: Purple / M.J: Manteigão Jalo / M.R Manteigão Rajado / Rs: Light Pink / Vr: Red); [1]Averages followed by the same letter, uppercase and lowercase in the column, do not differ by Scott-Knott's test at 5% significance; [2]ωi = recommendation index of the genotype; [3]Classification; [4]Coefficient of variation.

The behavior of the climatic factors in different harvests may have affected the behavior of the differentiated of the lineages, besides the genetic differences. High temperatures, for example, promote floral abortion, and consequently reduction of productivity (PEREIRA et al., 2014), directly affecting the parameters of adaptation and stability.

REFERENCES

ANNICCHIARICO. P. Cultivar adaptation and recommendation from alfafa trials in Northern Italy. Journal of Genetics and Breeding. v. 46. n. 1. p. 269-278. 1992.

CRUZ. C. D. GENES - a software package for analysis in experimental statistics and quantitative genetics. Acta Scientiarum. Agronomy. Maringá. v. 35. n. 3. p. 271-276. 2013.

CRUZ, C. D.; REGAZZI, A. J. Modelos biométricos aplicados ao melhoramento genético. 2. ed. rev. Viçosa: UFV, 2003.

PEREIRA, V.G.C.; GRIS, D.J.; MARANGONI, T.; FRIGO, J.P.; AZEVEDO, K.D.; GRZESIUCK, A.E. Exigências Agroclimáticas para a Cultura do Feijão (*Phaseolus vulgaris* L.). Revista Brasileira de Energias Renováveis, v. 3, 2014.

VIEIRA, C.; PAULA JR., T.J.; BORÉM, A. (eds.) Feijão. 2ed. amp./rev. Viçosa, Editora UFV, 2006. 600p.

TOWARDS UNDERSTANDING THE EFFECT OF PATHOGENS AND SALINITY ON ROOT ROT DISEASE IN COMMON BEANS

Dylan Lynch[1], Joey Cainong[1*], Girly Ramirez[2], Rubella S. Goswami[3], James Steadman[4], and Venu (Kal) Kalavacharla[1,5]

[1]College of Agriculture and Related Sciences, Delaware State University, Dover, DE 19901, [2]Dept. of Statistics, Kansas State University, Manhattan, KS 66506, [3]Division of Plant Protection, USDA-NIFA, Washington, DC 20024, [4]Dept. of Plant Pathology, University of Nebraska, Lincoln, NE 68583, [5]Center for Integrated Biological and Environmental Research (CIBER), Delaware State University, Dover, DE

INTRODUCTION: The root rot disease complex, caused mostly by *Fusarium* spp. and *Rhizoctonia solani*, limits common bean (*Phaseolus vulgaris* L.) production by causing root rot, damping-off, seedling blight and a general reduction in plant stand, root distribution, root vigor, and seed germination (Gossen *et al*., 2016). Salt-affected soils exist in nearly 20% of total cultivated and 33% of irrigated agricultural lands (Jamil *et al*., 2011). Salinity affects fungal mycelial production, conidial formation and spore germination (Jones *et al*., 2011). Our goal was to evaluate the effect of salinity on the mycelial growth of fungi when grown in sodium chloride (NaCl)-amended potato dextrose agar (PDA) plates.

MATERIALS AND METHODS: The isolates used were *F. graminearum* (31084), *F. solani* (Mi), *R. solani* (UD8, Wn11 and Wn293) and grown at 24 $\pm$ 1°C with 65% RH on NaCl-amended PDA plates to have final salt concentrations of 0, 50, 150, 200, 250 and 400 mM. Two mycelial growth measurements were taken perpendicularly at time points 3, 6 and 10 days after transfer. The collected data was treated as a longitudinal data or repeated measures data since the same Petri plate was used at different time points. Descriptive plots and statistical analyses were done in R 3.4.1 (R Core Team, 2017). The NLME package (Pinheiro *et al*., 2017) and R functions ***lm*** (linearized) and ***gls*** (generalized least square) were used to determine the best fit model for each isolate. The statistical models evaluated the dependent variable (mycelial growth) affected by salt, time of exposure (day), and their interaction. The best-fit model was indicated by the lowest Akaike's Information Criterion (AIC) value. Since data between time points were correlated, the structure of its covariance matrix was examined using corSymm (unstructured) and corAR1 (autoregressive 1).

RESULTS: The 270 data points collected were used in descriptive plot analysis and a non-linear growth pattern prompted the use of generalized least square (gls) in succeeding analysis. Figure 1 shows the plots generated with repeated measures ANOVA. **Fig 1A**. The *F. graminearum* (31084) isolate was least affected by salt, with plots of different salt concentrations having similar trends. The predicted growth (red line) were maximum at Day 10. **Fig 1B.** The *F. solani* (Mi) isolate was the most affected by salt with growth reduction starting at 150 mM. Maximum growth was at Day 10 for all salt treatments. **Fig 1C and 1D.** In *R. solani* (Wn11 and Wn293) isolates, maximum growth for the control (0 mM) was at Day 6. Higher salt concentrations (50 mM and above) had maximum growth at Day 10. **Fig 1E.** The mycelial growth of *R. solani* (UD8) was fastest among the isolates. At 0 and 50 mM, optimal growth was at Day 6 but the predicted growth was higher than 80 mm. The model did not completely fit the observed mycelial growth patterns for this isolate since growth was restricted by the diameter of the Petri dish.

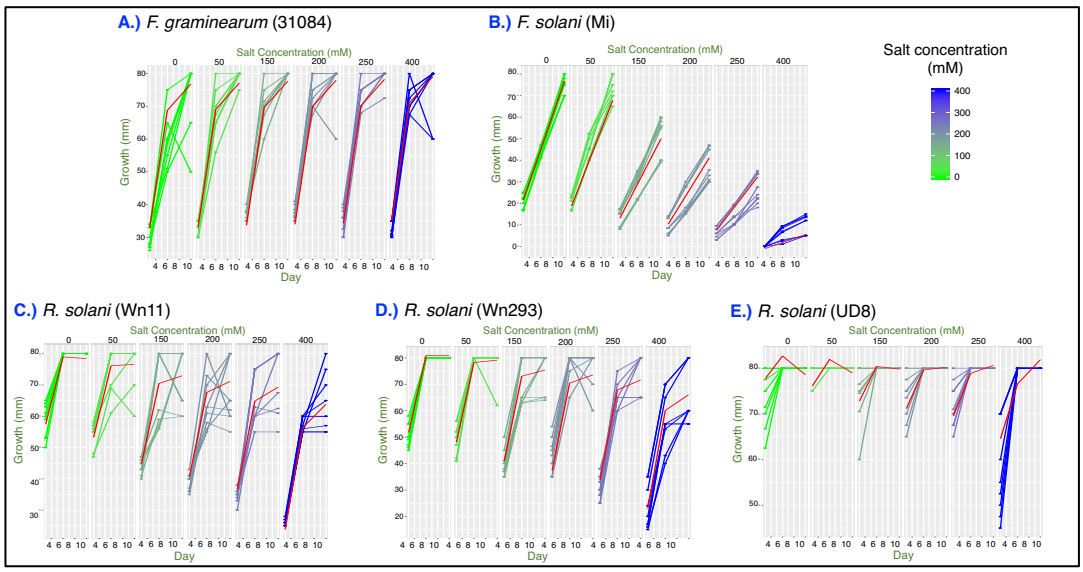


Figure 1. Mycelial growth (mm) as affected by salt concentrations at time points Day 3, 6 and 10. The growth line predicted by the statistical model is shown in red.

DISCUSSION AND CONCLUSIONS: The statistical analysis of longitudinal data or repeated measures allowed evaluation of how each isolate responds to salt concentrations over time. The non-linear growth of the fungi suggests that growth rates between time points were not constant. Maximum growth, seen as the highest point in the predicted growth line (red line), can be used to assess the time point until which growth measurements are to be taken. The curve suggested that for *R. solani* (UD8), mycelial growth needs to be measured earlier than Day 3 since some cultures have overgrown the plate even at Day 3. Also, time points less than 3 days apart are recommended due to their faster mycelial growth rate. Overall, mycelial growth at 0 and 50 mM have similar trends thereby suggesting that effect of salinity is minimal at salt levels below 50 mM, which is supported by the fact that soils are only considered saline at roughly 40 mM NaCl. In this study, the effects of salinity were primarily recorded at NaCl concentrations higher than 150 mM. These findings suggest that there is variation in sensitivity among fungal species, as well as isolates within each species, to different salt concentrations. The duration of the data collection could also affect conclusions drawn with regards to factors affecting mycelial growth rate. The statistical procedures used in this study could be applied to experimental data involving multiple variables common in agricultural studies.

ACKNOWLEDGEMENTS: For funding: NSF-Delaware EPSCoR #1301765. For isolates: USDA-ARS Culture Collection, Peoria, IL; Nancy Gregory, University of Delaware; James Steadman, University of Nebraska Lincoln.

REFERENCES:

Gossen, B. D., Conner, R. L., Chang, K. F., Pasche, J. S., McLaren, D. L., Henriquez, M. A., ... & Hwang, S. F. (2016). Identifying and managing root rot of pulses on the northern Great Plains. *Plant Disease*, *100*(10), 1965-1978.

Jamil, A., Riaz, S., Ashraf, M., & Foolad, M. R. (2011). Gene expression profiling of plants under salt stress. *Critical Reviews in Plant Sciences*, *30*(5), 435-458.

Jones, E. E., Stewart, A., & Whipps, J. M. (2011). Water potential affects Coniothyrium minitans growth, germination and parasitism of Sclerotinia sclerotiorum sclerotia. *Fungal biology*, *115*(9), 871-881.

Pinheiro J, Bates D, DebRoy S, Sarkar D and R Core Team (2017). _nlme: Linear and Nonlinear Mixed Effects Models_. R package version 3.1-131, <URL: https://CRAN.R-project.org/package=nlme>.

R Core Team (2017). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

IDENTIFICATION AND CHARACTERIZATION OF ROOT ROT PATHOGENS OF DRY BEAN IN MICHIGAN

Janette L. Jacobs, Devon R. Rossman, Kjersten Oudman, Hyunkyu Sang, and Martin I. Chilvers

Department of Plant, Soil and Microbial Sciences, Michigan State University
East Lansing, Michigan, USA

INTRODUCTION

Root rot, caused by a complex of *Fusarium* spp., *Pythium* spp., and *Rhizoctonia solani* is a constant disease problem in dry bean production in Michigan. Root rot often leads to poor stand establishment, reduced plant vigor, and potential yield loss. It is not uncommon for growers to replant sections or whole fields due to poor stand establishment as a result of root rot pathogens. Identification of the organisms responsible for stand loss and root rot would provide valuable information for plant breeding efforts and identifying the best seed treatments to manage disease. The objectives of this research were to i) conduct a survey of Michigan commercial dry bean fields to determine which pathogens are associated with stand loss and root rot symptoms; ii) characterize the isolates for pathogenicity and virulence in a seedling assay.

MATERIALS AND METHODS

Symptomatic plants were collected from 30 fields in eight counties from 2014 – 2017. Oomycete and fungal isolations were made on CMA-PARPB and WA amended with metalaxyl and streptomycin, respectively. DNA was extracted from lyophilized mycelium, and ITS and TEF-1α gene were utilized for identification of isolates. Pathogenicity screening was conducted with a representative panel of oomycete and *Fusarium* spp. on red kidney (cv. Red Hawk) and black bean (cv. Zorro) in a seedling assay (Rossman et al. 2017). *Rhizoctonia solani* isolates representing anastomosis groups (AG), AG2-2, AG2-3, AG4, AG5, and AG11 were tested for pathogenicity against dry bean, as well as soybean (cv. Sloan), and corn (cv. DK52-61).

RESULTS AND DISCUSSION

A total of 859 isolates were recovered during the survey period. *Fusarium* spp. were isolated in the greatest abundance across years, with the exception of 2014. The majority of isolates were identified as belonging to the *Fusarium solani* species complex (FSSC), with the second most abundant being the *Fusarium oxysporum* species complex (FOSC). *Rhizoctonia solani* isolates representing anastomosis groups (AG), AG2-2, AG2-3, AG4, AG5, and AG11 were identified. A total of 28 different oomycetes species were isolated representing *Pythium* and *Phytopythium* genera.

CONCLUSIONS

Fusarium spp. were the most commonly isolated dry bean pathogens in three of four survey years. Isolates within the FSSC and FOSC produced the highest disease severity index (DSI) on both bean seed types. *Rhizoctonia solani* isolates within AG2-2 and AG4 were the most aggressive on dry bean and soybean resulting in the greatest disease severity and decreased root dry weight. *Pythium ultimum*, *Pythium myriotylum*, and *Phytopythium* aff. *vexans* significantly reduced emergence of both seed types. *Pythiu*m aff. *diclinum*, *Pythium irregular*, *Pythium lutarium*, and *Pythium sylvaticum* significantly reduced dry root weight, root length, and root area in both Andean and Mesoamerican germplasm.

ACKNOWLEDGEMENTS
We gratefully acknowledge the financial support of Project GREEEN, the Michigan Bean Commission, and Michigan Department of Agriculture and Rural Development.

RELATED PUBLICATION
Rossman, D.R., Rojas, A., Jacobs, J.L., Mukankusi, C., Kelly, J.D., Chilvers, M.I. 2017. Pathogenicity and virulence of soilborne oomycetes on *Phaseolus vulgaris*. Plant Disease 101:1851-1859

PHENOTYPING FUNGAL AND OOMYCETE ROOT ROT PATHOGENS FOR FUNGICIDE SENSITIVITY

Alex Witte, Zachary A. Noel, J. Alejandro Rojas, Janette L. Jacobs, Martin I. Chilvers

Department of Plant, Soil and Microbial Sciences, Michigan State University, East Lansing, Michigan, USA

INTRODUCTION

Oomycetes and Fusarium cause significant damage to legume crops worldwide. Fungicide seed treatments are a standard method used to manage root rot pathogens. It is useful to test the active compounds used in seed treatments against target pathogens.

The traditional technique for evaluating *in vitro* fungicide sensitivity is the poison plate assay. However, this method limits both the number of isolates and number of chemistries that can be tested at one time, and is labor-intensive. A poison plate assay was used to evaluate the efficacy of fluopyram, a succinate dehydrogenase inhibitor, against 8 *Fusarium* species. A high-throughput microtiter assay was used to screen oomycetes against mefenoxam and ethaboxam.

The objectives of this research were to i) use a poison plate assay to evaluate the sensitivity of Clade 2 members of the *Fusarium solani* species complex (FSSC) to the SDHI fungicide fluopyram; ii) screen isolates across 8 FSSC species for sensitivity to fluopyram; iii) establish EC50 values for 61 oomycete isolates consisting of 20 *Pythium* spp. and *Phytophthora sansomeana*, isolated from Michigan; iv) screen oomycete isolates across two fungicidal chemistries, mefenoxam and ethaboxam, using a high-throughput technique

MATERIALS AND METHODS

Fusarium solani species complex

Isolates were grown for 2 weeks on ½ strength potato dextrose agar. Fluopyram (Luna Privilege, Bayer Crop Sciences) was amended into half-strength potato dextrose agar to final concentrations of 0, 0.5 1.0, 3.0, 5.0, 7.0, 10.0, 50.0 µg ml-1. Inoculated poison plates were incubated at 24°C for one month. Plates were scanned at 3, 10, 17, and 28 days after inoculation using an Epson Perfection V600 Series scanner. Colony areas were measured using APS Asses 2.0 Plant Disease Quantification Software. Colony area growth rates were calculated as the change in cm2/day following the day 3 measurements over a 7, 14, or 25 day period, depending on isolate growth rate. Percent growth values relative to the control were loaded into a 4-parameter log-logistic model, using the "drc" package in R, and dose-response curves were generated. EC50 values (fungicide concentration inhibiting mycelia growth by 50%) were estimated from dose-response curves.

Oomycetes

Isolates grown 2-3 days on ¼ strength potato carrot agar medium. 2-3 day old colonies macerated through a 10-ml syringe and inoculated into 96-well microtiter plate wells filled with ½ strength V8 broth. Wells contained final fungicide concentrations of 0, 0.01,0.1, 0.5, 1.0, 10, and 100 µg mL-1. Plates incubated for 48 hrs at 24°C. Optical density (OD) values determined using a microplate reader. EC50s estimated from OD readings using a 4-parameter logistic curve

RESULTS AND DISCUSSION

Fusarium solani species complex

Of the 80 isolates screened against fluopyram, 75 were sensitive within 10 µg ml-1. *Fusarium javanicum*, 3 isolates of *Fusarium solani*, and 1 isolate of *Fusarium virguliforme* were significantly less sensitive (EC50 > 50 µg ml-1) to fluopyram. *Fusarium tucumaniae* (n=4) was the most sensitive species, with a mean EC50 of 0.116 µg ml-1.

Oomycetes

47 isolates had EC50 < 1 µg ml-1 to ethaboxam. For ethaboxam, *Pythium* aff. *torulosum* and *Pythium rostratifingens* were found to be significantly less sensitive (EC50 > 100 µg ml-1). 57 isolates had EC50 < 1 µg ml-1 to mefenoxam. 3 isolates of *Pythium ultimum* var. *ultimum* were significantly less sensitive to mefenoxam (EC50 > 85 µg ml-1).

CONCLUSIONS

Fusarium solani species complex

All 8 members of FSSC Clade 2 tested are sensitive to fluopyram. In the future outliers will be addressed with sequencing and further identification. We are investigating mechanism that allows *F. tucumaniae* to respond to fluopyram at lower doses than other species.

Oomycetes

Isolates were less sensitive to ethaboxam (mean EC50) than to mefenoxam (mean EC50). Higher EC50 values in response to ethaboxam were strongly correlated within *Pythium* aff. *torulosum* and *Pythium rostratifingens*. Variability was detected among isolates of *Pythium ultimum* var. *ultimum*, therefore further identification will be addressed.

ACKNOWLEDGEMENTS

We gratefully acknowledge the financial support of Project GREEEN, the Michigan Bean Commission, Michigan Department of Agriculture and Rural Development, and Valent Crop Sciences.

RELATED PUBLICATIONS

Rojas, J.A, Jacobs J., Napieralski, S., Karaj, B., Bradley, C.A., Chase, T., Esker, P., Giesler, L., Jardine, D., Malvick, D. Markell, S., Nelson, B., Robertson, A., Rupe, J., Smith, D., Sweets, L., Tenuta, A., Wise, K., Chilvers, M.I. 2017. Oomycete species associated with soybean seedlings in North America – Part I: Identification and pathogenicity characterization. Phytopathology 107:280-292

Rossman, D.R., Rojas, A., Jacobs, J.L., Mukankusi, C., Kelly, J.D., Chilvers, M.I. 2017. Pathogenicity and virulence of soilborne oomycetes on *Phaseolus vulgaris*. Plant Disease 101:1851-1859

Wang, J., Bradley, C.A., Stenzel, O., Pedersen, D.K., Reuter-Carlson, U., Chilvers, M.I. 2017. Baseline sensitivity of *Fusarium virguliforme* to fluopyram fungicide. Plant Disease 101:576-582

TOTAL DIETARY FIBER CONTENT OF DRY BEAN, DRY PEA, CHICKPEA, AND LENTIL CULTIVARS USING AOAC 2011.25 INTEGRATED ASSAY

Chen Y[1], Thompson HJ[1], Brick[2] MA, Vandemark GJ[3], McGee RJ[3], and B. Ogg[2]

[1]Colorado State University, Department of Horticulture and Landscape Architecture
[2]Colorado State University, Department of Soil and Crop Sciences
[3]USDA/ARS, Pullman WA

INTRODUCTION

The human health benefits of dietary fiber (DF) in food crops are well documented. Pulse crops were highlighted in 2016 during the "Year of the Pulse" for their contribution to food security worldwide and health beneficial effect (Brick and Fisher, 2016). However, the consumption of DF in the human diet is lower than recommended, and the gap between actual consumption and recommended intake represents an unrecognized health risk. Many dieticians propose an increase in pulses as a practical way to close the dietary fiber gap. A systematic measurement of the contents of DF and its constituents in pulses has not been conducted since the AOAC 2011.25 method for measuring DF was adopted in 2011. A better understanding of DF content and its components in pulse crops may promote pulse consumption as whole grain and as an ingredient in pulse crop flour.

MATERIALS AND METHODS

Total dietary fiber was measured in 26 cultivars of dry bean (*Phaseolus vulgaris* L.), 11 cultivars of dry pea (*Pisum sativum* L), 13 cultivars of lentil (*Lens culinaris* Medik) and 24 cultivars of chickpea (*Cicer arietinum* L), each grown at two locations. Dietary fiber analysis was determined using the AOAC 2011.25 method as modified by Kleintop et al. (2013).

RESULTS

Mean insoluble dietary fiber (IDF), soluble dietary fiber (SDF), oligosaccharide (OLIGO) and total dietary fiber (TDF) content varied among the four pulse crops (Table 1). Mean TDF content for chickpea was 21.8%, dry bean 25.8%, dry pea 24.7%, and lentil 20%. The range in the components of DF also varied among pulse crops (Table 2). Total dietary fiber among cultivars within crop ranged from 19.5 to 24.9% for chickpea, 24.1 to 27.4% for dry bean, 22.3 to 28% for dry pea, and 18.4 to 21.3% for lentil. Dietary fiber components (IDF, SDF, and TDF) also varied ($P < 0.05$) among cultivars within pulse crops (data not shown). Location effects were not significant for most components of DF. These results indicate that genetic variation occurs among pulse crops and within cultivars of the four pulses. In general, all four pulses had high TDF relative to cereal crops and possessed adequate genetic diversity to improve DF by selection. Consumption of daily servings of any of these pulse crops would reduce the dietary fiber gap that exists today. In fact, one serving per day would supply approximately 25 to 30% of the minimum daily requirement in the human diet. This data will also contribute to a better understanding of the variation for DF in pulse crops when used as whole grains or as ingredients in processed food.

Table 1. Mean insoluble dietary fiber (IDF), soluble dietary fiber (SDF), oligosaccharide (OLIGO), total dietary fiber (TDF) content, and g of total dietary fiber in one serving of pulse crop.

Crop	% IDF[a]	% SDF[a]	% OLIGO[a]	% TDF[a]	TDF/serving g
Chickpea	15.8^{A}	3.5^{A,C}	2.5^{C}	21.8^{B}	7.1
Dry bean	13.9^{B}	7.7^{B}	4.2^{A}	25.8^{A}	8.1
Dry pea	16.2^{A}	3.9^{A,C}	4.6^{A}	24.7^{A}	4.3
Lentil	13.6^{B}	3.1^{A}	3.3^{B}	20.0^{C}	6.0
p-value	<0.001	<0.001	<0.001	<0.001	

[a]Values are expressed as percent of dry weight total. Means with different superscripts within a column are significantly different (P<0.05). Abbreviations: OLIGO = raffinose + stachyose + verboscose; TDF = IDF + SDF + OLIGO.

Table 2. Range in percent insoluble dietary fiber (IDF), soluble dietary fiber (SDF), total oligosaccharides (OLIGO), and total dietary fiber (TDF) among four pulse crops grown at two locations.

Crop	% IDF[a]	% SDF[a]	% OLIGO[a]	% TDF[a]
Chickpea	14.4 to 17.1	2.0 to 5.8	1.0 to 3.5	19.5 to 24.9
Dry bean	12.3 to 15.7	5.8 to 9.8	3.6 to 5.2	24.1 to 27.4
Dry pea	14.2 to 19.8	3.3 to 5.3	4.0 to 5.4	22.3 to 28.0
Lentil	12.3 to 4.7	2.7 to 3.9	3.0 to 3.7	18.4 to 21.3

[a]Values are expressed as percent of total seed dry weight. Abbreviations: OLIGO = raffinose + stachyose + verbascose; TDF = IDF + SDF + OLIGO.

REFERENCES:

1. Brick, M., and M. Fisher. 2016. 2016 is the International Year of Pulses. Crops Soils 49:38-38. doi:10.2134/cs2016-49-1-9.

2. Kleintop AE, Echeverria D, Brick LA, Thompson HJ, Brick MA. Adaptation of the AOAC 2011.25 integrated total dietary fiber assay to determine the dietary fiber and oligosaccharide content of dry edible beans. J.Agric.Food Chem. 2013 Oct 9;61(40):9719-26.

3. Chen, Y., R. McGee, G. Vandemark, M. Brick, and H.J. Thompson. 2016. Dietary Fiber Analysis of Four Pulses Using AOAC 2011.25: Implications for Human Health. Nutrients, 8: 829; doi:10.3390/nu8120829.

MATING SYSTEM IN THREE LANDRACES AND TWO IMPROVED VARIETIES OF *Phaseolus coccineus* L. ISOLATED FROM POLLINATORS

Ma. Luisa P. Vargas-Vázquez[1]; J. Kohashi-Shibata[1]; E. Uscanga-Mortera[1] and A. García-Esteva[1]

[1]Postgrado en Botánica, Colegio de Postgraduados, Montecillo, Edo. de México. 56230. México.

INTRODUCTION. *Phaseolus coccineus* L. is an allogamous species that requires carpenter bees and humming birds for pollination and seed production (Búrquez y Sarukhán, 1980) possibly due to floral mechanics or in causes of incompatibility not yet discovered (Búrquez y Sarukhán, 1984). Escalante *et al.* (1994) found that the levels of genetic variation in wild and cultivated *P. coccineus* are similar to that of wild species with mixed mating systems, and González *et al.* (2014) produced progeny by hand pollination during five generations and confirmed that this species displays mating systems of outcrossing and self-fertilization. The objective of this work was to study the mating system in three landraces and two improved varieties of *Phaseolus coccineus* L. isolated from pollinators**.**

MATERIALS AND METHODS. Three Mexican landraces of *Phaseolus coccineus* L. collected in Zitlaltepetl, Tlax., Nombre de Dios, Dgo., and Tlayacapan, Mor., as well as two improved varieties from Mexico (Blanco Tlaxcala), and Germany (Butler). Seeds were sown in containers of 20 liters capacity with soil, on September 9, 2016, in "Colegio de Postgraduados", Montecillo, in Estado de México. The experimental unit was one plant per container. The plants of indeterminate growth climbing type, were grown in a greenhouse, and each one was covered with a net during the flowering period. The experimental design was a complete random, with five treatments (landraces and varieties) and six replications (one plant per replication). The flowering period duration, the number of daily abscised flowers per plant, and the number of mature pods per plant were registered. The data obtained were subjected to ANOVA, and means were compared by the Tukey test ($p<0.05$) using the SAS program (SAS, 2012).

RESULTS AND DISCUSSION. The flowering period in collections from Zitlaltepetl was 151 days, Tlayacapan 151, Nombre de Dios, 85 and Blanco Tlaxcala 111; and in Butler it was only 36 days. The number of flowers per plant was statistically equal in Mexican collections, and statistically different in the Butler variety (Figure 1a). Only two seeds were obtained from the plants: one in Blanco Tlaxcala, and one in Butler variety, both seeds were viable. These landraces are not self-pollinated, so did not produce fruits without the active intervention of a pollinator agent. In addition, in Tlayacapan, Nombre de Dios, and Zitlaltepetl two different patterns of flower initiation: late flowering > 100 days, and early flowering < 100 days, were recorded (Figure 1b).

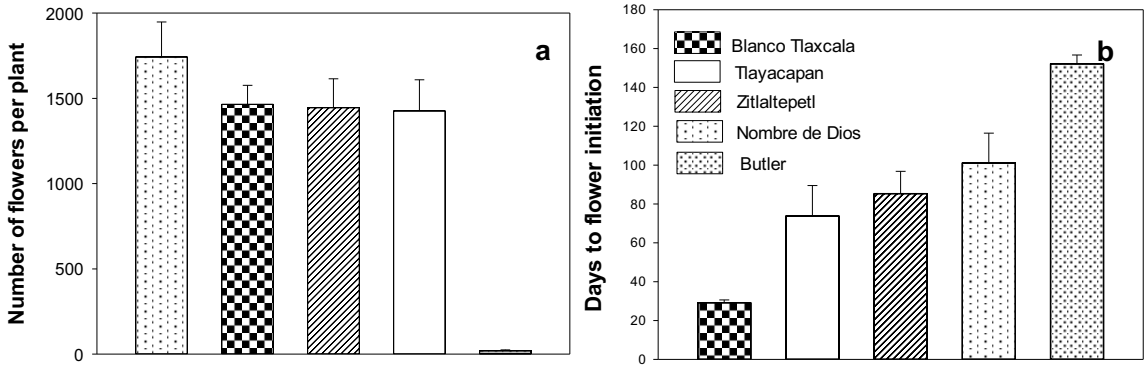


Figure 1. Number of flowers per plant (a) and flowering initiation time (b) in three landraces and two varieties of *Phaseolus coccineus* L.

REFERENCES

Búrquez, A. and J. Sarukhán. 1980. Biología floral de poblaciones silvestres de *Phaseolus coccineus* L. I. Relaciones planta-polinizador. Boletín de la Sociedad Botánica de México 39:5-25.

Búrquez, A. and J. Sarukhán. 1984. Biología floral de poblaciones silvestres de *Phaseolus coccineus* L. II. Sistemas reproductivos. Boletín de la Sociedad Botánica de México 46:3-12.

Escalante, A.M., Coello, G., Eguiarte, L.E., Piñero, D. 1994. Genetic structure and mating systems in wild and cultivated populations of *Phaseolus coccineus* and *P. vulgaris* (Fabaceae). Am. J. of Bot. 81:1096-1103.

González, A.M., De Ron, A.M., Lores, M., Santalla, M. 2014. Effect of the inbreeding depression in progeny fitness of runner bean (*Phaseolus coccineus* L.) and its implications for breeding. Euphytica 200:413-428.

Statistical Analysis System (SAS Institute). 2012. SAS version 9.3 for Windows. SAS Institute Inc. Cary, N.C., USA.

BREEDING RUNNER BEAN FOR CANNING INDUSTRY IN EASTERN AFRICA

Kimani. P. M.[1], Serah Nyawira Njau[1], Mwangi Njiru[2] and Steve Omondi[3]

[1]Department of Plant Science and Crop Protection, University of Nairobi, P.O Box 29053-00625 Nairobi, Kenya; [2] Operations Department, Trufood Ltd, P.O Box P.O Box 41521-00100 Nairobi; [3]Agronomy Department, Njoro Canning Ltd, P.O Box 7076-20110, Nakuru, Kenya

INTRODUCTION

Canning bean industry in eastern Africa is constrained by inadequate supply of grain because available landraces are low yielding and susceptible to diseases. Demand for canned bean products exceeds supply due to preference for fast cooking off-shelf products, high cost of cooking fuel and population growth. Little has been done to develop improved runner bean varieties for the canning industry due to lack of research investment for breeding runner bean and the limited local capacity to assess canning characteristics during variety development. Several new, grain-type runner bean lines with superior agronomic traits were recently developed at the University of Nairobi (Kimani et al, 2016). However, their potential for use by the processing industry is not known.

The objective of this study was to evaluate the canning quality of the new breeding lines and identify those that meet industry canning criteria and acceptability to consumers.

MATERIALS AND METHODS

Four populations were developed from crosses between a long day variety (White Emergo) and four short-day grain type runner bean landraces at Kabete Field Station. The F_2 populations were advanced to F_5 generation as population bulks at Ol Jorok, Subukia and Kabete experimental sites in Kenya. Starting F_5, single plants were selected and advanced through single pod descent method. About 139 $F_{6.8}$ lines were evaluated for grain yield, disease resistance and adaptation to short day conditions at Kabete (1860 masl) and Ol Jorok (2300 masl) in 2012 and 2013 (Kimani et al, 2016). In 2014, 43 advanced lines, three checks and one reference variety (TruFood RB) grown at Ol-Joro-Orok and Kabete Field Station were evaluated for canning quality. The beans were soaked, blanched, canned in brine and incubated for seven days, and subsequently evaluated for canning quality attributes including hydration coefficient (HC), washed drained weight (WDWT) and percentage washed drained weight (PWDWT) following procedures described by Khanal et al (2014) and van der Mwerwe et al (2006). Physical properties (size, shape, uniformity) and visual appearance properties (splits, clumping and brine clarity) were determined subjectively using seven point hedonic scale. Each trial was replicated three times. Data was analysed using Genstat software (v15).

RESULTS AND DISCUSSION

Results showed significant ($P<0.05$) differences in all traits evaluated (Table 1). The best performers at Kabete were KAB-RB13-327-92/1, KAB-RB13-326-207/1B and KAB-RB13-326-207/1B. However, the best performers at Ol-Joro-Orok were KAB-RB13-471-117/1, SUB-OL-RB13-275-248/3 and KAB-RB13-310-161/5 suggesting a genotype x environment interaction for canning traits. More than 30 lines met or exceeded industry canning standards for common bean (Khanal et al, 2014; van der Mwerwe *et al*., 2006). The texture of 31 lines was better than 72N standard for beans, and 16 better than industry reference check variety used in Kenya (Table 1). We used standards for canning beans as a reference because equivalent standards for dry grain runner bean were are not available. These lines are probably the first grain type runner bean lines bred in east, central, southern and west Africa.

Utilization of the new lines can increase productivity, incomes of smallholder farmers, and ensure regular supply and diverse, high quality value added products for the processing industry and consumers. Data generated provides a baseline for further breeding of runner beans for the canning industry

Table 1. Summary of canning quality characteristics of the new runner bean lines developed and evaluated at two locations in Kenya.

Trait	Range/Locations		Industry reference variety	Lines ≥ reference
	Kabete	**Ol-Joro-Orok**		
Soakability 16 h (%)	55-155	70-125	115	27
Water uptake after blanching (%)	95-174	111-187	131	31
Hydration coefficient	1.7-2.7	2.1-2.6	2.3	25
Fresh wt to yield solids (g)	111-115	110-112	110	31
Brine pH	5.8-6.0	6.0-6.1	5.7	31
WDWT (g)	253-298	256-312	269	31
PWDWT (%)	59-69	57-73	58	31
Texture (N)	93.7-194	101.9-219	125	16 (31)
Seed size (score)	3-7	5-7	4	30
Uniformity (score)	1-3	1-3	2	25
Seed shape (score)	1-4	1-5	2	22
Splits (score)	3-7	4-7	6	27
Clumping (score)	5-7	3-7	6	31

WDWT- washed drained weight; PWDT-percent washed drained weight

REFERENCES

Kimani, P.M., M.M. Mulanya, S. Njau and R.D. Narla. 2016. Breeding runner bean for grain yield, disease resistance and short-day adaptation in Eastern Africa. BIC 59: 207-208.

Khanal, R., Balasubramaniann et al. 2014. Genotypic associations of parameters commonly used to predict canning quality of dry bean. Crop Science 15:2564-2573

Van der Mwerwe et al. 2006. Comparison of canning quality of small white beans by small-scale and industrial method. J. Sci. Food Agric 86: 1046-1056

BREEDING SECOND GENERATION BIOFORTIFIED BEAN VARIETIES IN EASTERN AFRICA

Kimani. P. M., Ahmed Warsame and Serah Nyawira Njau

Department of Plant Science and Crop Protection,
University of Nairobi, P.O Box 29053-00625 Nairobi, Kenya

INTRODUCTION

Micronutrient malnutrition, also known as 'hidden hunger', is a serious health challenge facing vast sectors of Africa's population particularly resource-poor women and children. Worldwide, it affects over 2 billion people. Fe, Zn and Vitamin A deficiencies are the most frequent. For example, iron deficiency anaemia (IDA) prevalence varies from 6 to 88% in east and southern Africa, and 7 to 58% in west and central Africa (IDRC, 2001). Development and utilization of drought tolerant, biofort varieties is probably the most effective, sustainable and potentially long-lasting strategy for reducing micronutrient deficiencies and coping with frequent droughts.

Our objective was to develop second generation biofortified bean varieties combining drought tolerance, multiple disease resistance and higher grain iron and zinc concentration than the first generation varieties currently grown by farmers in east, central and west Africa. Targets for the first generation varieties was a grain concentration of 70ppm Fe and 30ppm Zn, which were fully achieved (Kimani et al, 2016). The target for the second generation varieties is 90ppm Fe and 40ppm Zn.

MATERIALS AND METHODS

Five genotypes with high micronutrient concentration but lacking in preferred agronomic traits and susceptible to major diseases, were crossed with multi-parent male gametes to generate 47 F_2 populations segregating for mineral density, resistance to biotic and abiotic stress factors, marketable grain types and yield potential at Kabete Field Station between 2009 and 2010. F_2's were advanced to F_4 as population bulks. In 2010, the F_4 families were evaluated for grain iron and zinc concentration using the wet digestion method (perchloric acid and nitric acid). $F_{4.5}$ progenies were subsequently evaluated under drought stress and no-stress conditions at two locations. Selected drought tolerant families were evaluated for disease resistance and agronomic traits at Kabete and Thika in 2011 and 2012. In 2013, promising lines were evaluated for adaptability and grain yield at five locations representing major bean production environments. The second screening for mineral density was conducted in 2014 for lines with good agronomic potential and resistance to diseases. Data was analyzed using Genstat version 15.

RESULTS AND DISCUSSION

Results showed significant ($P<0.01$) variation for mineral density, drought tolerance, disease resistance, growth habit, grain type and maturity among the populations and their progenies. Iron concentration varied from 30 to 130 ppm (Fig 1). Zinc concentration varied from 10 to 60 ppm (Fig 2). Outstanding lines were selected from BF08-01, BF08-07, BF08-16 and BF08-36 populations (Fig 1). Eighty-four lines had 50% higher yield under stress and no-stress conditions compared with the parental lines, suggesting transgressive segregation.

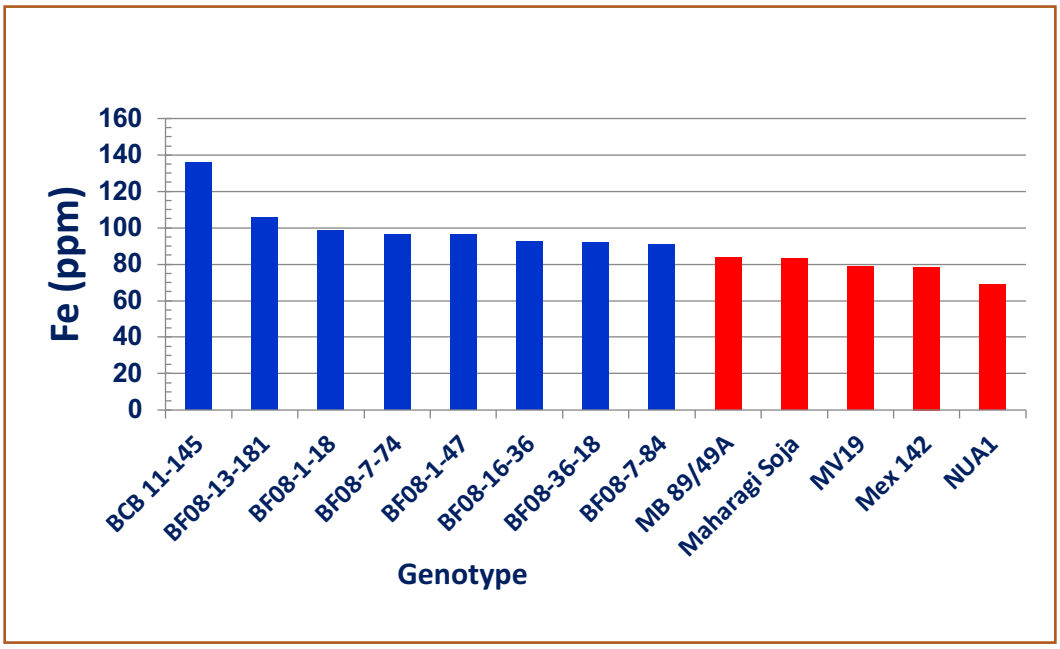


Figure 1. Fe concentration of F_8 lines grown at Thika and Kabete. MB 89/49, Maharagi Soja, MV 19, Mex 142 and NUA1 were used as checks.

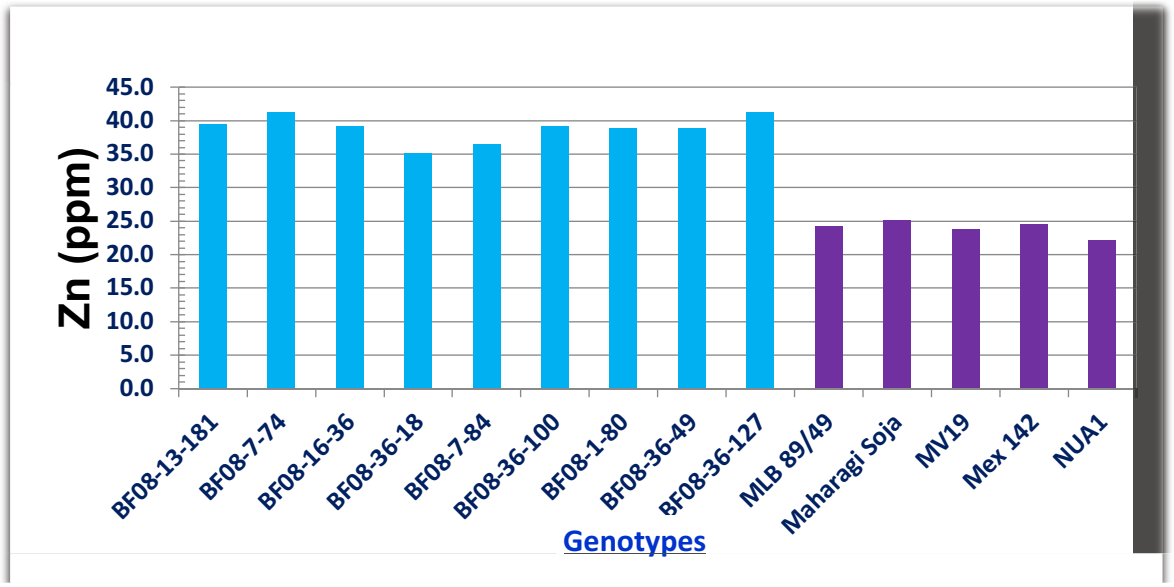


Figure 2. Grain zinc concentration of second generation F_8 biofort lines.
Results indicated second generation lines had up to 85.7% more Fe, 100 % more Zn, 50% higher grain yield and were more drought and disease tolerant than the first generation lines.

REFERENCES

Kimani, P.M., P. Mamiro, M. Ugen, and A. Musoni. 2016. Development and release of new biofortified bean varieties in Eastern Africa. *Bean Improvement Cooperative,* Vol.59:147-148.

ASSESSMENT OF TOFU PREPARED WITH NAVY AND GARBANZO BEANS

Hui Wang, Muhammad Siddiq, and Mark A. Uebersax

Dept of Food Science & Human Nutrition, Michigan State University,
East Lansing, MI 48824

INTRODUCTION: Tofu, the gelled curd derived from soybean with a consistency of soft white cheese, has traditionally been prepared from coagulation and pressing of soymilk (Zhao et al. 2016). This non-fermented, semi solid gelled product has wide acceptability, especially, among health-conscious consumers. Tofu preparation could potentially utilize alternate legumes, i.e., Navy beans (*Phaseolus vulgaris*) and Garbanzo beans (*Cicer arietinum*) to partially replace soybean in the formulation used for tofu production. In recent years, non-traditional legumes, particularly Garbanzo beans have received increasing acceptance by the consumers (e.g., hummus has become a popular side dish due to its no-fat and high-protein composition). Although Navy beans are processed as canned products, they offer a potential to be used in tofu formulation due mainly to neutral color and flavor. Therefore, the objective of this study was to investigate the feasibly of using Navy and Garbanzo beans in soy-based tofu and to assess its yield and quality attributes.

METHODS: ***Tofu preparation:*** Tofu was prepared with whole beans on a lab-scale using the method described by Wang (1993). Soybeans in tofu formulation were replaced at 25% to 75% levels with either Navy or Garbanzo beans. Briefly, the beans were soaked overnight in water at ambient temperature, followed by blending. The soluble fraction thus obtained was heated at 99.3 °C for 5 minutes and then let to cool to 70 °C. Curd was formed by adding and stirring 0.85%t $CaSO_4$ salt, followed by pressing in a cheese-cloth lined cylindrical tube.

Tofu Quality Evaluation: Following quality attributes of experimental tofu were assessed.
Proximate analysis: Moisture, protein, fat, ash, and fiber content was determined using standard AOAC methods. Yield: Tofu yield was expressed as grams of gelled tofu produced from the initial formula weight of the dry seeds. Texture: The instrumental texture of tofu was determined using a TMS-90 shear press equipped with a parallel plate compression cell TPA–1 (Food Technology Corporation, Maryland, USA). The results were expressed as peak firmness (hardness value) in Newton (N-force). Sensory Evaluation: A trained 6-member panel consisting of males and females of oriental origin, familiar with tofu quality characteristics, performed the sensory evaluation of flavor and overall acceptability using a 7-point hedonic scale.

RESULTS AND DISCUSSION

The proximate analysis of soybean (control) and Navy and Garbanzo bean tofu is shown in **Table 1**. Generally, adding Navy or Garbanzo beans to tofu did not affect proximate composition, with the exception of noted increase in fiber content (0.73–0.93% versus 0.69% in control).

Table 1: Proximate analysis (%) of different tofu formulations

Formulation	Moisture	Protein	Fat	Ash	Fiber
100SB	81.8	51.6	22.0	15.0	0.69
75SB:25NB	79.7	48.2	19.7	16.5	0.75
50SB:50NB	78.2	47.5	20.1	18.7	0.81
75SB:25GB	77.7	51.5	21.7	17.1	0.81
50SB:50GB	79.8	52.1	20.5	18.3	0.87
25SB:75GB	78.5	47.9	17.2	25.2	0.93

SB=soybean, NB=Navy Bean, GB=Garbanzo bean

The yield and texture data of different tofu formulations are presented in **Fig. 1**. Yield and texture values for both non-soy substituted tofu were consistently reduced with increased levels of substitution. Preliminary studies demonstrated 100% formulation of the alternate beans did not produce tofu. At the 75% level of substitution navy beans formed limited gel and eliminated from tests, whereas, garbanzo beans formed a gel with substantial yield reduction.

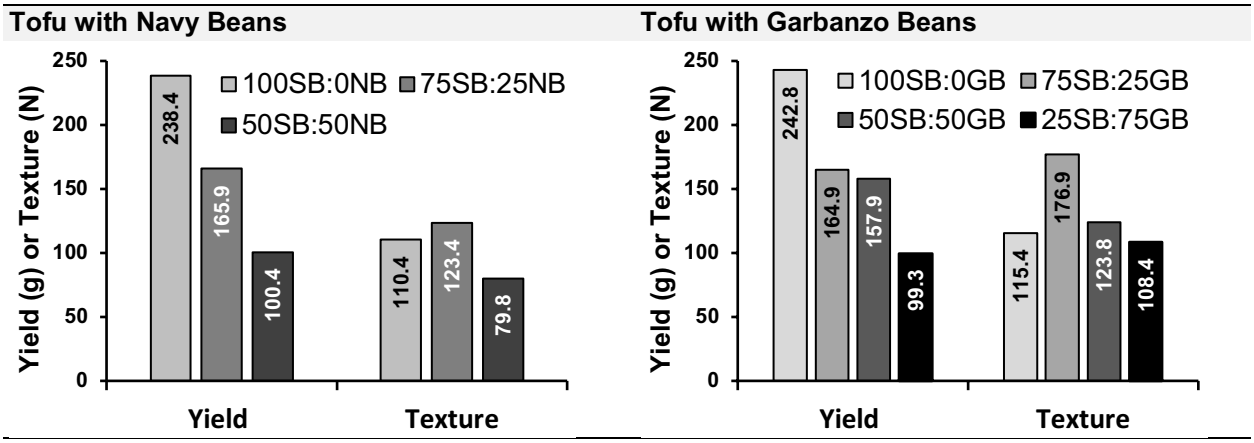


Fig. 1: Yield and texture of tofu with soybean (SB), Navy bean (NB) and Garbanzo bean (GB)

The sensory evaluation results of different tofu formulations are presented in **Fig. 2**. Both flavor and acceptability scores were reduced significantly with the addition of Navy or Garbanzo beans at all levels. The only exception was the flavor scores (≥ 3.0) for 25 and 50% Garbanzo bean tofu.

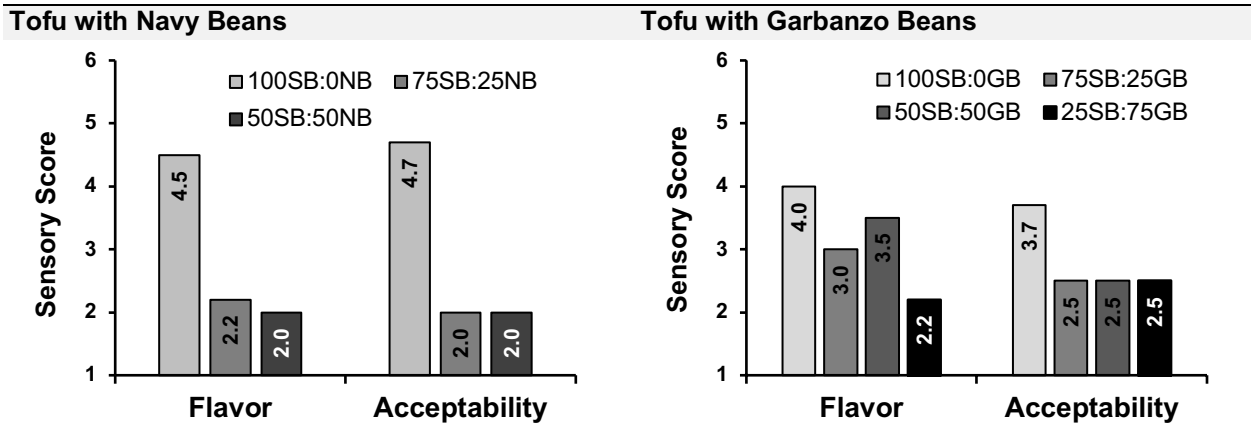


Fig. 2: Sensory scores of tofu with soybean (SB), Navy bean (NB) and Garbanzo bean (GB)

CONCLUSIONS

The yield and firmness of tofu produced were significantly reduced with substituted Navy bean or Garbanzo beans. These quality attributes are strong determinants of consumer acceptance and commercial feasibility of tofu. Results demonstrate that Navy or Garbanzo beans generally lack the ability to form high yielding firm gels required for high quality tofu products.

REFERENCES

Wang H. (1993). Evaluation of tofu from selected soy cultivars and breeding lines. *MS Thesis*, Department of Food Science & Nutrition, Michigan State University, East Lansing, 121p.

Zhao H, Li W, Qin F, Chen J. (2016). Calcium sulfate-induced soya bean protein tofu-type gels: influence of denaturation and particle size. *Intl J Food Sci & Technol* 51:731-741.

GENOMIC SELECTION ON A PANEL OF ELITE ANDEAN BREEDING LINES

D. Ariza-Suarez[1], J.F. De la Hoz[1], E. Macea[1], V.M. Mayor[1], B. Raatz[1]

[1]International Center for Tropical Agriculture CIAT

INTRODUCTION: Common bean breeding in the tropics has benefited from the use of marker assisted selection to accelerate the breeding cycle. It uses individual markers previously identified to be significantly associated with targeted QTLs. However, some limitations arise when identifying and quantifying a large number of minor effect loci governing quantitative traits through standard QTL analysis and marker tagging. Genomic selection (GS) offers a promising approach for breeding by using many markers as predictors of the performance of individuals in a population[1,2]. This alternative strategy does not provide direct information about the underlying loci and genes that control quantitative traits, but instead offers a valuable selection criterion to be implemented in a breeding scheme. Different GS studies have been recently reported in plant breeding, targeting complex traits for many species with interesting results. In this study we want to assess the prediction accuracy of different genomic selection models in a collection of Andean bean breeding lines from CIAT.

MATERIALS AND METHODS: We used field data from a panel of elite Andean breeding lines in this study. This panel has been evaluated in 6 different yield trials carried out between 2013 and 2016 in Palmira (Colombia), managed under irrigated (irr) or drought (drt) conditions. We fitted a mixed linear model for alpha lattice designs to calculate BLUPs, broad-sense heritability and phenotypic correlations. We genotyped this panel using the *ApeKI* GBS protocol and used the NGSEP[3] pipeline for SNP calling on the sequencing data, which produced in a set of 7000 high quality SNP markers. We used the BGLR package[4] to fit the GS models BayesB, RKHS (Bayesian reproducing kernel Hilbert spaces) and GBLUP (Genomic BLUP). Prediction accuracy is given as a Pearson correlation between the observed and predicted phenotypic value. To assess prediction ability we made 100 random population partitions, with 70% training - 30% validation sets on the BLUPs from each trial to predict its own performance.

RESULTS AND DISCUSSION: Mean yield on the field trials ranged from 761 kg/ha (Pal15C_drt) to 2290 kg/ha (Pal15C_irr). Phenotypic correlations among yield trials ranged from 0.30 to 0.56, which shows the degree of phenotypic stability across seasonal conditions for this location. The calculated broad-sense heritability was between 0.35 and 0.73 (figure 1). We observed significant phenotypic variability across trials that may be related with different environmental effects.

We obtained prediction accuracies with median values ranging from 0.31 to 0.55 (figure 1). The GS models tested vary in their assumptions when treating the variance of complex traits. This may generate a differential response based on the genetic architecture of the trait, marker density and the span of linkage disequilibrium[2]. In general, all GS models tested showed the same behavior on prediction ability, but the RKHS approach had slightly higher correlations for yield.

We observed a positive correlation between prediction accuracies and the broad-sense heritability calculated from the mixed linear model, which shows that GS models are properly capturing the genetic variance provided by the BLUPs. Thus, the variation in prediction accuracies between seasons corresponds to the same remaining environmental effects, and suggests that additional improvements in their modeling could enhance prediction accuracies as well.

This work tested different GS models to predict yield on a highly structured panel of Andean bean breeding lines across different trials on a single location with seasonal variability. Yield is a complex trait that is affected by numerous biotic and abiotic factors. The phenotypic response is controlled by many individual loci with variable small effect on each genotype, and difficult to predict. Despite that, we obtained promising prediction accuracies for yield, but further work should be performed to improve this strategy for implementation in a breeding scheme.

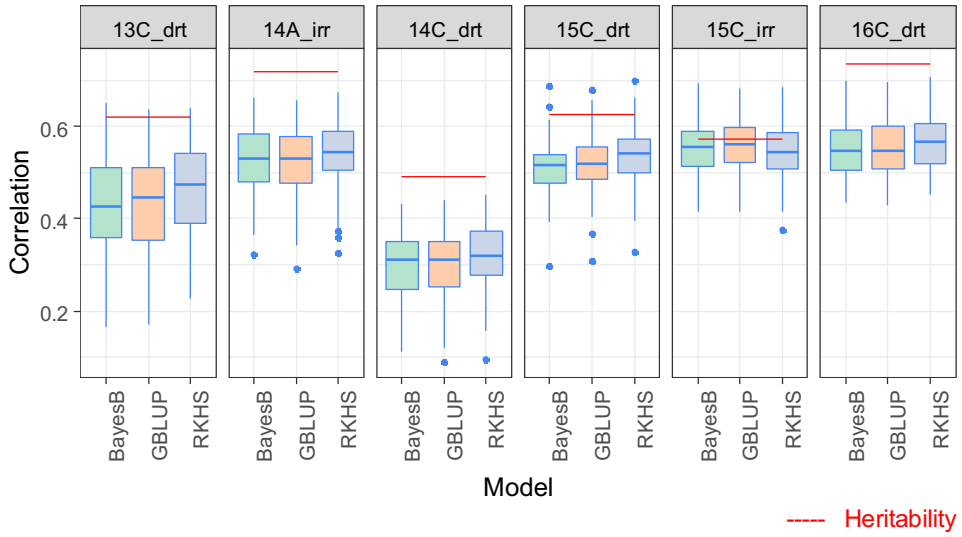


Figure 1. Prediction accuracy of yield (kg/ha) using the genomic selection models BayesB (green), GBLUP (orange) and RKHS (blue) on six different yield trials managed under drought (drt) or irrigated (irr) conditions. Every box shows the distribution of correlations for 100 random population partitions. Red lines shows the calculated broad-sense heritability.

CONCLUSIONS We assessed the prediction accuracy of different GS models on yield with a highly structured Andean bean breeding panel. We obtained correlation values ranging from 0.13 to 0.55, with the RKHS model having slightly higher values compared to other models. Future work involves the assessment of prediction ability with variable numbers of markers and genotypes. This work is part of the first approaches to implement GS into the Bean breeding strategy at CIAT.

REFERENCES

1. Crossa, J. *et al.* Genomic Selection in Plant Breeding: Methods, Models, and Perspectives. *Trends Plant Sci.* **22,** 961–975 (2017).
2. Desta, Z. A. & Ortiz, R. Genomic selection: Genome-wide prediction in plant improvement. *Trends Plant Sci.* **19,** 592–601 (2014).
3. Duitama, J. *et al.* An integrated framework for discovery and genotyping of genomic variants from high-throughput sequencing experiments. *Nucleic Acids Res.* **42,** (2014).
4. Pérez, P. & De Los Campos, G. Genome-wide regression and prediction with the BGLR statistical package. *Genetics* **198,** 483–495 (2014).

EARLY SELECTION OF COMMON BEAN PROGENIES THROUGH REML/BLUP MIXED MODELING

Reche DL[1], Vidigal Filho PS[1*], Elias JCF[1], Moda-Cirino V[2], Valentini G[1], Martins VSR[1], and Gonçalves-Vidigal MC[1]

[1]Departamento de Agronomia, Universidade Estadual de Maringá, Maringá, PR, Brazil;
[2]Instituto Agronômico do Paraná, PR, Brazil; *E-mail: vidigalfilhop@gmail.com

INTRODUCTION

In Brazil 80% of the producers do not use improved common bean cultivars and, therefore, obtaining progenies that present high productive capacity in the shortest time is essential. According to Resende and Duarte (2007), and Henderson (1984) the parameters: Adjusted heritability of the genotype mean assuming complete progeny survival ($\hat{h}^2_{mc}$) and Accuracy of genotype selection (Acclon), indicate the possibility of success with the early selection in progenies. Therefore, the objective of this study was to evaluate the agronomic performance of common bean $F_{2:3}$ families through analyzes of mixed models REML/BLUP.

MATERIAL AND METHODS

A total of 94 $F_{2:3}$ families from the LP97-28 line × IPR Uirapuru cross, and the cultivars SCS Guará, Ouro Negro, Pérola and IPR Juriti (controls), were evaluated at the Centro de Treinamento em Irrigação (CTI), Universidade Estadual de Maringá, PR, Brazil. Each experimental plot consisted of a line of 1.0 m length, spaced 0.50 m, containing 12 plants of each family. The experimental design was a 10 × 10 Lattice, with three replications. The families were evaluated for the characteristics: height plant (HP), number of days to flowering (DF), number of days to maturity (DM), number of pods per plant (NPP), number of seeds per pod (NSP), 100 seeds weight (SW) and grain productivity (PROD). Statistical analyzes, applying a selection pressure of 20%, were performed using the Model 17, Selegen REML/BLUP software, and the significance of the effects of the models was estimated through the Deviance Analysis (Resende 2016).

RESULTS AND DISCUSSION

The results indicated significant effects for HP, DM, SW and PROD evidencing the existence of genetic variability among the evaluated families.

Table 1. Estimative of the variance components for the characteristics height plant (HP), days to flowering (DF), days to maturity (DM), pods per plant (NPP), seeds per pod (NSP), 100 seeds weight (SW) and grain productivity (PROD) in common bean progenies

	Components of Variance REML						
Effects	HP	DF	DM	NPP	NSP	SW	PROD
$\hat{\sigma}^2_g$	23.79	0.35	1.98	0.47	0.01	2.48	194,958.66
$\hat{\sigma}^2_b$	19.42	0.80	0.61	0.64	0.00	0.92	48,589.48
$\hat{\sigma}^2_e$	53.35	10.83	4.48	2.56	0.77	1.04	124,972.68
$\hat{\sigma}^2_f$	96.56	11.98	7.07	3.67	0.77	4.44	368,520.82
$\hat{h}^2_{mc}$	0.47	0.06	0.47	0.27	0.02	0.83	0.76
Acclon	0.69	0.25	0.69	0.52	0.13	0.91	0.87
μ	61.37	44.43	84.87	9.81	5.85	19.00	2,525.41

$\hat{\sigma}^2_g$ - genotypic variance; $\hat{\sigma}^2_b$- environmental variance between blocks; $\hat{\sigma}^2_e$ - residual variance; $\hat{\sigma}^2_f$ - individual phenotypic variance; $\hat{h}^2_{mc}$ - Adjusted heritability of the genotype mean assuming complete progeny survival; Acclon - accuracy of genotype selection; μ - general average of the experiment.

The mean heritability estimates ($\hat{h}^2_{mc}$) ranged from mean values (0.47 for DM and HP) to high values (0.76 for PROD and 0.83 for SW). Similar results were obtained by Gonçalves-Vidigal et al. (2008) for DM and WS. Likewise, the accuracy values (Acclon, Table 1) were moderate for DM and HP (0.69) and high for SW (0.91) and PROD (0.87). Based on in the experimental average of grain productivity (PROD) observed (2,525.41 kg ha^{-1}), a selection pressure of 20% provided a gain of 69.45%. Among the best 20 progenies selected (Table 2), 13 were superior to the best control (SCS Guará), evidencing the potential use of early selection by REML/BLUP for the selection of superior families.

Table 2. Performance of the best 20 families and controls for the prediction parameters of genotypic effects ($u + g$), gain and new average for the grain productivity (Kg ha^{-1})

Order	Families	g	u + g	Gain	New average
1	323	1,214.45	3,843.81	1,214.45	3,843.81
2	289	1,122.09	3,751.45	1,168.27	3,797.63
3	349	966.12	3,595.48	1,100.89	3,730.24
4	333	927.09	3,556.45	1,057.44	3,686.80
5	344	896.66	3,526.02	1,025.28	3,654.64
6	365	842.64	3,472.00	994.84	3,624.20
7	418	808.31	3,437.67	968.19	3,597.55
8	398	794.48	3,423.83	946.48	3,575.84
9	358	763.04	3,392.40	926.10	3,555.46
10	409	747.44	3,376.80	908.23	3,537.59
11	305	734.73	3,364.09	892.46	3,521.82
12	391	645.21	3,274.57	871.85	3,501.21
13	239	576.05	3,205.41	849.10	3,478.46
15	265	563.68	3,193.04	811.65	3,441.01
16	335	560.54	3,189.90	795.96	3,425.32
17	274	560.32	3,189.68	782.10	3,411.46
18	345	560.28	3,189.64	769.77	3,399.13
19	272	524.08	3,153.44	756.84	3,386.20
20	399	505.39	3,134.75	744.27	3,373.63
21	431	484.75	3,114.11	731.91	3,361.27
14	SCS Guará	572.80	3,202.15	829.36	3,458.72
31	IPR Uirapuru	271.81	2,901.17	613.09	3,242.45
41	Ouro Negro	127.05	2,756.41	509.64	3,139.00
62	Pérola	-173.65	2,455.71	320.96	2,950.32
79	IPR Juriti	-436.31	2,193.50	190.63	2,819.99
98	LP97-28	-911.34	1,718.02	21.59	2,650.95

ACKNOWLEDGEMENTS: This research was financially supported by CNPq and Capes**.**

REFERENCES

Gonçalves-Vidigal et al.(2008). Euphytica 164: 551-560
Henderson (1984). University of Guelph 462p
Resende (2016). Crop Breeding and Applied Biotechnology 16:330-339
Resende and Duarte (2007). Pesquisa Agropecuária Tropical 37:182-194.

A HIGH-DENSITY SNP CONSENSUS MAP REVEALS SEGREGATION DISTORTION REGIONS IN COMMON BEAN

Valentini G[1], Gonçalves-Vidigal MC[1]*, Vidigal Filho PS[1], Gilio TAS[1], Hurtado-Gonzales OP[2], Song Q[2], Pastor-Corrales MA[2], Sousa LL[3], Gepts P[4]

[1]Departamento de Agronomia, Universidade Estadual de Maringá, Maringá, PR, Brazil; [2]Soybean Genomics Improvement Laboratory, USDA-ARS, Beltsville, MD, USA; [3]Universidade Federal de Goiás; [4]Department of Plant Sciences, University of California, Davis, CA, USA; *E-mail: mcgvidigal@uem.br

INTRODUCTION

Molecular markers, such as single-nucleotide polymorphisms (SNPs), are now widely used for construction of linkage maps in all major crops. A genetic linkage map of common bean based entirely on SNPs is useful for the identification of genes/QTL controlling traits and marker-assisted selection. High-density common bean linkage maps containing thousands of SNP markers were constructed by Song et al. (2015). These SNPs were identified by aligning millions of reads to the Andean reference sequence (G19833) of common bean (Schmutz et al. 2014). Common bean germplasm originates from two gene pools, Andean and Mesoamerican, which are characterized by distinctive morphological traits, seed storage proteins, and DNA sequences. Distorted segregation of markers in the progeny derived from the Andean and Mesoamerican crosses has been frequently observed. In order to verify the presence of segregation distortion regions (SDRs) in common bean, a high-density consensus map was constructed using single nucleotide polymorphism (SNP) markers, by merging a genetic map developed among 110 recombinant-inbred lines (RILs) population developed from the California Dark Red Kidney (CDRK) × Yolano (CY population) cross. However, their progenies have not been characterized for the presence of segregation distortion regions (SDRs), where alleles at a locus deviate from the 1:1 Mendelian expectation. The present study provides an initial high-density SNP map using California Dark Red Kidney (CDRK) × Yolano (CY population) recombinant-inbred line population and the identification of segregation distortion in this population.

MATERIAL AND METHODS

A total of 110 recombinant inbred lines (RILs) from the mapping population California Dark Red Kidney (Andean) × Yolano (Mesoamerican) were used in this study. The development of the CY population was described by Johnson and Gepts (1999; 2002). Seeds of each line were propagated at the Núcleo de Pesquisa Aplicada à Agricultura (Nupagri) of the Universidade Estadual de Maringá, Maringá, Paraná, Brazil. Leaf tissue was harvested from single plantlets and high-quality genomic DNA for SNP genotyping was isolated with the PureLink® Genomic DNA Mini Kit, following the manufacturer's instructions. The 110 CY RILs and parents were screened with 5,398 SNP markers of the Illumina BeadChip BARCBEAN6K_3 (Song et al. 2015) following the Infinium HD Assay Ultra Protocol (Illumina, Inc. San Diego, CA). The fluorescence intensity obtained by the BeadChip was visualized using Illumina BeadArray Reader. SNP alleles were automatically called using Illumina GenomeStudio V2011.1 (Illumina, San Diego, CA). Allele calls were visually inspected and errors in allele calling were corrected manually. Molecular analysis was performed at the USDA-ARS Soybean Genomic and Improvement Laboratory in Beltsville, MD. For linkage map construction, a pre-selection of SNPs was carried out in Microsoft Excel. Based on the expected allelic ratio of 1:1 of all polymorphic SNPs in the RIL population, the segregation distortion of each locus was investigated according to the P-values of Chi-square

($P < 0.05$). The data from Chi-square probability was graphically represented in a Manhattan plots using the QQman package of R software (Turner 2014). Distances between markers were calculated using Kosambi's mapping function with default parameters in JoinMap 4.1.

RESULTS AND DISCUSSION

The CDRK × Yolano RIL population was genotyped with 5,398 SNP markers. After elimination of SNPs with high frequency of missing data or loci with a minor allele frequency of 30%, 3,277 SNPs out of 5,398 SNP markers segregated in this population and the final map spanned 936 cM in genetic distance with an average interval of 0.3 cM. This map covered 512.15 Mbp of the genome, based on the proportion of the physical distance (bp) between the first and last SNPs mapped to each chromosome. The average recombination rate (Kb/cM), measured by the physical (Kb) and genetic (cM) position of the last marker mapped in each chromosome, was 565.7 Kb, similar to an earlier observation around the *Phaseolin* locus (Llaca and Gepts 1996). Genetic position of most SNPs in the linkage maps was consistent with the physical positions along each chromosome of the *Phaseolus vulgaris* genome assembly V1.0. The number of markers that showed distorted segregation in the CY population was 665 (20%), from a total of 3.335 SNPs polymorphic between parents CDRK and Yolano (Figure 1). Most of the distorted markers (451) were located on chromosomes Pv01 (CDRK alleles in excess) and Pv10 (Yolano alleles in excess), which accounted for 13.5% of markers. This information will facilitate understanding of the genetic architecture of common bean and identification of SNPs tightly linked to QTL for many important traits in common bean.

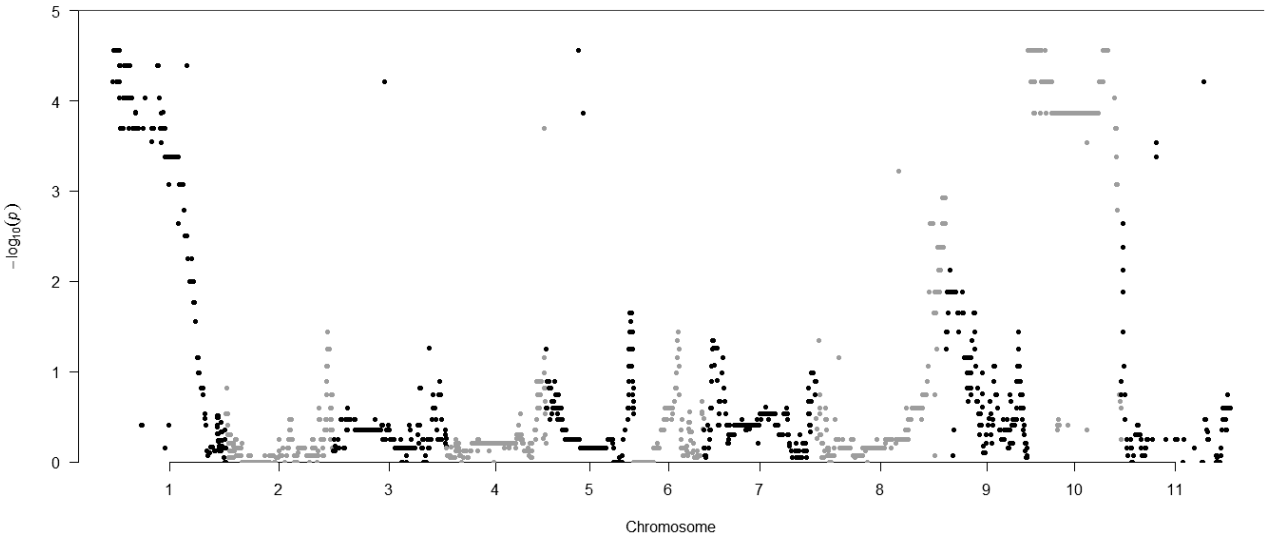


Figure 1. Manhattan plot of segregation distortion probability of 3,335 SNP markers in the RIL population CDRK × Yolano.

REFERENCES

Johnson and Gepts (1999). Euphytica 106: 45-56
Johnson and Gepts (2002). Euphytica 125: 69-79
Llaca and Gepts (1996). Genome 39: 722-729
Schmutz et al. (2014). Nature Genetics 46: 707-713
Song et al. (2015). G3 11: 2285-2290
Turner (2014). BiorXiv: biorXiv DOI: 10.1101/005165.

REACTION TO THE ANGULAR LEAF SPOT OF CREOLE BEANS ACCESSIONS CULTIVATED IN NORTH OF MINAS GERAIS, BRAZIL

Sanglard, D. A*.; Machado, M. A. M.; Batista, F. E. R.; Barbosa, M. H. C. and Nunes, C. F.

Biotechnology Lab, Agriculture Science Institute (ICA),
Universidade Federal de Minas Gerais (UFMG), 39.404-547, Montes Claros, MG, Brazil.
*E-mail: demerson.ufmg@gmail.com

Belonging to the *Leguminosae* family, the bean (*Phaseolus vulgaris* L.) is one of the most widespread crops in the world, being grown in almost all tropical and subtropical countries. In addition, it is possible to produce grains up to three times a year in many regions. One of the factors that limit bean productivity and performance is diseases. Among the diseases of major importance, it is worth highlighting the angular bean leaf spot, incited by *Pseudocercospora griseola* (Sacc.) Crous & U. Braun. Given the great progress of plant breeding and the loss of some characteristics of interest, the rescue and maintenance of the genetic diversity of the creole species are considered environmental services and are strategic basis for an agriculture that seeks sustainability (Penteado, 2010). Thus, the aim of this study was to verify the reaction of creole beans accessions to *P. griseola*.

It was evaluated, for the reaction to the angular leaf spot, 24 creole beans accessions, which are commonly cultivated by family farmers of the North of Minas Gerais, Brazil. The seeds come from properties of the following cities Montes Claros, Porteirinha, Pai Pedro, Juramento, Lagoa dos Patos, Itacambira, Bocaiúva and São Francisco. It was used eight races (isolates) known to be virulent and highly aggressive, all kept in the mycological bank of the Biotechnology Laboratory (ICA/UFMG). The inoculums of each race (isolate) were obtained by multiplying the fungus in Petri dishes containing a mix of distilled water, tomato sauce, agar and calcium carbonate ($CaCO_3$) (Sanglard et al., 2009). Using scissors previously sterilized in alcohol, one of the primary leaves of the plants tested was cut ten days after sowing, when it presented approximately 2/3 of its complete development. Subsequently, it was immersed in an inoculum suspension, in concentration of 2.0×10^4 conidia/mL. Then, the leaves were transferred individually to Petri dishes (90 x 15 mm) containing filter paper previously moistened with 3.0 mL of distilled water. These Petri dishes were then incubated in a climatic chamber (B.O.D.) at 20°C, in periods of 12 hours of light per day, with a light incidence of approximately 28 μmoles/$m^2 \cdot s$. Every three days from the inoculation, 1.5 mL of distilled water was added to the filter paper of each petri dish until the disease symptoms were verified (Ragagnin et al., 2005). Thus, it was possible to maintain humidity high inside the plates. The severity of the disease was visually evaluated at 15, 18 and 21 days after inoculation, using a nine-degree severity scale proposed by Pastor-Corrales & Jara (1995). Twelve leaves of each accession, collected from individual plants, were inoculated. Reactions in the leaves with grades from one to three were considered resistant; intermediate resistance with grades between three and six; and susceptible with grades between six and nine.

The results confirm the great difficulty in the identification of genotypes with wide resistance to the angular leaf spot disease. Among the 24 Creole accesses tested, 21 were intermediate and / or susceptible to at least five races (isolates) (Table 1). However, the accesses 'Andu Indiano', 'Moiacho' and 'Rosa Precoce' were resistant to seven, seven and six of the eight races tested, respectively. In addition, they were also those that showed significantly lower averages of severity. Orozco & Araya (2005) have already demonstrated that, in places where Mesoamerican-type beans

are exclusively cultivated, selection on the prevailing populations of *P. griseola* occurs, which leads to the enhancement of correspondent races to the host gene pool.

This behavior type confirms that, in the North of Minas Gerais, there are predominance of races of the Mesoamerican group, as it happens in the rest of the country. With the observed data, that is, considering the presence of few genotypes with resistance to a large number of races, it can be concluded that only the use of complete vertical resistance is not the best strategy to be used to control this disease. Therefore, it is suggested that genotypes with the highest degrees of horizontal and partial resistance should also be used as source of resistance. Another relevant possibility to control the angular leaf spot would be the use of multilines cultivars (Sartorato, 2006). The results obtained in this study are relevant for genetic plant breeding programs, since they direct the researches of resistance sources identification.

Table 1. Reaction of 24 creole bean accessions, collected in Minas Gerais, to races (isolates) of *P. griseola*.

Races (Isolates) of *P. griseola*	**Accessions of creole beans cultivated in North of Minas Gerais, Brazil**																						
	Andu Indiano	Andu Manteiga	Andu Preto Precoce	Azuk Claro	Azuk Roxo	Bonina	Branco Mineiro	Campeiro Preto	Chamego Preto	Catador Grande	Jalo Precoce	Mangalô	Meguito	Moiacho	Ouro Velho	Parto Mineiro	Radiante Preto	Rajado Precoce	Rosa Precoce	Rosinha Mineiro	Roxim Mineiro	Sangue de Boi	Santo Antônio	Tocão
63.63	1.4	5.8	2.5	7.0	8.0	6.6	6.5	8.3	6.8	6.0	6.9	7.2	2.6	2.2	5.2	6.0	5.6	7.7	7.1	4.0	2.5	5.2	3.8	4.2
63.47	2.5	7.3	5.9	3.6	2.8	4.0	3.8	5.4	6.1	5.0	3.5	4.1	4.0	2.5	5.2	4.9	6.2	5.5	2.5	1.4	7.9	5.0	4.6	3.6
63.23	1.0	2.5	3.6	3.4	7.1	5.5	4.0	5.0	3.2	2.1	7.1	6.0	4.6	1.5	4.1	5.9	7.2	7.1	1.7	3.7	3.7	4.0	5.8	7.0
63.7	2.7	3.8	2.9	6.7	5.4	3.5	1.9	4.1	1.9	3.8	3.4	5.5	7.4	3.0	7.6	4.0	4.0	3.6	2.6	7.0	5.6	3.8	6.0	4.2
47.39	5.5	6.1	6.0	2.5	3.1	7.9	3.9	3.1	4.0	3.0	1.7	5.1	3.3	1.7	5.1	3.8	4.2	3.6	2.6	1.1	4.0	3.1	6.2	3.1
31.7	2.1	5.0	6.0	7.0	4.8	5.5	7.2	5.0	7.1	2.2	3.4	3.1	4.6	1.9	6.0	5.5	3.5	8.6	4.2	5.0	4.3	6.0	2.7	3.5
31.4	2.2	4.5	2.2	3.8	8.5	4.7	3.3	5.6	4.5	4.3	3.7	5.2	4.2	2.9	7.1	3.1	4.4	3.6	1.6	4.5	4.3	3.5	5.1	1.5
23.23	3.0	7.0	3.9	4.4	2.0	4.6	2.9	5.7	3.3	8.0	3.8	2.5	3.6	3.1	4.5	5.5	8.4	4.5	2.4	4.1	8.0	3.5	4.9	5.1

Severity averages calculated by the evaluation of 12 leaves of each access. Reactions in the leaves with grades from one to three were considered resistant; intermediate resistance with grades between three and six; and susceptible with grades between six and nine.

References

Orozco, S; Araya, CM. Variabilidade patogênica de *Phaeoisariopsis griseola* na Costa Rica. Fitopatologia Brasileira, 30(6):589-593, 2005.

Pastor-Corrales, MA; Jara, CE. La evolución de *Phaeoisariopsis griseola* com el frijol común em América Latina. Fitopatologia Colombiana, 19(1):15-24, 1995.

Penteado, SR. Manual prático de agricultura orgânica – Fundamentos e Técnicas. Campinas. SP. Edição do autor, 2ª edição, 2010, 232 p.

Ragagnin,VA et al. A new inoculation procedure to evaluate angular leaf spot disease in bean plants (*Phaseolus vulgaris* L.) for breeding purposes Annual Report of the Bean Improvement Cooperative, East Lansing, MI, USA, 48:90-91, 2005.

Sanglard, DA et al. An efficient protocol for isolation, sporulation and maintenance of *Pseudocercospora griseola*. Annual Report of the Bean Improvement Cooperative, 52:62-63, 2009.

Sartorato, A. Novas fontes de resistência do feijoeiro comum à mancha-angular. Fitopatologia Brasileira, 31:92-194, 2006.

GENETIC VARIABILITY AMONG ISOLATES OF *Colletotrichum lindemuthianum*

Coêlho M[1], Gonçalves-Vidigal MC[1*], Vidigal Filho PS[1], Valentini G[1], Lacanallo GF[1], and Martiniano-Souza MC[2]

[1]Dep. Agronomia, Universidade Estadual de Maringá, Maringá, PR, Brazil, 87020-900
[*]E-mail: mcgvidigal@uem.br; [2]Instituto Agronômico de Pernambuco, PE, Brazil.

INTRODUCTION

Anthracnose, caused by the fungal pathogen *Colletotrichum lindemuthianum* (Sacc & Magnus) Briosi & Cavara, is one of the most devastating diseases in commom bean (*Phaseolus vulgaris* L.), especially when environmental conditions are favorable for the development of the disease (Singh and Schwartz 2010). The incidence of several races of *C. lindemuthianum* is due to its wide intra-and inter-pathogenic variability observed among the races. The investigation of internal transcribed spacer (ITS) regions of fungal ribosomal DNA (rDNA) is one of the most successful strategies to identify genetic variability of this pathogen. These regions are highly variable in comparison to other genic regions of rDNA and specific fungal sequences can be amplified via PCR with taxon-specific primers. The objective of this work was to characterize isolates of *C. lindemuthianum* from several states of Brazil, through sequencing of ITS regions.

MATERIAL AND METHODS

This research was conducted at the Núcleo de Pesquisa Aplicada a Agricultura (Nupagri) of the Universidade Estadual de Maringá. Forty-one isolates of *C. lindemuthianum* from the states of Mato Grosso, Paraná and Santa Catarina were used for genotyping of the ITS regions. Small fungal spore discs were transferred from petri dishes containing BDA medium to erlenmeyers, containing liquid medium BD (Dextrose potato). Genomic DNA extraction was performed following the modified SDS protocol (Cárdenas et al. 2012). The amplification of the ITS1, 5.8S and ITS2 regions of the rDNA was performed using the ITS1F (5' CTTGGTCATTTAGAGGAAGTAA 3') and ITS4 (5' TCCTCCGCTTATTGATATGC 3') primers. The amplification products of *C. lindemuthianum* isolates were purified using the PureLink PCR Purification Kit (Invitrogen®), sequenced and analyzed with the BioEdit Sequence Alignment Editor Program version 7.2.5.

RESULTS AND DISCUSSION

The PCR products obtained by the amplification of the ITS1, 5.8S and ITS2 rDNA regions of the 41 *C. lindemuthianum* isolates showed fragments of 600 base pairs (bp) (Figure 1). The ITS regions showed high genetic variability, with the presence of 163 SNPs (single nucleotide polymorphisms), wherein the ITS1 region showed 74 SNPs and the ITS2 region exhibited 89 SNPs. Similar results were obtained by Balardin et al. (1999), when analyzing sequences of 14 isolates of *C. lindemuthianum* detected highly variability in the ITS2 region. The genetic variability within and among the studied isolates revealed the importance of ITS regions to determine the diversity among *C. lindemuthianum*.

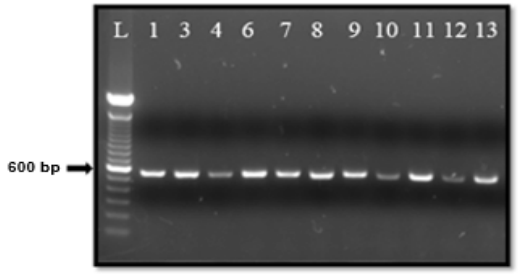


Figure 1. Agarose gel electrophoresis at 2% showing the DNA band of approximately 600 bp. L: 100 bp DNA ladder (Invitrogen). Numbers 1 to 13 correspond to the isolates of *C. lindemuthianum*.

Variability of *C. lindemuthianum* isolates was inferred from the sequence comprising ITS 1 and ITS 2 regions. The analysis of sequences of 41 isolates of this study were compared with the sequence of race 17. According to Figure 2, the ITS1 region reveals the presence of SNPs at positions 77 and 165 bp for the isolates 6, 7, 8, 11, 12, 16, 18, 23, 25, 27, 29, 36 and 40, occurring a substitution mutation of **G** to **T** and **G** to **A**, respectively. In turn, the isolates 2, 5, 7, 9, 15, 18, 19, 22, 23, 25, 26, 29, 31, 34, 36, 37, 38 and 40, presented an insertion of **C**, an insertion of **A** and a substitution of **C** to **A** at positions 157, 158 and 159 bp, respectively. In the ITS2 region at position 487 bp in the isolates 2, 7, 9, 15, 18, 19, 23, 29, 31, 33, 34, 36, 37, 38 and 40 the insertion of **A** occurred. Also at the position 515 bp, was observed a substitution of nucleotide **C** to **A** in the isolates 6, 7, 8, 11, 16, 18, 23, 25, 27, 29, 34, 36 and 40.

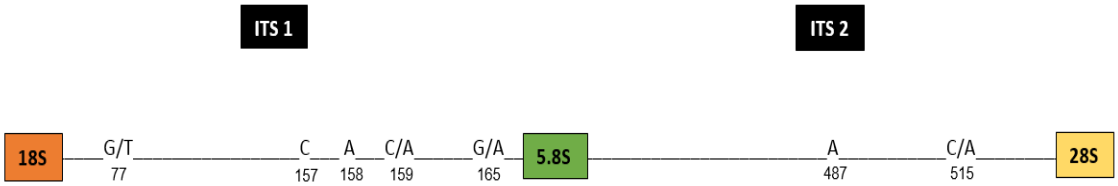


Figure 2. Identification of SNPs that occurred most frequently among the 41 isolates of *C. lindemuthianum*.

Among the 41 isolates studied, a total of 28 presented SNPs, the isolates with the highest amount of SNPs were 10, 22, 37 and 41, with 59, 36, 19, 38 SNPs, respectively. The association of the *C. lindemuthianum* pathogen with common bean cultivars from different origins and the exposure to different environmental conditions may result in broad pathogenic variability (Talamini et al. 2006). The results obtained in this study revealed the existence of high genetic variability among the isolates of *C. lindemuthianum* through analysis of ITS regions.

ACKNOWLEDGEMENTS

This research was financially supported by CNPQ and Capes, Brazil. M. Coêlho and G. Valentini were supported by a scholarship from Capes. M.C.M. Souza was supported by a scholarship from CNPq. M.C. Gonçalves-Vidigal and P.S. Vidigal Filho are grateful for Grants from CNPq and are sponsored by CNPq.

REFERENCES

Balardin et al. (1999). Mycological Research 103:841-848
Cárdenas et al. (2012). Plant Disease 96:456
Singh and Schwartz (2010).Crop Science 50:2199-2223
Talamini et al. (2006). Fitopatologia Brasileira 31:545-550.

ANTIBACTERIAL ACTIVITY OF CLOVE EXTRACTS ON COMMON BACTERIAL BLIGHT ISOLATED FROM PINTO SALTILLO DRY BEAN CULTIVAR

Erika Cecilia Gamero-Posada[1], Ixchel Abby Ortiz-Sánchez[1], Sonia Valdez-Ortega[1], Rigoberto Rosales-Serna[2], Mirka Maily Acevedo-Romero[1*]

[1]Instituto Tecnológico del Valle del Guadiana, km 22.5 Carretera Durango-México, Villa Montemorelos, Dgo., México. C. P. 34371. [2]INIFAP-Durango, km 4.5 Carretera Durango-El Mezquital, Durango, Dgo. México. C. P. 34170. *e-mail: mirkamar.itvg@hotmail.com

INTRODUCTION. Common beans (*Phaseolus vulgaris*) is an important staple food crop in Durango, México. Several plant pathogens, including common bacterial blight (*Xanthomonas axonopodis* pv. *phaseoli*), cause significant reductions on yield and seed quality in common beans. Disease control is difficult due to reduced genetic diversity for genetic breeding in common beans and the lack of effective and eco-friendly bactericides. Alternative organic and low cost bactericides are required for disease control in order to reduce pathogen effects on common beans production. Clove (*Syzygium aromaticum*) present antibacterial properties against food borne pathogens such as *Pseudomonas aeruginosa* (Pandey and Singh, 2011) and *Xanthomonas campestris* pv. *citri* (Chudasama and Thaker, 2012). The objective was to evaluate *in vitro* antibacterial activity of different clove extracts for CBB control under controlled laboratory conditions.

MATERIAL AND METHODS. Seeds were harvested in Canatlán, México, in plants of Pinto Saltillo common bean cultivar showing typical CBB symptoms under field conditions. 10 g of seed samples were sealed at the hilar region using cyanoacrylate based glue. Seeds were disinfected using a 2 % sodium hypochlorite solution, shaken constantly during 3 min, and then rinsed with distilled water. Dried seeds were milled with 90 mL phosphate buffer in a domestic blender during 1 min to allow the leakage of microorganisms from infected seed tissues. Bacteria were isolated by dilution plate technique in YCA culture medium. CBB isolates were confirmed considering colonial macroscopy, microscopic morphology, metabolism and growth curve by spectrophotometric method at 660 nm. Clove extract was obtained by steam distillation using 250 g of crushed dry seeds combined with 1 L of water. The essential oil was separated from the aqueous phase and the remaining infusion was also included in the test. The tea extract was prepared by boiling 10 g of clove seeds in 250 mL of water for 10 min and then paper-filtered. Clove extracts were evaluated in agar plates with different inoculum levels for CBB. Agar disk-diffusion method was used including filter paper discs impregnated in each treatment and then placed over the bacteria inoculated surface. A Completely Randomized experimental design with a 6 x 5 factorial arrangement and three replicates was used. Factor A included five type of clove extracts: essential oil, diluted oil (1:1 oil-water), aqueous extract, infusion and tea. A negative control was also included for comparison. Factor B consisted in five dilution levels of bacterial inoculum (0.1, 0.01, 0.001, 0.0001, 0.00001) starting from a colony and using phosphate buffer as diluent, in order to simulate different levels of disease infestation. The inhibition halo around the filter paper disc was calculated using its diameter (mm) after 48 h incubation period at 30 °C. Data were subjected to an analysis of variance (ANOVA) and when significant difference among means were found comparisons were carried out using Tukey's test ($p \leq 0.05$). Both procedures were performed using the InfoStat® package.

RESULTS AND DISCUSSION. Significant differences were observed ($p \leq 0.05$) among treatments and the clove essential oil was the most effective treatment for CBB control (Figure 1), followed by clove aqueous extract. The presence of CBB was confirmed according to typical colony macroscopy, cellular morphology and the metabolic attributes described by Gamero *et al*., (2014). The growth curve showed that the exponential stage begins between 0 and 2 h of incubation, concluding at 14 h period when the stationary phase was observed. The mean generation time was 4.6 h, indicating rapid pathogen proliferation under controlled culture conditions and doubling its population during this time period. High bactericide efficacy of clove extracts (oil and aqueous extract) was observed under low bacterial concentrations. Results suggest that clove extract shows a potential use in preventive control of bacterial dry bean diseases such as CBB. Similar results were observed for bacterial inhibition using 10 to 100 % clove oil concentrations (Lucas *et al*., 2012). Further studies are required in order to determine chemical components of clove extracts, individual effects and potential use for CBB control under field conditions.

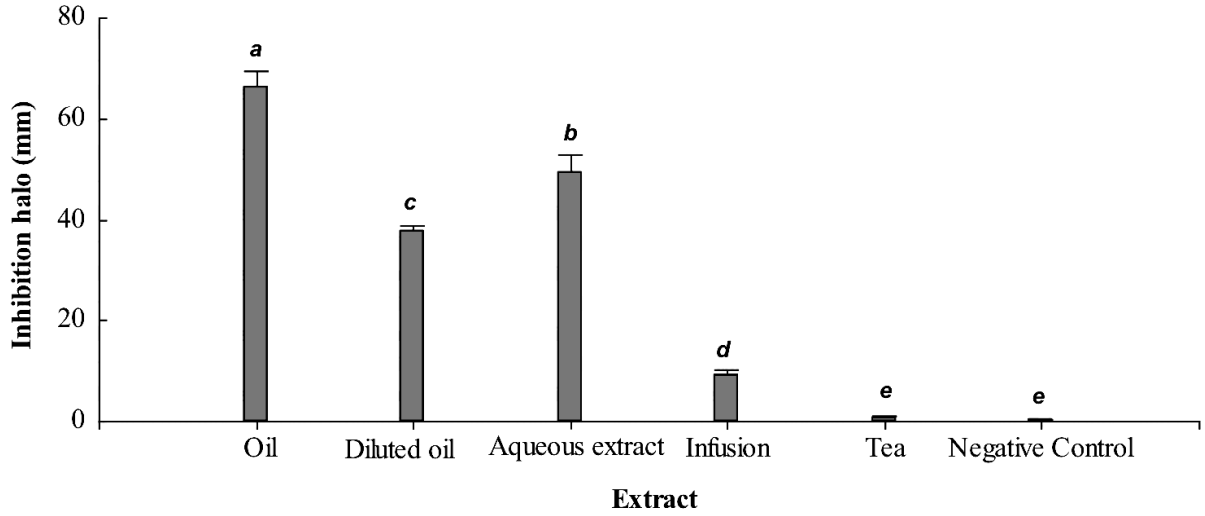


Figure 1. Efficacy of clove extracts on *in vitro* growth inhibition of common bacterial blight (CBB) isolated from Pinto Saltillo dry bean cultivar. Bars represent halo mean diameter ± standard error. [a-e]letters represent statistically significant differences among treatmets.

CONCLUSIONS. Clove essential oil and the aqueous extract showed significant inhibitory activity on *in vitro* growth of common bacterial blight CBB.

REFERENCES

Bao, L-M., Eerdunbayaer; A. Nozaki, E. Takahashi, K. Okamoto, H. Ito and T. Hatano. 2012. Hydrolysable tannins isolated from *Syzygium aromaticum*: Structure of a new *C*-Glucosidic ellagitannin and spectral features of tannins with a tergalloyl group. Heterocycles 85(2): 365-381.

Chudasama, K. S. and V. S. Thaker. 2012. Screening of potential antimicrobial compounds against *Xanthomonas campestris* from 100 essential oils of aromatic plants used in India: an ecofriendly approach. Archives of Phytopathology and Plant Protection 45(7): 783-795.

Gamero P., E. C., R. Rosales S., M. L. Reyes G., M. Amezcua R. y K. A. Silva R. 2014. Evaluación de la calidad sanitaria y nutricia del frijol producido en Vicente Guerrero, Durango. Semana Internacional de Agronomía. Durango. México. pp. 272-278.

Lucas, G. C., E. Alves, R. Borges P., F. J. Perina e R. M. de Sousa. 2012. Antibacterial activity of essential oils on *Xanthomonas vesicatoria* and control of bacterial spot in tomato. Pesquisa Agropecuária Brasileira 47(3): 351-359.

ANTIBACTERIAL ACTIVITY OF OREGANO EXTRACTS ON HALO BLIGHT ISOLATED FROM COMMON BEANS GROWN IN DURANGO, MÉXICO

Ixchel Abby Ortiz-Sánchez[1], Sonia Valdez-Ortega[1]*, Erika Cecilia Gamero-Posada[1], Rigoberto Rosales-Serna[2] and Mirka Maily Acevedo-Romero[1]

[1]Instituto Tecnológico del Valle del Guadiana, km 22.5 Carretera Durango-México, Villa Montemorelos, Dgo., México. C. P. 24371. [2]INIFAP-Durango, km 4.5 Carretera Durango-El Mezquital, Durango, Dgo., México. C. P. 34170. *Corresponding author: sonia_valdez@hotmail.com.

INTRODUCTION. Common beans (*Phaseolus vulgaris*) have economic, social and nutritive importance due to its daily consume for the majority of farmers in Durango, México. Several abiotic and biotic factors reduce common bean yield and seed quality. In México plant diseases such as halo blight (HB) (*Pseudomonas syringae* pv. *phaseolicola*) cause leaf damage, seed abortion and yield reduction in common beans (Prudencio *et al*., 2008). In Durango organic products need to be identified in order to increment the use of safe and eco-friendly bactericides in common bean production. Mexican oregano (*Lippia graveolens*) is an endemic and abundant plant species in Durango. Antibacterial properties in oregano extracts has been reported (Hernández *et al*., 2009) and is considered as a potential and low cost bactericide for halo blight control in common beans. The objective was to evaluate antibacterial activity of oregano extracts on *in vitro* growth of halo blight isolated from common beans grown in Durango.

MATERIALS AND METHODS. Oregano extracts were obtained by the steam distillation technique using 50 g of Mexican oregano plants (leaves) combined with 750 mL of water in a distillation flask. Readings were taken for the inhibition level of *in vitro* HB growth for each oregano extract. A Completely Randomized Design (CRD) with factorial arrangement (2 x 6) and three replicates were used. Factor A included two different infestation levels obtained by bacterial inoculum dilutions (1:10 and 1:100) using Butterfield´s phosphate buffer as diluent. Factor B consisted in five oregano extracts: oregano oil (O), diluted oil (DO) (oil:water 1:1), aqueous extract (AE), infusion (IN) and tea (TE). A negative control (NC) was also included and consisted in a sterile filter paper disk. Paper diffusion technique (antibiogram) was used (Gamero *et al*., 2014) and a 2 cm in diameter sterile filter paper disk containing three drops of each oregano extract was placed in the center of each Petri dish newly planted. The Petri dishes were incubated at 28 °C during a 48 h time period. Antibacterial activity was found by measuring diameter (mm) of the clear zones of halo inhibition formed around the discs using a Vernier caliper (resolution of 0.01 mm). Data were subjected to an analysis of variance (ANOVA) and when significant difference among means were found comparisons were carried out using Tukey's test ($p \leq 0.05$). Both procedures were performed using the InfoStat® software.

RESULTS AND DISCUSSION. Significant differences were observed ($p \leq 0.05$) among treatments and the oregano essential oil was the most effective treatment for HB control (Figure 1), followed by oregano aqueous extract. The bacteria growth inhibition was associated with the oregano oil containing more than 25 volatile compounds (mainly carvacrol and thymol) showing antimicrobial and antifungal activity (Cueto, 2010). Extract dilutions with reduced content for each compound showed lower efficacy on halo inhibition compared to the essential oil.

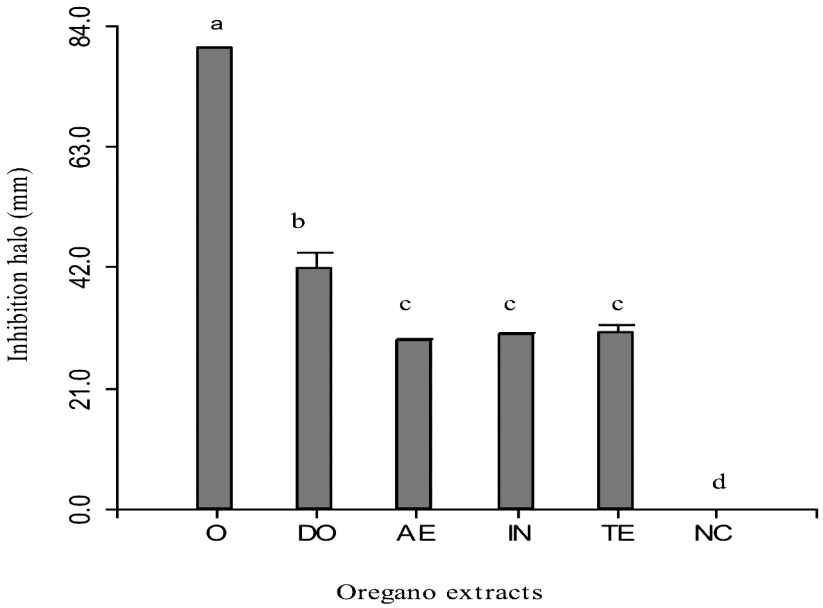


Figure 1. Antibacterial activity of oregano extracts on *in vitro* growth inhibition of common bacterial blight (CBB) isolated from Pinto Saltillo dry bean cultivar. Bars represent halo mean diameter ± standard error. [a-d]letters represent statistically significant differences among treatmets. Oil(O), diluted oil (DO), aqueous extract (AE), infusión (IN), tea (TE) and Negative control (NC).

CONCLUSIONS. Outstanding results were obtained for the *in vitro* test using oregano essential oil which inhibited by 83 % the halo growth for *P. syringae* pv. *syringae*. Additional studies are necessary in order to encourage the use of oregano essential oil as a bactericide for organic and sustainable production of common beans.

REFERENCES

Cueto, W. M. C. 2010. Determinación del efecto inhibitorio del aceite escencial y diferentes extractos de orégano (*Lippia berlandieri Schauer*) sobre el crecimiento de *Fusarium oxysporum* tanto *in vitro* como en plántula de tomate. Tesis de Doctorado. Universidad Autónoma de Nuevo León. México. Páginas.

Gamero P., E., E. Merlín B., H. Sarmiento L. y L. Tapia B. 2014. Evaluación *in vitro* del efecto de productos químicos y orgánicos sobre el crecimiento de *Erwinia amylovora* causante del tizón de fuego en el manzano (*Malus sylvestris*). Memoria de la XXVI Semana Internacional de Agronomía FAZ-UJED. pp. 1018-1022.

Hernández, T., M. Canales, J. G. Ávila, A. M. García, S. Meraz, J. Caballero, R. Lira. 2009. Composition and antibacterial activity of essential oil of Lippia graveolens H.B. K. (Verbenaceae). Boletin Latinoamericano y del Caribe de Plantas Medicinales y Aromáticas 8(4): 295-300.

Prudencio S., J. M., R. Navarrete M., J. Navarrete M. y J. A. Acosta G. 2008. Dinámica de los tizones común y de halo del frijol en el Valle de México. Agricultura Técnica en México 34(2): 201-212.

HERITABILITY OF COMMON BACTERIAL BLIGHT RESISTANCE IN COMMON BEAN

Wilson Nkhata[1], Deidré Fourie[2], Rob Melis[1]

1 School of Agricultural, Earth and Environmental Sciences, University of KwaZulu-Natal, X301, Scottville 3209, Pietermaritzburg, South Africa. 2 Agricultural Research Council-Grain Crops Institute, Private Bag X1251, Potchefstroom, 2520, South Africa

INTRODUCTION

Common bacterial blight (CBB), caused by *Xanthomonas axonopodis* pv *phaseoli* (Xap) (Smith) Vauterin, Hoste, Kosters & Swings and its fuscans variant, *X. fuscans* sbsp. *fuscans* (Xff), is an important seed-borne disease of common beans (*Phaseolus vulgaris* L.) worldwide (Singh and Schwartz, 2010). Although several sources of resistance to CBB have been identified, the disease remains a major challenge in common bean production. The study was initiated using crosses between two navy bean cultivars, Teebus-RCR 2/Teebus-RR 1, and two red speckled sugar bean cultivars, RS 7/Tygerberg) to investigate the mode of gene action governing inheritance of resistance to CBB and to estimate the heritability in CBB resistance.

MATERIALS AND METHODS

Teebus-RR 1 and Tygerberg (CBB susceptible) Teebus-RCR 2 and RS 7 (CBB susceptible) were used in crosses and a total of six generations were developed for each cross (F_1, RF_1 F_2, RF_2, BCP_1 and BCP_2). Reaction to CBB was evaluated in a greenhouse at ARC-GCI, Potchefstroom during the 2015/2016 growing season. Mean day- and night temperatures of the greenhouse were 24 ^{0}C and 18 ^{0}C, respectively. The trial was planted on 30th March 2016 and laid out in a randomised complete block design (RCBD) with three replications. The six generations of each cross, including the parents) were raised in sterilised polythene plastics pots of 30 cm in diameter. The number of plants varied depending on the generation. A mixture of two local isolates (Xf260 and Xf410) were used for inoculation. The inoculum was prepared by suspending 48 to 72-h-old cultures in distilled water. The inoculum density was 10^8 CFU/ml. The multiple needle technique (Andrus, 1948) was used to inoculate fully first expanded trifoliate leaves. Plants were rated for CBB reaction 14 DAI using a 1-9 scale with 1 being highly resistant and 9 highly susceptible. Gene effects were estimated using the method of Cavalli, 1952.

RESULTS AND DISCUSSION

Data for the reaction to CBB (Table 1) for Teebus-RCR 2/Teebus-RR 1 fitted the six-parameter model and all gene effects were significant at ($P<0.001$). The reaction to CBB for RS 7/Tygerberg did not fit the six-parameter model or the simple additive dominant model. Additive gene effects were non-significant. Both dominance and additive gene effects were significant at $P< 0.05$ and $P<0.01$ respectively. In both crosses dominance gene effect predominate additive gene effects, this implied that early generation selection could not be effective. The failure of the model to fit simple additive model in both crosses is an indication that CBB is quantitatively inherited. The significance additive x additive epistatic effects implies that resistance can be fixed and exploited. Cytoplasmic effects (Table 2) were significant in both crosses. However, data did not fit both the simple and six parameter model for maternal gene effect. The non-additive maternal gene effects were absent an indication that maternal effect is governed by fixable gene effects only. Narrow sense heritability in both crosses was moderate in both crosses (36% for Teebus-RCR 2/Teebus-RR 1 and 59% for RS 7/Tygerberg).

Table 1. Estimates of gene effects of reaction to CBB for the two common bean crosses

Gene effects	Teebus-RCR 2/Teebus-RR 1	RS 7/Tygerberg
	Estimates ±SE	Estimates±SE
[m]	-35.76±6.25***	5.371±0.407***
[a]	8.73±2.43***	2.31±2.32
[d]	21.69±1.04***	3.19±1.57*
[aa]	28.6±4.69***	9.65±4.78*
[ad]	-31.08±5.75***	-15.01±5.49**
[dd]	-9.252±0.822***	-
Epistasis type	Duplicate	-

Significant at: * P<0.05; ** P<0.01; *** P<0.001. SE = Standard error, [m] = Mid parent, [a] = Additive gene action, [d] = Dominance gene action, [aa] = Additive x additive gene action, [ad] = Additive x dominant gene action, [dd] = Dominant x dominant gene action

Table 2. Estimates of maternal gene effects of reaction to CBB for the two crosses of dry bean

Maternal gene effects	Teebus-RCR 2/Teebus-RR 1	RS 7 / TYGERBERG
	Estimates ±SE	Estimates ±SE
[m]	-58.22± 3.98***	5.76±0.17***
[c]	0.27±0.11**	0.79±0.06***
[a_m]	-3.53±0.32***	0.41±0.10***
[aa_m]	27.19±16.73***	-

Significant at: * P<0.05; ** P<0.01; *** P<0.001. SE = Standard error, [m] = Mid parent, [c] = Cytoplasmic effect, [a_m] = Additive maternal gene action, [aa_m] = Additive x additive maternal gene action.

CONCLUSION

Common bacterial blight is moderately inherited and governed by additive dominance and epistasis effects. Maternal effects were found to be of importance in common bacterial blight resistance. The overall implications of these findings in CBB resistance breeding programme is that it will affect the selection strategy to be deployed and also the choice of female parent. Backcross breeding, recombinant breeding, delayed selection and choosing a resistant parent as a female parent would register positive result in CBB resistance breeding programme.

REFERENCES

Andrus C (1948) A method of testing beans for resistance to bacterial blights. Phytopathology 38 : 757-759.

Cavalli L (1952) An analysis of linkage in quantitative inheritance. In: Reeve C and Waddington C (eds), Quantitative inheritance. HMSO, London. p. 135-144.

Singh SP and Schwartz HF (2010) Breeding common bean for resistance to diseases: a review. Crop Science 50 : 2199-2223.

IDENTIFICATION OF COMMON BEAN LINES USEFUL FOR THE DIFFERENTIATION OF *XANTHOMONAS AXONOPODIS* PV. *PHASEOLI* RACES

Mildred Zapata

Department of Agroenvironmental Sciences, University of Puerto Rico,
Mayagüez, Puerto Rico, 00680

In the attempt to identify lines of common bean useful for the differentiation of *Xanthomonas axonopodis* pv. *phaseoli* races (Xap), four improved common bean breeding lines were selected (W-BB-11, W-BB-20, W-BB-52 and W-BB-56) for leaf inoculation of Xap strains under greenhouse conditions. Bacterial strains were represented by two strains from Puerto Rico (484 and 3353) and two from Central America (1930 from Nicaragua and 1934 from Costa Rica). A complete block design with 3 replicates was used. The inoculation and the evaluation methods has been described previously by Zapata.

The bean release W-BB-11 showed susceptibility to the four Xap strains beginning at 14 days after inoculation in contrast with the other releases **(Table 1).** Puerto Rico strains showed similar virulence on the releases. In contrast, W-BB-56 showed an immune response to all the Xap strains. W-BB-52 showed immunity to the Puerto Rico strains and very low susceptibility to Xap 1930 from Nicaragua.

At 21 days, a systemic infection was shown by W-BB-11 for strain Xap 484. WBB-52 showed an immune response to Xap 484 and Xap 3353 **(Table 2).** The difference between the Puerto Rico Xap strains was related to the severity of infection. Xap 484 induced systemic infection on W-BB-11and Xap 3353 was not systemic at that period of time. W-BB- 56 showed good resistance to all the strains.

At 35 days, W-BB-11 was systemically invaded by all the Xap strains **(Table 3).** W-BB-52 showed resistance to the strains from Puerto Rico but was systemically invaded by Xap 1934. W-BB-20 was systemically invaded by Xap 3353 and Xap 1930 but showed resistance to Xap 484. W-BB-56 showed resistance to all the Xap strains except to Xap 1930.

In general, W-BB-52 was most useful to differentiate reactions among the Xap strains. It was resistant to the Xap strains from Puerto Rico and susceptible to the isolates from Nicaragua and Costa Rica. W-BB 20 was useful to differentiate between the strains from Puerto Rico. It was resistant to Xap 484 and highly susceptible to Xap 3353. The Xap strains from Nicaragua and Costa Rica were more virulent than the ones from Puerto Rico. All the bean releases tested except for W-BB-11 can be used for the differentiation of the four Xap strains evaluated in this study.

Table 1. Evaluation of bean breeding lines to leaf inoculation with four Xap strains from the Caribbean and Central América at 14 days after inoculation.

Identity	***Bacterial strains***			
Bean breeding Line	*Puerto Rico 484*	*Puerto Rico 3353*	*Nicaragua 1930*	*Costa Rica 1934*
W-BB-52	1.00a	1.00a	2,33a	1.33a
W-BB-20	1.17a	1.00a	2.17a	2.00b
W-BB-56	1.00a	1.33a	1.67a	1.00a
W-BB-11	4.00b	3.00b	3.00a	3.00c

$p \leq 0.05$

Table 2. Evaluation of bean breeding lines to leaf inoculation with four Xap strains from the Caribbean and Central América at 21 days after inoculation.

Identity	***Bacterial strains***			
Bean breeding Line	*Puerto Rico 484*	*Puerto Rico 3353*	*Nicaragua 1930*	*Costa Rica 1934*
W-BB-52	1.00a	1.00a	2.33a	3.00b
W-BB-20	1.17a	1.67a	2.33a	3.00b
W-BB-56	1.00a	1.33a	1.67a	1.00a
W-BB-11	9.00b	5.00b	6.00b	3.17b

$p \leq 0.05$

Table 3. Evaluation of bean breeding lines to leaf inoculation with four Xap strains from the Caribbean and Central América at 35 days after inoculation.

Identity	***Bacterial strains***			
Bean breeding line	*Puerto Rico 484*	*Puerto Rico 3353*	*Nicaragua 1930*	*Costa Rica 1934*
W -BB-52	1.00a	1.00a	6.00b	9.50c
W-BB-20	1.17a	9.00c	9.00c	3.00b
W-BB-56	1.00a	1.67b	3.50a	1.33a
W-BB-11	9.00b	9.00c	10.0c	9.00c

$p \leq 0.05$

REFERENCES

Zapata, M., Beaver J., and T.Porch. 2011. Dominant gene for common bean resistance to common bacterial blight caused by *Xanthomonas axonopodis* pv. *phaseoli.* Euphytica. Vol 179:373-382.

INDUCED *Bean Golden Mosaic Virus* SYMPTOMS BY VIRUS ISOLATES GATHERED FROM DIFFERENT COMMON BEAN GENOTYPES

Cantamessa[1] LA, Hoshino[1] AT, Oliveira[1] LM, Caviglione[2] JH, Kitzberger[2] CSG, Santos[2] AM, Ávila[2] MR, Androcioli[2] HG, Bianchini[2] A

[1]Universidade Estadual de Londrina - UEL, Londrina, PR, Brazil; [2]Instituto Agronômico do Paraná - IAPAR, Londrina, PR, Brazil; anesio@iapar.br

INTRODUCTION

The Bean Golden Mosaic Virus (BGMV), belonging to the Geminiviridae family and genus *Begomovirus*, transmitted by the whitefly (*Bemisia tabaci* Gennadius B biotype), has become one of the most limiting factors towards the common bean (*Phaseolus vulgaris* L.) cultivation in Brazil, causing losses up to 100% towards the crop's production (CIAT, 1990 e Ferreira; SALGUEIRO, 1993). The aim of this study was to evaluate the induced reaction towards BGMV isolates coming from different common bean genotypes, infected with a virus transmitted by *B. tabaci*.

MATERIALS AND METHODS

The experiment was conducted between the months of September and November at the Intituto Agronômico do Paraná (IAPAR), Lodrina – State of Paraná. Common bean plants located in a greenhouse expressing high mosaic (leaf yellowing), mild mosaic (leaf yellowing), high stunted and mild stunted symptoms, from the following genotypes: MD 1133, MD 1092 and IPR Campos Gerais were collected, totaling 12 isolate sources of BGMV. These plants were placed inside an enclosure containing 300 *B. tabaci*, which nurtured upon these plants for three days. Afterwards, five IPR Campos Gerais seedlings, planted in 275 mL tubes, were placed inside each enclosure, serving as susceptible test plants. After three days of nurturing, the seedlings were transferred to a temperature-controlled chamber (temperature of 25 ± 2°C, relative humidity (RH) of 70 ± 10%, 14h photoperiod). A randomized block design, containing 12 treatments (isolate source) and five repetitions (plants with symptoms) was employed. After 20 days the test plants were evaluated, being attributed a visual severity grade, depending on the mosaic and stunted symptoms expressed, between zero and five, where zero demonstrates the absence of symptoms and five demonstrates a maximum symptom manifestation. The grades were weighted as follows: mild expression (grade equal or bellow 2) by 1; moderate expression (grade between 2.5 and 3.5) by 2; high expression (grade equal or above 4) by 3, generating grades between 0 and 15. An analysis of variance by Tukey's test ($\alpha = 5\%$) was utilized.

RESULTS AND DISCUSSIONS

The derived genotype MD 1092 isolates expressed a mild degree of symptom severity, between 2.2 and 5.6, while the derived genotype IPR Campos Gerais isolates expressed a high degree of symptom severity, between 10 and 12.2 (Table 1).

Table 1. Symptom types and degree of severity (DS) induced by BGMV isolates derived from plants of different common bean genotypes and degrees of severity.

BGMV isolates		Grades of the susceptible test plants (DS)	
Symptom	Genotype	Mosaic	Stunting
HMS	MD1092	4.4 b	3.4 b
	MD1133	7.8 ab	7.2 ab
	IPR Campos Gerais	12.3 a	10.0 a
MMS	MD1092	2.8 b	2.3 b
	MD1133	8.0 a	4.4 ab
	IPR Campos Gerais	5.6 ab	8.2 a
HSS	MD1092	2.4 b	4.0 ab
	MD1133	9.3 a	6.8 a
	IPR Campos Gerais	8.8 a	8.0 a
MSS	MD1092	5.6 a	2.2 b
	MD1133	5.6 a	5.2 ab
	IPR Campos Gerais	4.5 b	2.9 b
CV (%)		15.5	22.8

HMS = high mosaic symptom; MMS = mild mosaic symptom; HSS = high stunted symptom; MSS = mild stunted symptom. * Averages followed by the same letter in the columns do not differ between themselves, Tukey ($\alpha = 5\%$).
The grades here shown are weighted, consisting of numbers between 1 and 15.

The symptoms and degree of severity did not differ between high symptom isolates from MD 1133 and IPR Campos Gerais genotypes. Every mild stunted isolate induced a lesser degree of severity compared to high stunted isolates of MD 1133 and IPR Campos Gerais. The lesser degree of severity induced by MD 1092 isolates and the mild stunted isolates, from the other genotypes, demonstrate that BGMV isolates differ from each other (NARDO & COSTA, 1986; BIANCHINI et al, 2004). The isolates taken from the MD 1092 genotypes differ statistically from the rest as they induce a lesser degree of severity of mosaic and stunted symptoms. Future studies which use a larger number of isolates deriving from other genotypes and sources, as well as molecular examination for confirmation are necessary.

REFERENCES

BIANCHINI, A., MOTOMURA, K., F., LEITE, R.P. Variabilidade de geminivirus que afetam o feijoeiro e outras espécies no estado do Paraná. Fitopatol. bras. 29 (Suplemento), agosto 2004.

CIAT. Research constraints provisionally identified by CIAT. In: Workshop on advanced *Phaseolus* beans. Research Network 11-14 de setembro, 1990. Cali, Colômbia, 1990.

SALGUERO, V. Perspectivas para el manejo del complejo mosca blanca-virosis. In: Tallerdelcenroamericano y del caribe sobre moscas blancas, Turrialba, Costa Rica. 1992. Las moscas blancas (Homoptera: Aleyrodidae) en America Central y Caribe: Memória. Turrialba: CATIE, 1993. p. 20-26. (Informe Técnico, 205).

NARDO, E. A. B., and COSTA, A. S. 1986. Diferenciação de isolados do complexo brasileiro do vírus do mosaico dourado do feijoeiro. Fitopatol. Bras. 11:655-666.

NEW SOURCES OF WHITE MOLD RESISTANCE DERIVED FROM WIDE CROSSES IN COMMON BEAN AND EVALUATED IN THE GREENHOUSE AND FIELD USING MULTI-SITE SCREENING NURSERIES COMPARING 2016 AND 2017 DATA

R. Higgins, Z. N. Kamvar, S.E. Everhart and J.R. Steadman

Department of Plant Pathology, University of Nebraska-Lincoln, Lincoln, NE 68583
Email: jsteadman1@unl.edu Data from M. Brick (CO), J. Kelly (MI), M. Wunch (ND), J. Myers (OR), P. Miklas (WA), E. Berghauer (WI), C. Urrea (NE)

The goal of our work over the past two years is to identify putative sources of resistance to white mold in adapted backgrounds at multiple sites located in most of the major bean-production areas of the northern states. Our approach combines evaluations in greenhouses using a straw test method that is consistent in identification of sources of resistance in adapted and non-adapted bean germplasm and a multi-site field evaluation at white mold nurseries in several locations throughout the major dry bean production regions of the United States.

In 2017, breeders sent 12 lines for greenhouse evaluations, and 9 lines were tested in the field nurseries. Tested lines included pinto, navy, black, and small red bean seed classes. Trials were conducted using these lines, plus the three controls G122 (partial resistance), Beryl (susceptible), and Bunsi (field avoidance).

Table 1. Greenhouse results from 2017 WMMN testing*

Line	NE	OR	WA	WI	CO	Mean	Grouping
ND121448	7.6	7.8	8.7	6.8	7.2	7.6	A
P14814	7.5	8.3	7.9	6.5	7.5	7.6	A
N14229	7.3	7.9	6.3	5.5	7.8	7	A B
B15430	7.6	7	7.8	6.1	6.2	6.9	A B
PT9-5-6	7.5	6.9	6.1	6.3	7.5	6.9	A B C
Beryl	7.2	5.6	7.9	4.3	7.7	6.5	A B C D
SR16-5	7.1	7.6	5.8	4.7	7.2	6.5	A B C D
NDZ14083	7.2	6.6	6.1	4.8	6.4	6.2	B C D E
Cayenne	4.7	5.2	5.9	5.5	6.9	5.7	C D E
Bunsi	6.6	5.1	6.3	4.2	5.8	5.6	D E
ND121630	5.8	3.2	6.1	4	6.8	5.2	E F
ND122386	4.4	5.3	3.8	3.5	4	4.2	F G
G122	5.2	2.8	3.8	4	4.6	4.1	F G
NE5-16-101	3.5	2.7	2.4	3.3	5.1	3.4	G
NE5-16-98	4	3.1	2.8	3.3	3	3.2	G

*Petzoldt & Dickson scale: 1-3 = resistant, 4-6 = intermediate, 7-9 = susceptible

Table 2. Field results from 2017 WMMN testing**

Line	NE	OR	WA	WI	MI	ND	Mean	Grouping
Beryl	4.5	6.7	7.5	8.5	4.3	8.1	6.6	A
SR16-5	5	7	4.4	6.5	2.3	6	5.2	A B
PT9-5-6	2	5	5.3	7.3	5	5.6	5	A B
P14814	4.5	2.3	5.5	6.5	3	7.1	4.8	A B
Cayenne	5	6.3	4.3	6.3	3	3.2	4.7	A B C
Bunsi	3.5	4.7	5.2	6	4	3.6	4.5	B C
NDZ14083	2	5.7	3.9	8.3	2.7	3.3	4.3	B C
NE5-16-98	3.7	5	3.9	7	3.7	2	4.2	B C
B15430	1.7	4.7	6.3	5.5	3	3.4	4.1	B C
N14229	1	1.7	6.3	4	4	4.2	3.5	B C
NE5-16-101	3.5	1.3	4.1	6.7	3.5	1.8	3.5	B C
G122	2	2	3.5	3	4	2.2	2.8	C

**CIAT scale- 1-9; 1= no disease, 9 = dead plant

In analysis of 2017 greenhouse data from five sites, we identified two pinto lines (NE5-16-98 and NE5-16-101) that had straw test ratings with better performance than the control (G122) and significantly reduced disease ratings compared to all other lines, except ND122386. Field trial data from six sites did not show a significant difference in lines (ANOVA; $p = 0.051$), however, five lines (NDZ14083, NE5-16-98, B15430, N14229, and NE5-16-101) had lower disease ratings than Bunsi, although none were lower than G122.

Table 3. Combined greenhouse results from 2016 and 2017 WMMN testing

Line	Year	NE	OR	WA	WI	CO	MI	Mean	Grouping
N15341	2016	8.8	8.4	7.9	9	8.4	8.7	8.5	A
P14815	2016	7.9	6.9	6.6	9	8.2	8	7.8	A B
NDF140436	2016	8.7	6.9	6.5	9	7.4	7.9	7.7	A B
ND121448	2107	7.6	7.8	8.7	6.8	7.2	NA	7.6	A B C
B15430	2016	8.9	7.8	7.5	9	5.2	7.3	7.6	A B C
P14814	2107	7.5	8.3	7.9	6.5	7.5	NA	7.6	A B C D
NDF140443	2016	8.8	7	5.6	9	6.2	8.6	7.5	A B C D
NDF140446	2016	8.6	6.4	6.3	8.6	6.6	8.4	7.5	A B C D
NDF140422	2016	8.4	6.3	6.8	8.6	5.7	8.7	7.4	A B C D E
NDF140433	2016	8.8	6.3	6.3	9	5.7	7.5	7.3	A B C D E F
N14229	2107	7.3	7.9	6.3	5.5	7.8	NA	7	B C D E F G
B15430	2107	7.6	7	7.8	6.1	6.2	NA	6.9	B C D E F G
WM91212-4-3	2016	7.3	5.9	6.5	8.3	7.4	6.2	6.9	B C D E F G
PT9-5-6	2107	7.5	6.9	6.1	6.3	7.5	NA	6.9	B C D E F G H
NDF140461	2016	8.3	6.6	6.9	6.6	4.2	8	6.7	B C D E F G H
ASR 1865	2016	7.1	4.8	5.2	7.8	6.1	8.9	6.6	B C D E F G H
NDF140423	2016	8.3	6	6.7	6.8	4.8	7.2	6.6	B C D E F G H
NDF140405	2016	7.3	6.1	6.6	7.6	5.2	6.7	6.6	B C D E F G H
Beryl	2107	7.2	5.6	7.9	4.3	7.7	NA	6.5	B C D E F G H I
SR16-5	2107	7.1	7.6	5.8	4.7	7.2	NA	6.5	B C D E F G H I
NDF141308	2016	7.6	4.2	5	8	7.2	6.7	6.4	C D E F G H I
R13752	2016	6.9	4.4	6.3	8.3	6.2	6.3	6.4	C D E F G H I
NDF140427	2016	7.1	5.4	5.7	8.3	4.8	7	6.4	C D E F G H I
Beryl	2016	7.7	5.1	5.8	5.8	5.8	8	6.4	C D E F G H I
NDF140409	2016	7.5	6.1	6.2	8.2	4.9	5	6.3	C D E F G H I
NDF140415	2016	7.5	6.1	5.4	7.3	5.4	6	6.3	D E F G H I
NDF140408	2016	6.8	6	6	7.4	5.2	6.1	6.2	D E F G H I
NDF140406	2016	8.2	5	6.3	4.9	5.1	7.9	6.2	E F G H I
NDZ14083	2107	7.2	6.6	6.1	4.8	6.4	NA	6.2	E F G H I J
NDF140460	2016	7.4	6	4.7	7.3	3.5	7.5	6.1	F G H I J
PS08-039A-5	2016	5.9	5	5.3	7.2	5.1	5.9	5.7	G H I J
Cayenne***	2107	4.7	5.2	5.9	5.5	6.9	NA	5.7	G H I J
Bunsi	2107	6.6	5.1	6.3	4.2	5.8	NA	5.6	G H I J
Bunsi	2016	7.8	4.9	5.8	5.6	4.3	5.1	5.6	H I J
R12844***	2016	5.5	5.2	5.7	5.8	4.6	6.8	5.6	H I J
ND121630	2107	5.8	3.2	6.1	4	6.8	NA	5.2	I J K
G122	2016	5.2	4.2	4.1	4.7	4.4	6.9	4.9	J K
ND122386	2107	4.4	5.3	3.8	3.5	4	NA	4.2	K L
G122	2107	5.2	2.8	3.8	4	4.6	NA	4.1	K L
USPT-WM-12	2016	4.2	3.7	3.8	4.3	3.3	5	4	K L
031-A-11	2016	4.2	3.7	4.4	3	3.3	5.2	4	K L
NE5-16-101	2107	3.5	2.7	2.4	3.3	5.1	NA	3.4	L
NE5-16-98	2107	4	3.1	2.8	3.3	3	NA	3.2	L

***R12844 = Cayenne

Collectively, these results suggest the two pinto lines with disease resistance in the greenhouse show promise for increased disease resistance in the field.

GENETIC PROGRESS IN LATE CYCLES OF RECURRENT SELECTION FOR RESISTANCE TO WHITE MOLD IN COMMON BEAN

Renato C. C. Vasconcellos, Juliana A. Dias, Fernanda S. Lopes, Antonio C. M. Porto, João B. dos Santos

Universidade Federal de Lavras (UFLA), CEP 37200-000, Lavras, MG – Brazil;
jbsantos@dbi.ufla.br

INTRODUCTION

The white mold (*Sclerotinia sclerotiorum* (Lib.) de Bary) is a fungus responsible for limiting the productivity of the common bean crop, causing considerable losses and consequent unfeasibility of the crop in certain regions and periods of the year. The interaction of the pathogen with its host is complex, and host tolerance is inherited as a quantitative trait with low to moderate heritability. An alternative to gradually accumulate a greater number of alleles of white mold resistance is through recurrent selection (Bernardo, 2010). This method is a cyclical and dynamic process that involves obtaining progenies, evaluating and recombining the best plants in order to increase the frequency of favorable alleles and, consequently, to improve the expression of character. The goal was to evaluate the genetic progress of the recurrent selection program to resistance to white mold on common beans and check if there are still significant gains with the selection for the continuity of the program.

MATERIAL AND METHODS

The best progenies of each recurrent selection cycle (IX to XI) of the recurrent selection program for resistance to white mold at the Federal University of Lavras (UFLA) were evaluated in the field, in the Brazilian dry season of 2015 using a triple 8 x 8 lattice design. It was evaluated the 4 best progenies $S_{0:4}$ of cycle IX, 24 best progenies $S_{0:3}$ of cycle X and 34 best progenies $S_{0:2}$ of cycle XI and the checks Corujinha (susceptible) and Cornell 605 (with partial high resistance). The plots consisted of a one-meter line and were inoculated with mycelium of the fungus and evaluated 10 plants per plot using the straw test (Terán and Singh, 2008). Among the progenies selected for resistance to white mold, selection for grain type and plant architecture was also performed. The analysis of variance, selective accuracy and genetic progress was performed.

RESULTS AND DISCUSSION

The success of recurrent selection was assessed by estimating the genetic progress. In the case of autogamous plants, one of the alternatives to estimate this progress is the evaluation of the best progenies obtained after each selective cycle. There were significant differences among the treatments and among the progenies within treatment for plant architecture and grain type, indicating that it is possible to identify superior progenies for these characteristics in these groups (Table 1). However, for resistance to white mold no variation was observed among treatments and among progenies within each cycle, because we selected the best resistant progenies in each cycle. Note that these progenies were not different from the resistant cultivar Cornell 605 (Check R), but differed from the susceptible line Corujinha (Check S), indicating that the selection per cycle was efficient and the progenies have a high partial resistance. In addition, a significant difference was observed at 5% level of probability for the progenies between cycles, indicating that even though there was no difference among the treatments in general, it is possible to observe differences among the cycles (Table 1).

The genetic progress for resistance to white mold was estimated considering the 4 best progenies from each of the 3 cycles, and it was estimated -6.07% per cycle, with an adjusted R^2 of 96.79%. The negative signal of the genetic progress indicates that there was a decrease in disease notes and, consequently, improvement resistance to the fungal. This indicates that the recurrent selection was efficient to obtain progenies with a high level of resistance to white mold with carioca grain type and upright architecture, and that it is possible to obtain additional gains with the selection for resistance to white mold, plant architecture and grain type, due to the presence of genetic variability, even after 11 cycles of recurrent selection.

Table 1. Analysis of variance of the progenies selected from the $S_{0:4}$ generations of cycle IX, $S_{0:3}$ of cycle X and $S_{0:2}$ of cycle XI of recurrent selection for resistance to white mold, plant architecture and grain type, using a grading scale of 9 notes for the three traits.

Source of Variation	DF	White Mold		Plant Architecture		Grain Type	
		MS	$r_{\hat{g}g}$	MS	$r_{\hat{g}g}$	MS	$r_{\hat{g}g}$
Rep	2	0.3526^{NS}		0.4375^{NS}		0.0016^{NS}	
Block (Rep)	21	1.0018^{**}		0.6438^{NS}		0.2102^{*}	
Treatments	63	0.3223^{NS}	0.4435	1.2734^{**}	0.7899	0.782^{**}	0.7966
Treatments (Groups)	59	0.2464^{NS}	0.4697	1.3296^{**}	0.8001	0.3323^{**}	0.8171
Prog. within CIX	3	0.2466^{NS}		1.3969^{**}		0.3235^{**}	
Prog. within CX	23	0.1663^{NS}		1.2702^{**}		0.2657^{**}	
Prog. within CXI	33	0.2068^{NS}		1.3517^{**}		0.3715^{**}	
Groups	4	1.2300^{**}		0.3694^{NS}		6.8468^{**}	
Check R vs Cycles	1	0.4055^{NS}		0.1264^{NS}		8.3938^{**}	
Check S vs Cycles	1	3.4072^{**}		0.2726^{NS}		14.199^{**}	
Between Cycles	2	0.8275^{*}		0.5827^{NS}		0.3320^{NS}	
Error	104	0.2096		0.4787		0.1104	

*. **: Significant at 5% and 1% level of probability; ^{NS}: Not significant by the F test.
$r_{\hat{g}g}$: selective accuracy.

REFERENCES

BERNARDO. R. Breeding for quantitative traits in plants. 2nd ed. Minnesota: Stemma. 2010.
TERÁN, H.; SINGH, S. P. Annual Report of the Bean Improvement Cooperative, v. 51, n. 42, 2008.

DETACHED LEAF ASSAY IN THE REACTION SCREENING OF BEAN GENOTYPES TO WHITE MOLD

Antonio C. M. Porto, Raoni Gwinner, Moacir Pasqual, João B. dos Santos

Universidade Federal de Lavras (UFLA), CEP 37200-000, Lavras, MG – Brazil;
jbsantos@dbi.ufla.br

INTRODUCTION

White mold is a major disease in the common bean production chain. The plant resistance to the generalist necrotrophic fungus *Sclerotinia sclerotiorum* (Lib.) de Bary is quantitative, what implies in a considerably large environmental effect during the selection for this trait. Several methods have been proposed to evaluate plant resistance in this pathosystem aiming a more efficient and cost-effective procedure. Moreover, methods with reduced evaluation time and indoor methodologies that keep the inoculum in a controlled environment have being developed (Terán & Singh, 2008). The detached leaves assay has been used efficiently for plant resistance evaluation (Elmor et al., 2017). Nevertheless, lacks information regarding common bean - *S. sclerotiorum* pathosystem characterization using this method for most of Brazilian common bean lines. In this paper, we aim to evaluate the reliability of this assay using the common bean - *S. sclerotiorum* bean pathosystem.

MATERIAL AND METHODS

Detached leaf assay: To measure lesion formation, we infected 12 diverse common bean genotypes with *S. sclerotiorum* plugs. We used a randomized block design with three independent biological replicates of each plant genotype. The whole experiment was repeated twice with independent randomization between experiments leading to six measurements per genotype. We randomly sampled adult leaflets per plant and each leaflet was used for inoculation of 1 *S. sclerotiorum* plug. Leaflets were placed on 1% phytoagar flats with humidity domes on top. Control leaves were mock-inoculated with sterile PDA plugs without pathogen. All leaflets infections were photographed at 60, 84, 108, 132 and 156 hours post inoculation for downstream image analysis.

Automated Image Analysis: We measured lesion areas using the ebimage and crimage packages (Pau et al., 2010; Failmezger et al., 2010) in the R statistical environment (R development core team, 2016). Leaflets were identified as objects with green hue, and lesions were identified as low-saturation objects within the leaflet. Images masks were generated for both the leaflet and lesion and manually refined by a technician to ensure proper object calling. The area of the leaflets and lesions were then automatically measured as pixels per lesion and converted to absolute area using a 1 cm control object within each image.

Data analysis: The AUDPC (area under the disease progress curve) was calculated at plot level using lesion size values across time. AUDPC mean values were compared by Tukey test (0.05) using R software (R development core team, 2016).

RESULTS AND DISCUSSION

Genotypes presented distinct AUDPC means (p=0.05) (Figure 1a). Therefore, the genotypes were clustered as: susceptible (AUDPC $\geq$ 800), moderate resistance (799 $\leq$ AUDPC $\geq$ 400) and resistant (AUDPC $\leq$ 399). Comparing the genotypes with higher and lowest AUDPC value, our data indicates that the susceptible genotype 64/8 reach the resistant AUDPC before 108 hours of inoculation (Figure 1b,1c). Furthermore, some genotypes previously characterized by the straw

test, presented non-similar results for resistance to white mold. For example, the genotype M20 described as susceptible by Carneiro et al. (2011) presented some level of resistance when phenotyped by the straw test.

The genotypes Corujinha and Cornell 605 were clustered as with moderate resistance. Nevertheless, Leite et al. (2017) classified Corujinha as susceptible, and Cornell 605 as resistant. Considering the differences of evaluation among experiments, the distinct plant behavior between experiments suggest distinct resistance mechanisms in a single genotype acting in different plant structures (stem and leaf), what can lead to a plant resistance gradient depending on the evaluated tissue. The detached leaves assay has the potential to be used for phenotyping the reaction of common bean to white mold, especially when field and greenhouse evaluation are not available. However, this method need to be better compared with other more reliable like straw test, and mainly select the plant structures to be evaluated that better represent the level of resistance of the genotype.

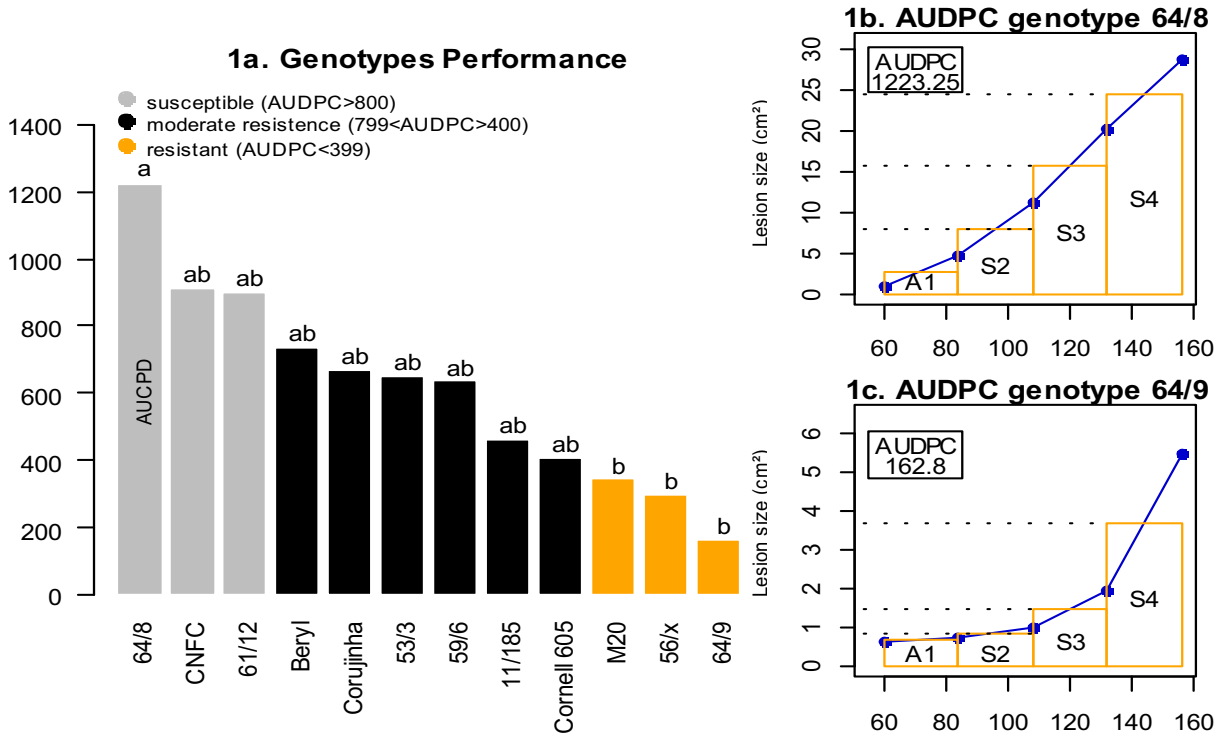


Figure 1. Genotypes performance for AUDPC values (1a), and curves of disease progress in the susceptible (1b) and resistant (1c) genotypes. *1a - genotypes with the same letter do not differ among themselves through the Tukey test at 5% probability.

REFERENCES

ELMOR, I. et al. Plant Disease, 2017.

TERÁN, H.; SINGH, S. P. Annual Report-Bean Improvement Cooperative, v. 51, n. 42, 2008

LEITE, M. E. et al. Bioscience Journal, v. 33, n. 5, 2017.

CARNEIRO, F. F. et al. Crop Breeding and Applied Biotechnology, v. 11, n. 2, p. 165-173, 2011.

PAU, G. et al. Bioinformatics, v. 26, n. 7, p. 979-981, 2010.

R Development Core Team. R Foundation for Statistical Computing, Vienna, Austria, 2016.

FAILMEZGER, H. et al. R Packag version 1.4.0, 2010.

NUMBER OF FUNGICIDE APPLICATIONS AND GENOTYPE MAY AFFECT INCIDENCE OF *Sclerotinia sclerotiorum* IN SEED OF COMMON BEAN

Teixeira, P.H.[1]; Sousa, L.R.V.[1]; Souza, A.F.F.[1]; Ferreira, T.C.[2]; Silva, P.M.L.[1]; Mendes, O.L.[1]; Rocha, B.M.[1]; Lima, R.C.[3]; Teixeira, H.[4]; Paula Júnior, T.J.[2]; Vieira, R.F.[5]

[1]Univ. Federal de Viçosa, 36570-000 Viçosa, MG, Brazil (pablo.teixeira@ufv.br); [2]Epamig, CP 216, 36570-000 Viçosa, MG, Brazil; [3]IFMT, 78890-000 Sorriso, MT, Brazil; [4]Epamig, CP 176, 37200-000 Lavras, MG, Brazil; [5]Embrapa/Epamig, Viçosa, MG.

INTRODUCTION. White mold (WM), caused by the fungus *Sclerotinia sclerotiorum* (Ss), is a devastating disease in irrigated areas of common bean in Brazil. Ss is able to survive in seeds as dormant mycelium in testa and cotyledons. Ss transmission by seed is an important mean of dispersion, especially to areas free of WM. Here we evaluated the effect of number of fungicide applications and genotypes on incidence of Ss in seed (ISsS) of common bean.

MATERIAL AND METHODS. We used seeds from three irrigated field trials in which treatments were arranged as a 3 x 4 factorial combinations of genotypes (VC 17, Pérola or Madrepérola) and number of fungicide applications (0, 1, 2 or 3). Two trials were conducted in Viçosa and one in Oratórios, Zona da Mata region, Minas Gerais State, Brazil, in a field naturally infested with sclerotia of Ss during the fall-winter season. Under field conditions, the line VC 17 has exhibited partial resistance to WM, Pérola has exhibited moderate resistance, and Madrepérola has exhibited susceptibility to WM. In these trials, the fungicide fluazinam (0.625 L ha^{-1}) was first applied at the beginning of the flowering stage. With two or three applications of fungicide, interval between applications was eight days. WM pressure was low in Viçosa (2015), low/moderate in Viçosa (2016) and moderate/high in Oratórios (2015). We used the method of Neon-R for detection of ISsS (Kawasaki and Machado, 2013). This method is based on the change of the substrate color from blue to yellow as the result of the oxalic acid action produced by the pathogen around the seeds. Four hundred normal seeds (chalky, discolored and shriveled seeds were removed) from each treatment were randomly placed on the top of the culture medium. We used 20 plastic Petri dishes (150 mm) per treatment and 20 seeds per Petri dish. A randomized block design with four replications was used. The Petri dishes were incubated at 20ºC in the dark. After two days, Petri dishes with yellow colors around seeds were isolated. The production of sclerotia around seeds for up 12 days confirms the presence of the fungus. Petri dish with presence of sclerotia indicates that at least one seed in 20 was infected with Ss.

RESULTS AND DISCUSSION. ISsS was lower in Viçosa (2015) than in the other trials (Fig. 1), indicating that higher WM pressure in the trial may imply higher ISsS. ISsS was 0.062% for Madrepérola, 0.099% for Pérola and VC17 (Fig. 2). ISsS was higher when no fungicide was used and lower when fungicide was applied twice (Fig. 3). Foliar-applied fungicide decreased ISsS by 9-fold for Madrepérola and from 2.8- to 11-fold for Pérola (Fig. 4). For line VC 17, results of ISsS in response to number of fungicide applications were inconsistent. Our results suggest that foliar-applied fungicide decrease ISsS in some genotypes of common bean.

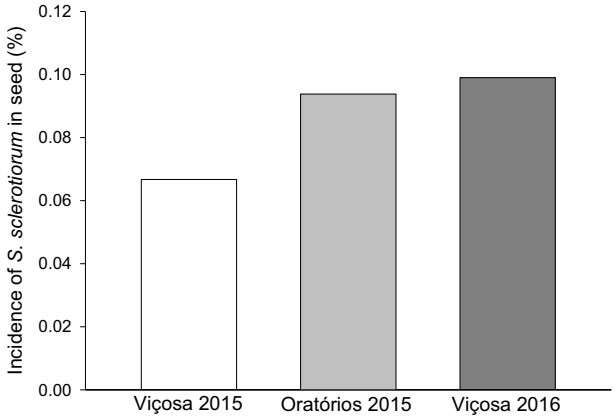


Figure 1. Incidence of *S. sclerotiorum* in seed in the three field trials was averaged across genotypes and number of fungicide applications.

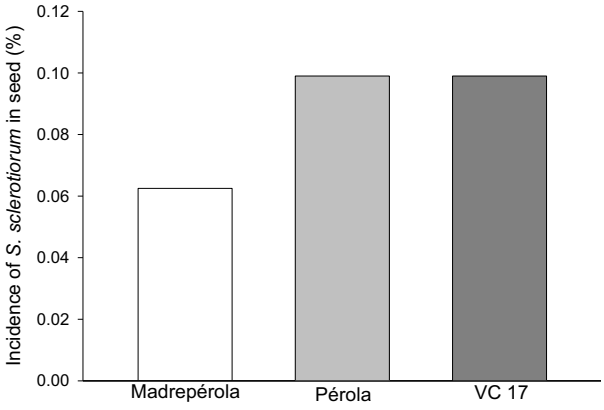


Figure 2. Incidence of *S. sclerotiorum* in seed in response to genotypes was averaged across number of fungicide applications and trials.

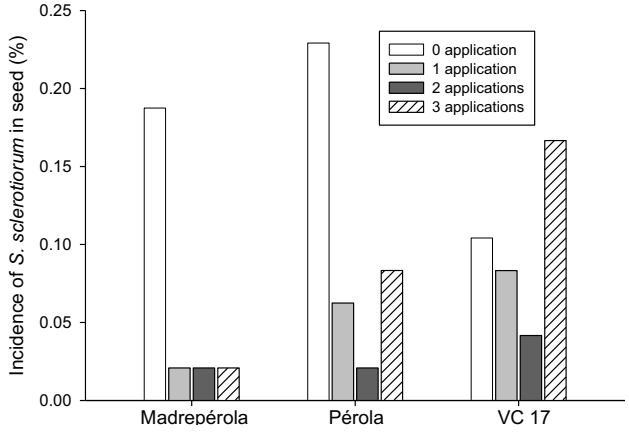


Figure 3. Incidence of *S. sclerotiorum* in seed in response to number of fungicide applications was averaged across genotypes and trials.

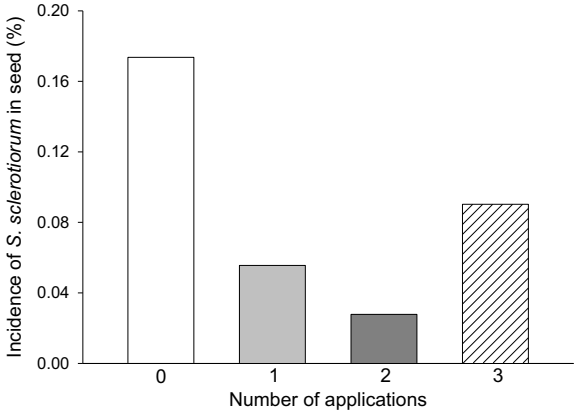


Figure 4. Incidence of *S. sclerotiorum* in seed in response to number of fungicide applications and genotypes was averaged across trials.

ACKNOWLEDGMENTS:
CNPq, FAPEMIG and EMBRAPA for financial support.

REFERENCE

Kawasaki, V.H.; Machado, J.C. 2013. Establishment of a semi-selective method for the detection of *Sclerotinia sclerotiorum* in dry bean and soybean seeds. Journal of Seed Science, 35:435-442.

PERFORMANCE IN FIELD OF COMMON BEAN GENOTYPES SELECTED FOR PARTIAL RESISTANCE TO WHITE MOLD

Teixeira, P.H.[1]; Souza, A.F.F.[1]; Silva, P.M.L.[1]; Sousa, L.R.V.[1]; Ferreira, T.C.[2]; Mendes, O.L.[1]; Lima, R.C.[3]; Rocha, B.M.[1]; Paula Júnior, T.J.[2]; Carneiro, J.E.S.[1]; Vieira, R.F.[4]

[1]Univ. Federal de Viçosa, 36570-000 Viçosa, MG, Brazil (pablo.teixeira@ufv.br); [2]Epamig, CP 216, 36570-000 Viçosa, MG, Brazil; [3]IFMT, 78890-000 Sorriso, MT, Brazil; [4] Embrapa/Epamig, Viçosa, MG.

INTRODUCTION

Genetic resistance can help farmers to manage white mold (WM), disease caused by *Sclerotinia sclerotiorum*. Our objective was to evaluate the performance in field of common bean genotypes screened for reaction to WM in the Value for Cultivation and Use (VCU) trials conducted between 2008 and 2016. In the VCU field trials, conducted by region or state in Brazil, elite lines are compared with registered control cultivars according to the National Register of Cultivars/Ministry of Agriculture, Livestock and Supply rules.

MATERIAL AND METHODS

Two irrigated trials (in the districts of Viçosa and Oratórios) with 20 common bean genotypes were conducted during the fall-winter season of 2017 in an area with a history of WM. Ten lines and the cultivars Ouro Branco and Vereda were selected in the VCUs trials for partial resistance to WM; the cultivars Pérola and Estilo for moderate resistance; and the cultivars Ouro Negro, Majestoso and Ouro Vermelho for susceptibility to WM. The following international genotypes with partial resistance to WM were also included for comparison: A195, G122 and Cornell 605. A randomized complete block design with four replications was used. Plots were two 3 m-long rows, spaced 0.50 m apart.

RESULTS AND DISCUSSION

WM pressure was moderate in Viçosa and moderate/high in Oratórios. VC 17 and Pérola were moderately attacked by anthracnose. For this reason they exhibited relatively low yield (Table), differently from those yield obtained in previous years. The association between WM incidence (WMI) and WM severity index (WMSI) was very high ($r > 0.90$). The correlation between yield and WMSI ($r = -0.28$, $p = 0.005$) was significant in Oratórios. The correlation between yield and plant height in the growth stage R7 ($r = 0.62$) was very highly significant in both sites. Canopy closure (100% indicates that the soil is not visible) at the end of flowering correlated with WMSI in Viçosa ($r = 0.36$, $p = 0.001$) and in Oratórios ($r = 0.48$, $p = 0.001$). Plant lodging at harvest correlated with WMSI in Viçosa ($r = 0.36$, $p = 0.001$) and in Oratórios ($r = 0.50$, $p = 0.001$). The association between plant height and WMSI was negative but non-significant. The lines CNFC 10432 and CNFP 11990 were in the group of the genotypes with the highest yield in both sites. These lines and the genotype A195 were in the group with lower WMSI. The cultivars Ouro Negro, Ouro Vermelho and Majestoso as well as the line CNFC MG11-08 were the genotypes most susceptible to WM. The carioca bean line CNFC 10432 and the black line CNFP 11990 confirm the good performance they have been since 2012 in areas with a history of WM.

Table. Grain yield, white mold incidence (WMI) and white mold severity index (WMSI) of genotypes screened from VCU field trials and three international genotypes with partial resistance to WM (A 195, G 122 and Cornell 605) in Viçosa and Oratórios, Zona da Mata region, Minas Gerais State, Brazil, 2017.

Genotype[1] (seed class)	Viçosa			Oratórios		
	Yield (kg/ha)	WMI (%)	WMSI[2] (%)	Yield (kg/ha)	WMI (%)	WMSI[2] (%)
CNFC 10720 (C)	3025 A[3]	51 B	30 B	3213 B	52 C	30 C
CNFP 10798 (B)	2950 A	52 B	32 B	3067 B	65 B	37 B
CNFC 10432 (C)	2921 A	50 B	32 B	3771 A	44 D	26 D
CNFC 10722 (C)	2821 A	46 B	25 B	3058 B	56 C	32 C
VC 26 (C)	2763 A	66 A	47 A	2742 C	64 B	35 C
BRS Estilo (C)	2750 A	45 B	37 A	2583 C	65 B	44 B
VC 27 (C)	2700 A	49 B	34 A	2358 D	72 B	43 B
CNFC MG11-08 (C)	2675 A	55 A	39 A	2713 C	81 A	53 A
CNFP 11990 (B)	2592 A	41 B	21 B	3846 A	39 D	22 D
Ouro Branco (A)	2463 B	40 B	21 B	2821 C	33 D	16 D
CNFC 11946 (C)	2392 B	42 B	28 B	3067 B	51 C	31 C
A 195 (A)	2346 B	38 B	23 B	2625 C	40 D	19 D
BRS Vereda (R)	2238 B	50 B	30 B	2288 D	72 B	39 B
Majestoso (C)	2200 B	71 A	48 A	2442 D	81 A	57 A
VC 17 (C)	2171 B	48 B	26 B	2533 C	54 C	23 D
G 122 (A)	2108 B	53 B	35 A	2196 D	36 D	24 D
Ouro Vermelho (R)	2033 B	63 A	42 A	2779 C	84 A	54 A
Cornell 605 (A)	2000 B	37 B	17 B	2186 D	64 B	38 B
Pérola (C)	1996 B	30 B	12 B	2304 D	33 D	15 D
Ouro Negro (B)	1921 B	65 A	49 A	2067 D	89 A	62 A
Mean	2453	49.6	31.4	2733	58.8	35.0
CV (%)	13	27	33	13	22	23

[1]Cultivars in italic + bold are moderately resistant to white mold in the field; cultivars in bold are susceptible. Between parentheses: C = carioca, B = black, A = Andean, R = red.

[2]At harvest, plants were rated from 0 to 4, in which 0 = no symptoms, and 4 = 76 to 100% of the plant with symptoms of white mold (Hall and Phillips, 1996). WMSI was calculated for each plot on a percentage basis by the following formula: Σ (scores of all plants) / [4 × (total number of plants)] × 100.

[3]Tested by Scott-Knott groupment analysis test ($p < 0.05$).

ACKNOWLEDGMENTS: CNPq, CAPES and FAPEMIG for financial support.

REFERENCE

Hall, R.; Phillips, L. G. 1996. Evaluation of parameters to assess resistance of white bean to white mold. Annual Report of the Bean Improvement Cooperative, 39:306-307.

EFFECT OF CLIMATE CHANGE ON THE POTENTIAL DISTRIBUTION OF FOUR WILD BEANS (*Phaseolus* spp.)

IM Cerda-Hurtado[1], S Hernández-Delgado[1], MH Reyes-Valdés[2], N Mayek-Pérez[3], and JM González-Prieto[1*]

[1]Instituto Politécnico Nacional, CBG. Reynosa Tamps, México 88710. [2]UAAAN, Coah, México 25315. [3]UMAN, Tamps, México 88630. *jmgonzalezp@ipn.mx

INTRODUCTION

Five *Phaseolus* species have been domesticated: *Phaseolus vulgaris* [L.], *Phaseolus lunatus* [L.], *Phaseolus coccineus* [L.], *Phaseolus polyanthus* [Green], and *Phaseolus acutifolius* [A. Gray]) (Hernández-Delgado *et al*. 2015). Crop wild relatives as well as other species of wild plants have experienced genetic erosion and extinction due to direct and indirect environmental changes by humans. Due the potentially devastating impacts of climate change on biodiversity, in urgent needed to conserve the diversity of wild relatives. The aim of this study was to evaluate the impact of climate change in the years 2020 and 2080 on the potential distribution model (DM) of wild *P. acutifolius, P. coccineus* subsp. *coccineus, P. coccineus* subsp. *striatus,* and *P. vulgaris* in Mexico.

MATERIALS AND METHODS

Occurrences with geographical information (920) were obtained from several herbarium databases and germplasm banks. Bioclimatic variables (19) were calculated for Mexico based on climate date of the periods 1961-2009 (reference climatology) and compared with 2020s (2015-2039) and 2080s (2075-2099), under two representative concentration pathways of greenhouse gases (RCP 4.5 and 8.5). Distributions were modeled by using Maxent (Philips *et al*., 2006). Potential distribution surface results were classified on three categories as percentages of environmental fitness: 0-0.3 probability LEA (Low Environmental Aptitude); 0.3-0.7 probability MEA (Medium Environmental aptitude) and 0.7-1.0 probability HEA (High Environmental Aptitude).

RESULTS AND DISCUSSION

All models had an average test AUC>0.93, suggesting a good aptitude of the models to discriminate the specie´s fundamental climatic niche. Table 1 shows the changes of patterns of distribution of MEA and HEA surface under future climate scenarios with respect to the current scenario. MEA surface decrease between 10.1% until 19.3% for *P. coccineus* subsp. *coccineus*, whereas taxa *P. acutifolius*, *P. coccineus* subsp. *striatus,* and *P. vulgaris* increase the surface between 1.1 and 163.3%. The changes in HEA surface varies between -13.1% for *P. vulgaris* and +145.2% for *P. coccineus* subsp. *striatus*. The projected changes in distribution are showed in Fig. 1.

Table 1. Percentage of variation on the surface of two categories of environmental fitness for potential distribution of wild *Phaseolus* in current climatology (1961-2009) according two climate change scenario (RCP 4.5 y 8.5) and two periods: 2020s and 2080s.

Model	Medium Environmental Aptitude					High Environmental Aptitude				
		RCP4.5		RCP8.5			RCP4.5		RCP8.5	
	1961-2009 Surface Ha	Δ% 2020s	Δ% 2080s	Δ% 2020s	Δ% 2080s	1961-2009 Surface Ha	Δ% 2020s	Δ% 2080s	Δ% 2020s	Δ% 2080s
P. acutifolius	37,020,606	1.1	3.5	4.3	9.7	4,069,879	21.0	**-8.2**	16.0	2.8
P. coccineus* subsp. *coccineus	23,440,185	**-10.1**	**-19.3**	**-13.6**	**-10.9**	2,901,410	15.9	36.5	17.8	13.5
P. coccineus* subsp. *striatus	6,555,397	156.2	70.6	163.3	49.5	1,182,451	145.2	78.1	115.5	102.2
P. vulgaris	20,399,135	17.1	28.4	19.8	28.7	1,665,740	40.4	2.3	**-13.1**	11.0

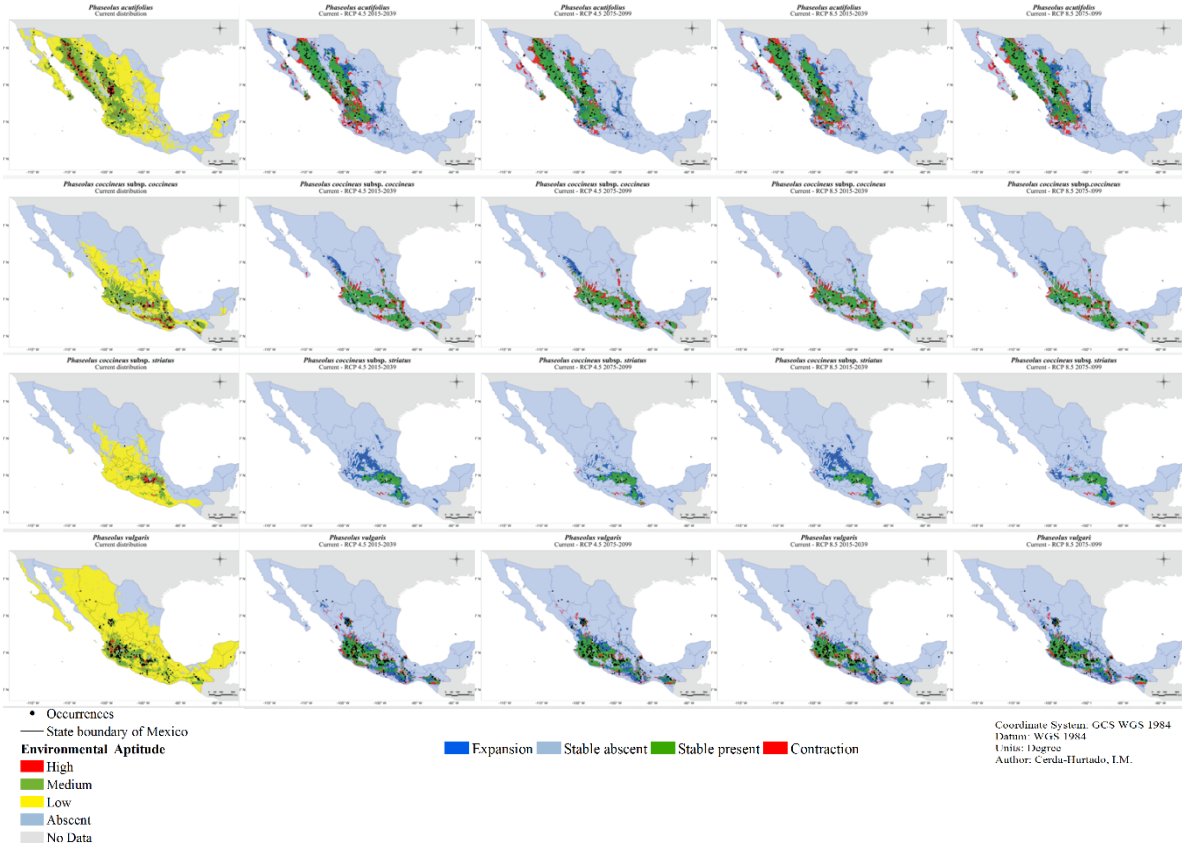


Figure 1. Predicted distribution of MEA and HEA area change under scenario RCP 4.5 and RCP 8.5 for the year 2015-2039 and 2075-2099 as compared to current model of wild *Phaseolus*.

CONCLUSIONS

Our study predicts contractions and range shifts towards northern areas for the studied species. These changes can be result in loss of genetic diversity. We therefore suggest to long term monitoring programs and collecting of CWRs for *ex situ* backing up.

REFERENCES

Hernández-Delgado, S., *et al*., 2015. In: M. Caliskan (ed.). Molecular approaches to genetic diversity. Pages 47-73.

Philips, S. J., *et al*., 2006. Ecological Modelling. 190:231-259.

AGRONOMIC AND PHYSIOLOGICAL PERFORMANCE OF COMMON BEAN LINES UNDER WATER DEFICIT

Gratão AS[1], Gonçalves-Vidigal MC[1], Elias JCF[1], Vidigal Filho PS[1], Martins VSR[1], Valentini G[1], Franzon RC[1], Moda-Cirino V[2]

[1]Departamento de Agronomia, Universidade Estadual de Maringá, Maringá, PR, Brazil;
[2]Instituto Agronômico do Paraná, Londrina, PR, Brazil. *E-mail: vidigalfilhop@gmail.com

INTRODUCTION

The cultivation of common bean can be greatly affected by hydric deficit limiting especially the grain yield. Great efforts from breeding programs have been made to obtain common bean drought-tolerant cultivars. In this context, the study had as objective to evaluate the common bean lines derived from the cross IAPAR 81 (drought tolerance) × LP 97-28 (low drought tolerance) submitted to hydric deficit under field conditions.

MATERIAL AND METHODS

The experiments consisted of 32 $F_{2:6}$ lines from the IAPAR 81 × LP 97-28 cross, the parents and controls Flor Diniz, Juriti, Tangará and BAT93, totaling 38 treatments, that were assessed under field conditions at Centro de Treinamento de Irrigação (CTI-UEM), Universidade Estadual de Maringá (UEM), Paraná, Brazil, from August to November 2014. Seeds were obtained at the Núcleo de Pesquisa Aplicada a Agricultura (Nupagri). The experiments were conducted in a randomized block design with three replications. Each plot was composed of two lines of 1.5 m in length, spaced 0.5 m between rows, with a density of 15 plants per linear meter. Two experiments were conducted under field condition: drought stress and no drought stress experiments. For the drought stress experiment, lines where submitted to hydric suppression for a period of 14 days during the reproductive phase R5 to R7, while for no drought stress experiment the water supplementation was normally carried out. Characteristics of agronomic interest were evaluated during the normal common bean cycle in both experiments: plant height (PH), days to flowering (DF), days to maturity (DM), number of seeds per pod (NSP), number of pods per plant (NPP), 100 seeds weight (SW) and grain yield. Physiological characteristics were observed 10 days after plants were submitted to hydric deficit: CO_2 concentration in the sub-stomatal internal cavity (ci), foliar transpiration rate (e), stomatal conductance (gs) and photosynthetic rate (a). Statistical analysis were performed using the Genes Computational Software (Cruz, 2013).

RESULTS AND DISCUSSION

The hydric suppression during common bean reproductive cycle provided a reduction of agronomic characteristics of interest, especially grain yield (Table 1 and 2). For no drought stress experiment, the mean grain yield was 1,936 kg ha^{-1}, while grain yield in the drought stress experiment was reduced to 1,089 kg ha^{-1}. Similarly, the number of pods per plant was severely reduced when genotypes was submitted to the drought stress condition. The number of seeds per pod and photosynthetic rate exhibited heritability, which magnitude values ranged from 15.31 to 91.38% (no drought stress) and from 10.72 to 91.37% for the the drought stress experiment, respectively. These heritability coefficients could be associated with higher additive genetic variances and reduced genotype × environment interaction (Fehr, 1987). The amount of genotypic

Table 1. Mean value, coefficient of variation (CV), heritability (h^2), genotypic variance (σ^2_G) and phenotypic variance (σ^2_F) observed for the 32 $F_{2:6}$ common bean lines from the IAPAR 81 × LP97-28 cross and controls in the **no drought stress** experiment

Characteristics	Mean	CV(%)	h^2 (%)	σ^2_G	σ^2_F
PG - Plant height (cm)	54.68	1.89	80.14	1.08	1.35
DF - Days to flowering	47.41	2.59	49.65	1.36	2.75
DM - Days to maturity	78.74	8.69	50.68	7.74	15.35
NSP - Number of seeds per pod	5.50	10.1	15.31	0.018	0.12
NPP - Number of pods per plant	10.82	17.18	41.12	0.80	1.95
SW - 100 seeds weight (grams)	17.73	6.92	74.77	1.48	1.99
Yield - Grain yield	1,936.25	19.89	53.74	57,469.52	106,931.24
Ci - CO_2 concentration in the sub-stomatal internal cavity (μmol CO_2 mol^{-1})	215.19	5.55	80.04	190.81	238.39
e - Foliar transpiration rate (mol m^{-2} s^{-1})	3.77	5.59	90.00	0.13	0.14
gs - Stomatal conductance (cm s^{-1})	0.39	11.88	89.58	0.006	0.007
a - Photosynthetic rate (μmol.m^{-2}.s^{-1})	20.42	6.25	91.38	5.77	6.31

variance in both experiments (Tables 1 and 2) demonstrated the possibility to obtain superior lines. Lines 22, 27, 29 and 102 were the most promising in relation the performance of the characteristics number of seeds per pod, number of pods per plant, mean 100 seed weight (SW), yield, CO_2 concentration in the sub-stomatal internal cavity, foliar transpiration rate and photosynthetic rate.

Table 2. Mean value, coefficient of variation (CV), heritability (h^2), genotypic variance (σ^2_G) and phenotypic variance (σ^2_F) observed for the 32 $F_{2:6}$ common bean lines from the cross IAPAR 81 × LP97-28 and controls in the **drought stress** experiment

Characteristics	Mean	CV (%)	h^2	σ^2_G	σ^2_F
PG - Plant height (cm)	49.89	1.79	-	-	4.88
DF - Days to flowering	47.83	1.95	61.67	0.39	0.64
DM - Days to maturity	75.78	10.8	70.09	1.89	2.62
NSP - Number of seeds per pod	5.10	12.67	10.72	0.016	0.15
NPP - Number of pods per plant	7.00	11.99	86.7	1.53	1.76
SW - 100 seeds weight (grams)	16.92	7.32	82.71	2.44	2.95
Yield - Grain yield	1,089.19	21.79	73.46	52,019.51	70,809.50
Ci - CO_2 concentration in the sub-stomatal internal cavity (μmol CO_2 mol^{-1})	212.03	6.74	87.06	459.61	527.89
e - Foliar transpiration rate (mol m^{-2} s^{-1})	3.17	6.06	93.25	0.17	0.18
gs - Stomatal conductance (cm s^{-1})	0.30	10.76	90.55	0.003	0.003
a - Photosynthetic rate (μmol.m^{-2}.s^{-1})	17.71	9.04	91.37	9.05	9.9

REFERENCES

Cruz (2013) Acta Scientiarum 35:271-276.
Fehr (1987) 465p.

PHYSIOLOGICAL PERFORMANCE AND VARIABILITY OF BEAN SEEDS UNDER WATER DEFICIT

L.V. de S. Cangussú[1], A.M.S. de S. David[1], F.H.B. Machado[1], C.D.da Silva[1], J.C. Figueiredo[1], R.A.Alves[1], J.L.R. Barbosa[1], R.A.N. Silva[1], E.V.C. Jorge[1], M.B.O. Silva[1]*, I. Aspiazú[1], A.J. de Carvalho[1]

[1]Universidade Estadual de Montes Claros
*Corresponding author: mariunim@yahoo.com.br

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is of great importance around world. However, genotypic constitution and cultivation environments influence the physiological performance of seeds, especially when submitted to water deficit condition. The low availability of water in the soil is a limiting factor for bean production, especially in three critical stages: germination, flowering and grain filling (SORATTO *et al*., 2003). Thus, the objective of this work was to evaluate the physiological performance and variability of seeds of bean cultivars under water deficit.

MATERIALS AND METHODS

The experiment was conducted at the Seeds Analysis Laboratory of the State University of Montes Claros, Janaúba-MG in September 2016. Two common bean cultivars (Pérola and BRS Estilo) of the carioca commercial group were used. Aqueous solutions of polyethylene glycol 6000 (PEG 6000), in the osmotic potentials of 0.0 - control (distilled water); -0.4; -0.8; and -1.2 MPa, were used to simulate water deficit according to the formula proposed by Villela *et al*. (1991). The experimental design was completely randomized, with four replicates per treatment. The evaluated characteristics were germination, first germination count (BRASIL, 2009) and germination speed index - GSI (MAGUIRE, 1962). The data were submitted to analysis of variance and when significant the effects of the cultivars were evaluated by the "F" test at 5% of significance, while the effects of osmotic potential levels were submitted to regression analysis.

RESULTS AND DISCUSSION

The results of the analysis of variance revealed isolated effect of osmotic potentials levels for all studied variables. It was also observed an isolated effect of the cultivars only for the GSI. Germination and seed vigor (first germination count and GSI) were adjusted in a linear regression equation of decreasing linearity, as the levels of osmotic potentials of the substrates decreased, that is, a larger amount of PEG 6000, the values of the variables were reduced (Figure 1). Increasing the concentration of salts in the substrate causes a reduction in the osmotic potential, resulting in a lower capacity of water absorption by the seeds, which generally influences germination capacity and plantlets development (CHAVES *et al*., 2009).

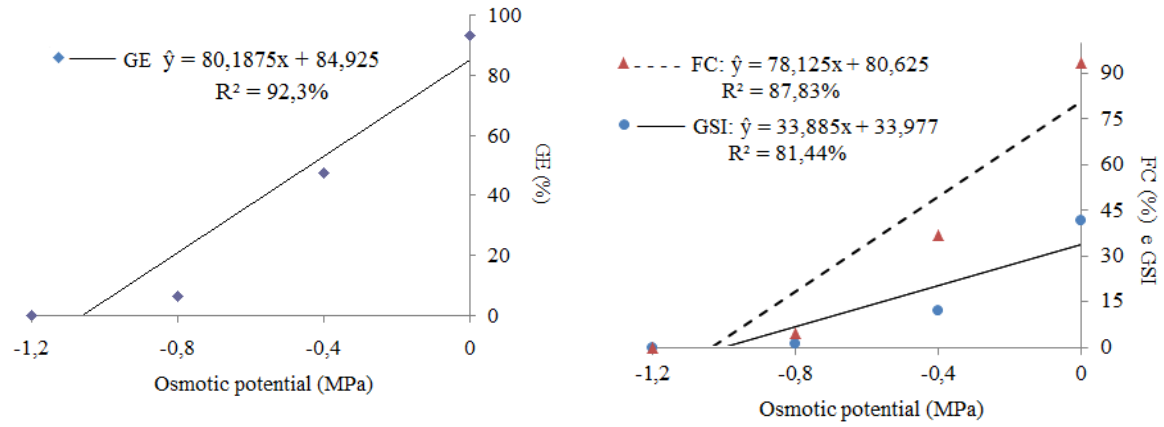


Figure 1. Germination (GE), first germination count (FC) and germination speed index (GSI) of seeds of carioca bean cultivars submitted to different levels of osmotic potential.

When comparing the cultivars (Table 1), it is observed that the Pérola cultivar presented higher GSI in relation to BRS Estilo cultivar. Higher values of GSI indicate that seeds germinate more rapidly and uniformly, and are therefore more vigorous. In addition, the detected difference can be attributed to the genetic constitution of the evaluated materials.

Table 1. Germination speed index (GSI) of bean cultivars of the carioca commercial group

Cultivars	GSI
Pérola	14,46 A
BRS Estilo	12,82 B
CV %	14,65

CONCLUSIONS

The physiological performance of bean seeds of Pérola and BRS Estilo cultivars is negatively affected by water deficit, simulated by PEG 6000. There is variability in vigor of the studied cultivars, being the Pérola cultivar more vigorous than the BRS Estilo cultivar.

REFERENCES

BRASIL. MINISTÉRIO DA AGRICULTURA, PECUÁRIA E ABASTECIMENTO. Regras para análise de sementes. Brasília: DNDV/CLAV, 2009. 365 p.

CHAVES, M. M.; FLEXAS, J.; PINHEIRO, C. Photosynthesis under drought and salt stress: regulation mechanisms from whole plant to cell. Annals of Botany, v. 103, p. 551-560, 2009.

MAGUIRE, D. J. Speed of germination-aid in selection and evaluation for seedling emergence and vigor. Crop Science, Madison, v. 2, n. 2, p. 176-177, 1962.

SORATTO, R. P *et al*. Resposta do feijoeiro ao preparo do solo, manejo de água e parcelamento do nitrogênio. Acta Scientiarum: Agronomy, Maringá, v. 25, n. 1, p. 89-96, 2003.

VILLELA, F. A.; DONI-FILHO, L.; SEQUEIRA, E. L. Tabela de potencial osmótico em função da concentração de polietileno glicol 6000 e da temperatura. Pesquisa Agropecuária Brasileira, Brasília, v. 26, n. 11/12, p. 1957-1968, 1991.

CHANGES ON THE NON-DIGESTIBLE CARBOHYDRATES CONCENTRATION OF COMMON BEAN IN RESPONSE TO PLANT WATER SUPPLY

Mayra Denise Herrera, Mónica Mariana Lozada Carlos, Miguel Servín Palestina

INIFAP-CEZAC. Km 24.5 Zacatecas-Fresnillo. Calera de Víctor Rosales, Zac., México. C.P. 98500.

INTRODUCTION

Common bean yields are limited by inadequate water supply during the cropping season which leads to a water-stress condition; in addition, the nutraceutical compounds concentration may be altered. Although the seed phytochemical compounds synthesis is related to defense mechanism, they are also considered as bioactive substances with health benefits. The aim of this study was to evaluate the effect of different regimes of water supply during plant growth and development, over the non-digestible carbohydrates content of the harvested seeds.

MATERIAL AND METHODS

Common bean seeds cv Dalia were sown at the INIFAP-Zacatecas Research Center under a rainout shelter to protect against rainfall; plants were subjected to different water supply regimens: 50/50 % of water availability in the vegetative/reproductive stage, as well as 50/100, 100/50 and 100/100 %. Harvested seeds were cooked and freeze-dried with its cooking water. All samples were processed in a domestic grinder and kept at 4 °C until analyses. Determination of total, soluble and insoluble dietary fiber was assessed with the Total Dietary Fiber Assay kit (Sigma-Aldrich, Saint Louis, Missouri, USA) and resistant starch was determined using the methodology described by Saura-Calixto et al. (1993).

RESULTS AND DISCUSSION

Table 1 shows the concentration of dietary fiber and its soluble and insoluble fractions. Results indicate a significant difference ($P<0.05$) among treatments; plants subjected to treatment 50/100 had the outstanding results in comparison to the remaining treatments, which indicates that plants subjected to a water restriction during exclusively the vegetative period tends to increase to production of the total dietary fiber and both its fractions.

Table 1. Content of dietary fiber in common bean obtained from plants subjected to different water supply regimes.

Compound (g/100 g)	Treatment			
	50/50	50/100	100/50	100/100
Total dietary fiber	36.3 ± 1.5 b	41.8 ± 1.9 a	37.1 ± 0.4 b	37.4 ± 0.2 b
Insoluble ditary fiber	27.2 ± 0.2 d	29.7 ± 0.2 a	29.1 ± 0.4 b	28.3 ± 0.3 c
Soluble dietary fiber	9.1 ± 1.2 bc	12.1 ± 1.8 a	7.9 ± 0.4 c	9.1 ± 0.5 b

Different letters in same row indicates significant difference ($P <0.05$) with Tukey's test.

Drought stress conditions during plant grown and development induced a higher synthesis of non-digestible carbohydrates. Accordingly, different studies have linked the incidence of water stress with a higher concentration of total fiber in diverse crops (Zhou et al., 2014). In terms of health benefits, both fibers complement each other, and their beneficial effect depends on the proportions of each fraction. A 70:30 (insoluble:soluble) ratio is considered a well-balanced proportion, and the same treatment (50/100) lead to a proportion similar to the recommended, with 71:29, whereas

treatment 100/50 lead to a proportion of 78:22, and with treatments 50/50 and 100/100, a proportion of 75:25 was obtained.

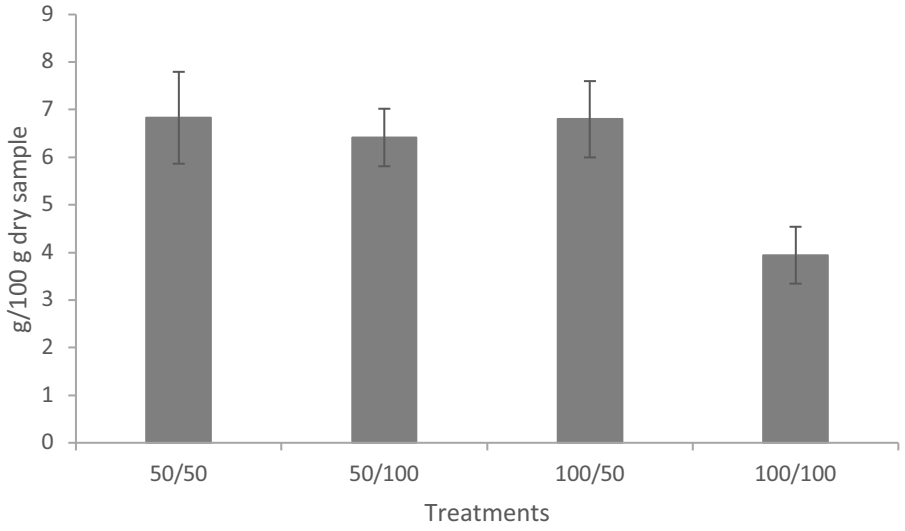


Figure 1. Content of resistant starch in common bean obtained from plants subjected to different water supply regimes.

In regard to the content of resistant starch, some authors suggest that its consumption decreases blood glucose, triglycerides and LDL cholesterol, and the resulting short-chain fatty acids of its fermentation play an important role in the prevention of colorectal and colon carcinoma (Le-Leu et al., 2007). Figure 1 shows no significant differences between treatments that involve a restriction of water supply, moreover, seeds of plants that were full-irrigated during the whole crop cycle had the lowest concentration of resistant starch. It may be suggested that the restriction of water supply in one or both periods of the common bean developmental cycle increases the concentration of non-digestible carbohydrates, thus functional quality of this products might be enhanced.

REFERENCES:

Le-Leu RK, Brown IL, Hu Y, Morita T, Esterman A and Young, GP. Effect of dietary resistant starch and protein on colonic fermentation and intestinal tumorigenesis in rats. Carcinogenesis. 28: 240-245 (2007).

Saura-Calixto F, Serrano J, Goñi I. Intake and bioaccessibility of total polyphenols in a whole diet. Food Chem. 101: 495-501 (2007).

Zhou, M., Sun, G., Zhanmin, S., Tang, Y. & Wu, Y. 2014. Cotton proteomics for deciphering the mechanism of environment stress response and fiber development. Journal of Proteomics 105: 74-84.

DIFERENTIAL CHANGES ON THE PHENOLIC CONTENT OF COMMON BEAN GROWN UNDER WATER RESTRICTION

Mayra Denise Herrera, Mónica Mariana Lozada Carlos, Miguel Servín Palestina

INIFAP-CEZAC. Km 24.5 Zacatecas-Fresnillo. Calera de Víctor Rosales, Zac., México. C.P. 98500.

INTRODUCTION:

Besides their nutritional importance, the common bean also serve as a rich source of bioactive substances, including phenolic compounds that play metabolic roles in human organism by its health-promoting effects in relation to the prevention of chronic diseases including cancers, cardiovascular diseases, obesity and diabetes. However, the quality of bean seed reaching the consumer depends on the characteristics of the seed at harvest time, which are determined by biotic and abiotic environmental factors such as drought. Therefore, the aim of this study was to subject common bean plants to different levels of soil moisture availability to determine its effect on nutraceutical seed constituents such as polyphenols.

MATERIAL AND METHODS:

Common bean seeds cv Dalia were sown under a rainout shelter and plants were subjected to different water supply regimens. According to water supply, four treatments were tasted base on soil water availability during the vegetative/reproductive stage of the whole crop cycle (50/50, 50/100, 100/50, 100/100 %). Seeds from plants subjected to the experimental treatments were cooked and freeze-dried with its cooking water. All samples were processed in a domestic grinder and kept at 4 °C until analyses. Extraction of phenolic compounds was assessed as described by Xu et al. (2007) and content of total phenols (mg GAE/g), total flavonoids (mg CAE/g), condensed tannins (mg CAE/g) and anthocyanins (mg C3GE/g) was determined.

RESULTS AND DISCUSSION:

The phenolic compounds content among treatments is shown in Figure 1. It results interesting that the concentration of total phenols (Figure 1A) and flavonoids (Figure 1B) is slightly higher in seeds from plants grown under the 50/50 treatment, although its content of anthocyanins and proanthocyanins (condensed tannins) were lower due to the stress caused by this treatment (Figure 1C and D). Results suggest that the phenylpropanoids biosynthetic pathway react differently among its different branches, while a type of proanthocyanins such as condensed tannins, tends towards a considerable diminution, total flavonoids quantified in this study tend to increase. In this regard, André et al. (2009), mentions that drought induces changes of the expression of PAL gene from the core phenylpropanoid pathway; likewise, genes HCT and C3H from the chlorogenic acid pathway are regulated, mentioning that the polyphenol content under normal and drought stress conditions may be correlated with the expression of these genes.

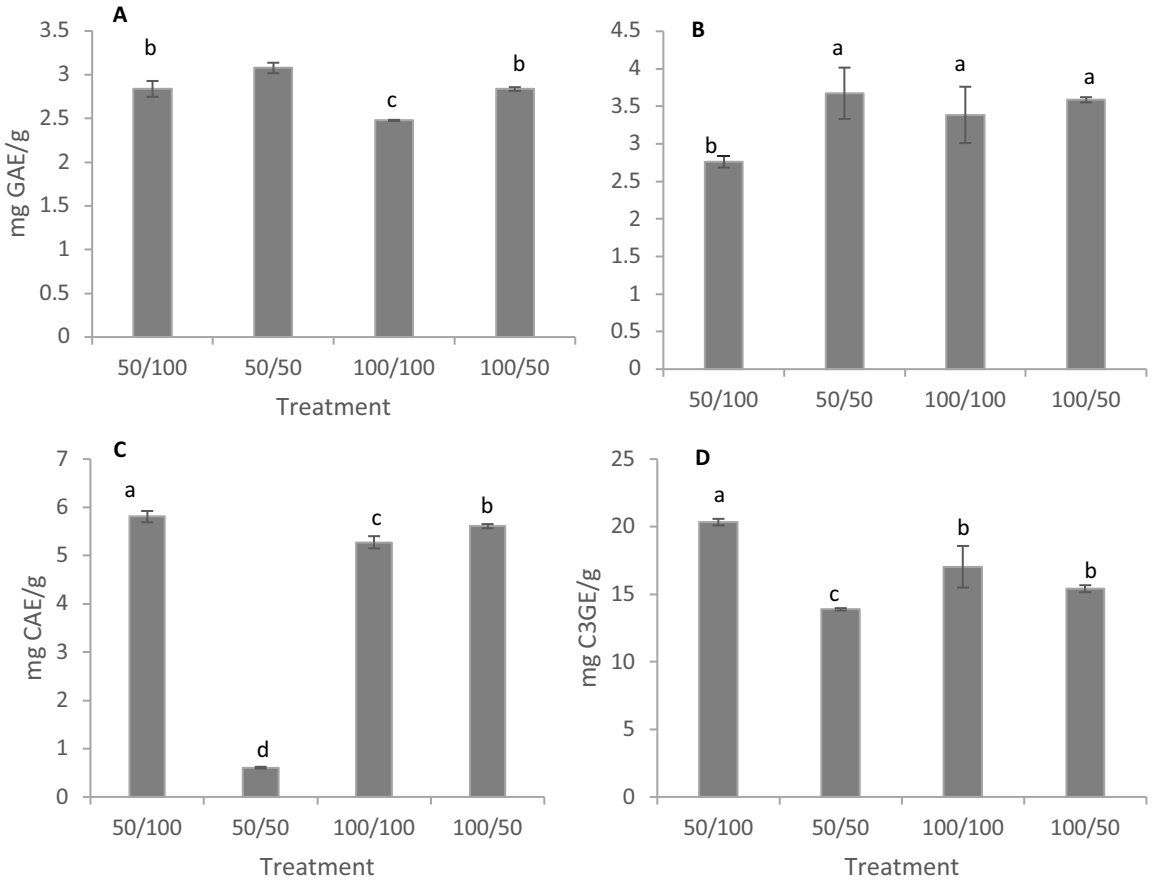


Figure 1. Polyphenol concentration on common bean from plants grown under different water supply regimes. A, total phenols; B, total flavonoids; C, condensed tannins; D, total anthocyanins. Since the phenolic compounds are considered antioxidants, and in fact, the most abundant in our diet, some authors attribute to their biological properties the prevention of chronic degenerative diseases such as cancer, cardiovascular disease and diabetes (Saura-Calixto et al., 2007). The results indicate that a water limitation during bean seed development may result in higher production and synthesis of different phenolic compounds.

REFERENCES:

André, C.M., Schafleitner, R., Legay, E., Lefévre, I., Alvarado, A.A., Norberto, G., Hoffman, L., Hausman, J-F., Larondelle, Y. & Evers, D. 2009. Gene expression changes related to the production of phenolic compounds in potato tubers grown under drought stress. Phytochemistry 70:1107-1116.

Saura-Calixto F, Serrano J, Goñi I. Intake and bioaccessibility of total polyphenols in a whole diet. Food Chem. 101: 495-501 (2007).

Xu BJ, Chang SKC. 2007. A comparative study on phenolic profiles and antioxidant activities of legumes as affected by extraction solvents. Journal of Food Science. 2: 159-166.

DIFFERENCES IN RD22 AND CA1 GENE EXPRESSION BETWEEN DELAWARE AND NEBRASKA

Isaac Fisher[1], Carlos Urrea[2], Venu Kalavacharla[1]

[1]Delaware State University-College of Agriculture and Natural Resources, Dover DE,
[2]University of Nebraska at Lincoln, Panhandle Research and Extension Center, Scottsbluff NE

INTRODUCTION: Common bean is one of the most important legumes produced worldwide, but under drought stress both quality and quantity can be reduced (Beebe et al. 2008). A deeper understanding of genes involved in drought-response can aid breeders who aim to combat this abiotic stress. Our experiment wanted to see if there would be differences in gene activity between two different states. The two genes being investigated were RD22 (Phvul.009G105300) and CA1 (EC997016). RD22 was one of the first studied drought responsive genes, has commonly been used to assess drought stress in many different crops and acts to restrict the degradation of chlorophyll under moisture stress (Xu 2010, Harshavardhan 2014,). CA1 is a transcription factor that responds to ethylene (Kavar 2007).

MATERIAL AND METHODS:
A developmental line, NE28-15-16 (bred by Dr. Carlos Urrea[2]), was grown in two different states, Delaware and Nebraska, during the same year. Both locations had a fully irrigated plot as well as a plot that received terminal drought stress. Leaf and root tissues were harvested at the same stage of development (R7) and were flash-frozen in liquid nitrogen. RNA isolation, cDNA synthesis, and then reverse-transcriptase PCR was conducted on previously stated genes. Actin 11 (KF569629) was used as a housekeeping gene to check for normalized expression. Monthly temperature and rainfall averages were acquired from NCDC.NOOA (Fig. 1/2).

RESULTS AND DISCUSSION:
Both genes showed varying transcription levels between the two states. RD22 had a higher expression in Nebraska's drought-treated leaf (LD) as well as control leaf (ND) compared to Delaware samples (Fig. 3) (Fig. 4), but drought-treated root (RD) in Delaware showed a much higher increase, while the control root sample (RN) in Delaware showed no expression at all. CA1 expression followed a similar pattern for the Nebraska leaf samples (Fig.5), but the Nebraska drought-treated root showed a much higher increase as opposed to Delaware samples (Fig. 6). It is interesting that in Delaware RD22 expressed higher in roots under stress, but CA1 expressed higher in Nebraska roots under stress. There must be other underlying factors in the locations affecting the expression of these genes (Elevation, temperatures, and differing rainfall).

REFERENCES:
- Beebe SE, et. all (2008) Selection for drought resistance in common bean also improves yield in phosphorus limited and favorable environments. Crop Sci 48:582–592
- Harshavardhan VT, et. all (2014) AtRD22 and AtUSPL1, members of the plant-specific BURP domain family involved in Arabidopsis thaliana drought tolerance. PLoS ONE 9(10):e110065
- Hongliang Xu, et. all (2010) Genome-scale identification of Soybean BURP domain-containing genes and their expression under stress treatments
- Tatjana Kavar et. all (2008) Identification of genes involved in the response of leaves of Phaseolus vulgaris to drought stress

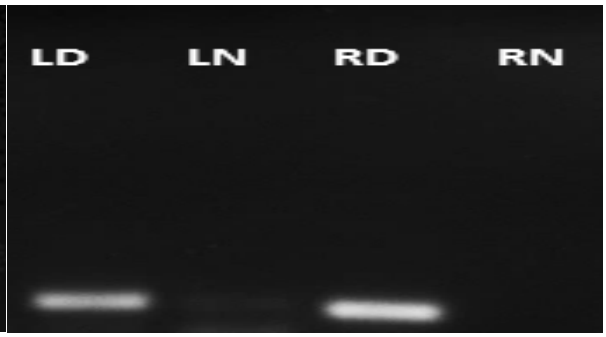
Location	June	July	August	September	Average
Smyrna, DE	73 F	80 F	80 F	73 F	77 F
Scottsbluff, NE	74 F	76 F	71 F	64 F	71 F

(Figure 1: Average temperatures during growth)

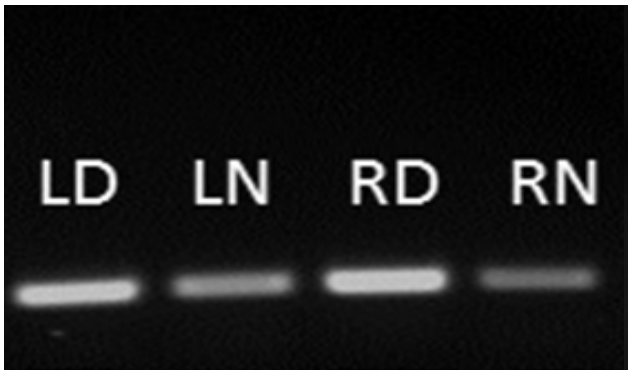
Location	June	July	August	September	Average
Smyrna, DE	104.14mm	190.5	71.12	261.62	629.92
Scottsbluff, NE	27.94	33.02	48.26	35.56	142.24

(Figure 2: Average rainfall during growth, in millimeters)

(Figure 3: RD22 Nebraska samples)



(Figure 4: RD22 Delaware samples)



(Figure 5: CA1 Nebraska samples)

(Figure 6: CA1 Delaware samples)

HEAT TOLERANCE OF COMMON BEAN LINES IN HONDURAS

Juan C. Rosas[1], James S. Beaver[2], Timothy G. Porch[3], Stephen E. Beebe[4], Jonathan P. Lynch and James Burridge[5]

[1] Escuela Agricola Panamericana, Zamorano, P.O. Box 93, Tegucigalpa, Honduras
[2] Dept. of Agroenvironmental Sciences, University of Puerto Rico, Mayagüez, PR 00680
[3] USDA-ARS-TARS, 2200 P.A. Campus Ave., Suite 201, Mayagüez, PR 00680
[4] Centro Internacional de Agricultura Tropical (CIAT), A.A. 67-13, Cali, Colombia
[5]Pennsylvania State University, 102 Tyson Bldg., University Park, PA

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) production in Central America is threaten by increases in temperature caused by climate change. Traditional bean cultivars are more affected due to their lack of adaptation to higher temperatures. Improved bean cultivars released during the past 20 years are better adapted to warmer conditions in the tropical lowlands, but overtime could lose adaptation due to continued increases in temperature. Germplasm accessions and breeding lines were tested in high temperature locations to identify genotypes with potential for release as cultivars or use as breeding parents.

MATERIALS AND METHODS

More than 300 Andean and Mesoamerican cultivars and breeding lines were evaluated under high day/night temperatures in the Nacaome valley of southern Honduras. One-hundred twenty lines BASE (Bean Adaptation Stress Environment) trials were planted in a lattice design with three replications and 24 lines from the ERSAT (acronyms in Spanish for Heat Tolerant Regional Bean Trial) were planted using a randomized complete block design with three replications. Experimental units were 2.5 m long x 1.2 m wide raised beds planted with two rows of bean plants separated by 40 cm. Plants were fertilized with 130 kg/ha of 18-46-0 at planting and 65 kg/ha of urea at 25 days after planting. Insect pests and diseases were controlled with chemical pesticides and weed populations were managed with herbicides before planting and at the pre-flowering stage. Adequate soil moisture during the trials were maintained using drip irrigation. Phenological data including days to flowering, pod formation, pod filling and physiological maturity were recorded. Temperature and relative humidity during the trials were registered using *IButtons*® (Hygrochron DS-1923) sensors. At the pod filling stage, five plants samples were taken and dried in an oven at 70°C x 48 h to determine plant biomass (BDW) and pods dry weights (PDW) and to estimate pod partition index (PPI) using the formula PPI= PDW/(BDW + PDW). At harvest maturity, 10 plants samples were taken and dried to determine seed dry weight (SDW) and PDW and to estimate harvest index (HI) using the formula HI= SDW/(SDW + PDW). Seed yield and seed weight (100 SDW) were also determined.

RESULTS AND DISCUSSION

Temperatures during 2015-16 field trials were higher than those normally present in most bean production areas in Central America and adequate for testing the tolerance to heat in common beans (Table 1). In general, high temperatures caused abortion of flowers and small pods, and extended the flowering period in sensitive genotypes, which then produced few pods with a fewer number of smaller seeds or no seeds at all. Tolerant genotypes, on the other hand, set pods and seeds at normal maturity and produced good yields under heat stress conditions.

Table 1. Maximum and minimum average (range) temperatures during the high temperature field trials. Nacaome, Honduras, 2015-16.

Planting month and year[z]	Average and range temperatures (°C) Maximum	Minimum
February 2015	39.1 (29.2- 42.6)	24.2 (20.7- 28.2)
June 2015	37.9 (36.9- 41.0)	23.0 (18.0- 28.0)
October 2015	36.8 (31.1- 40.6)	22.2 (17.6- 25.1)
February 2016	37.8 (31.6- 43.6)	22.9 (17.1- 27.6)
June 2016	39.3 (32.6- 43.6)	23.3 (20.6- 25.6)
November 2016	37.9 (31.3- 42.6)	20.4 (15.6- 26.6)

[z] Seasons: Summer (Feb-April), First (June-August) and Second (October-December).

The range of seed yields of common bean and tepary check lines evaluated at the Nacaome site during 2015-16 are presented in Table 2. As expected, tepary lines performed well under higher temperature environments. A small group of common bean lines also showed good adaptation to heat stress and several of these lines possess good agronomic adaptation, disease resistance and good commercial seed types. The small red bean FBN1211-66 and the white bean Beniquez had superior performance across planting dates and years. The pink stripped landrace Indeterminate Jamaica Red was the most heat tolerant Andean bean in the trials. Heat stress was more severe during the June planting dates resulting in lower yields than in other seasons. Commercial production in lowland, warmer regions of Central America is currently feasible due to heat tolerant genotypes such as SJC730-79 which was recently released as a cultivar in El Salvador.

Table 2. Seed yield (kg/ha) of common bean and tepary lines from BASE and ERSAT trials conducted under high temperatures. Nacaome, Honduras 2015-16.

Trial	Planting date	Range of seed yield (kg/ha)	Promising lines[z]
BASE 2014	February 2015	0 to >4,000	TARS-Tep22, 23, 29 & 30[y], USMR20, IJR, SB-DT1, Beniquez, SEF15, Matterhorn, H9657-27-10, INB841.
BASE 2015	February 2016	0 to >4,000	G40001[y], INB 841, PR9920-171, TARS-Tep22, IJR, SER78, Cedron, SB-DT1, Cardenal, ICTAZAM, DEORHO, Morales.
ERSAT 1 and others	June 2015	0 to >1,000	TARS-Tep22, MER2212-28, Beniquez, IJR, MEN934-28, SJC730-79, BIOF4-70, FBN1211-66.
ERSAT 2	October 2015	60 to 1,887	SJC730-79, TARS-Tep22, MHR311-17, MER2212-28, FBN1211-66, ALS0532-6, BRT103-182, SB-DT1.
ERSAT 2	February 2016	150 to 2,989	TARS-Tep 22, FBN1211-66, SJC730-79, MEN934-68, Beniquez, IJR, SB-DT1, MEN2212-28.
ERSAT 3	June 2016	0 to 1,202	TARS-Tep22, Cedron, ICTAZAM, PR9920-171, Beniquez, Amadeus77, FBN1211-66, MER2212-28.
ERSAT 3	November 2016	1,066 to 3,084	FBN1211-66, BIOF2-106, Morales, MHR311-17, Beniquez, SER78, SB793, DEORHO.

[z]Top 10% in seed yield from BASE and top 30% from ERSAT trials.
[y] Tepary (*Phaseolus acutifolius* L.) lines.

NON-PREFERENCE FOR OVIPOSITION OF BEAN GENOTYPES TO *Tetranychus urticae* KOCH (ACARI: TETRANYCHIDAE)

C. C. Melville[1], L. Nogueira[1], Oliveira. N.T[2], A. L. Boiça Júnior[1] Daniel J. A[1]

[1]Universidade Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias, Departamento de Fitossanidade, Laboratório de Acarologia Agrícola, 14884-900, Jaboticabal, SP, Brazil.
[2]Universidade Federal de Lavras, Programa de Pós-Graduação em Agronomia/Fitotecnia, 37200-000, Lavras, MG, Brazil.
E-mail: ciranomelville@outlook.com

INTRODUCTION

Various pests affect common bean production, among wich the two-spotted spider mite, *Tetranychus urticae* Koch, is one of the most important phytophagous in many bean-growing areas around the world (Zhang, 2003). Therefore, considering the increasing difficulty to control mites by acaricides, it would be important to recognize bean genotypes that can affect the development and survival or at least delay outbreaks of *T. urticae*.

Host plant resistance are powerful tool for analyzing and understanding the adverse effects of the plant on the arthropod. This control measure holds some advantages over the other methods, as it does not require sophisticated technology by farmers for use, does not interfere with the environment, and has persistent, cumulative, and non-polluting effects (Boiça Júnior et al., 2013). The objective of this study was to evaluate non-preference for oviposition in common bean genotypes to the two-spotted spider mite.

MATERIALS AND METHODS

The experiment was conducted in the Laboratório de Acarologia of the Departamento de Fitossanidade, FCAV/UNESP, São Paulo, Brazil, under controlled conditions of 25 ± 2 ºC temperature, 70 ± 10% relative humidity, and 12 h photophase. The following common bean genotypes were tested: BRS-Supremo, BRS Pérola, BRS Talismã, IAC Harmonia and IPR Campos Gerais. Seeds of the common bean genotypes were sown in 5-L-pots containing a mixture of soil, manure, and sand at 2:1:1 ratio, and were kept in a greenhouse until use. Plants of common beans were used at the vegetative stage V3-V4.The preference oviposition by *T. urticae* in free-choice assay was designed in randomized blocks and, no-choice test completely randomized, with 10 replications each.

In the free-choice each replication consisted of a Petri dish arena (15 cm diameter x 1 cm height) containing the leaf discs (2.5 cm diameter) of each genotype distributed equidistantly. To prevent escape of mites and maintenance of moisture, the margins of Petri dishes were surrounded with water-soaked cotton and then covered with plastic film. Thereafter, 25 newly emerged *T. urticae* female were released in the ratio of five females to each genotype.

For the no-choice test, the arena (10 replication) consisted of a Petri dish (6 cm diameter x 1 cm height) with an insect pin fixed in the center, where a supernatant leaf disc (2.5 cm diameter) was placed in water attached through the insect pin in order to avoid leaf movement and mite scape. Then, five newly emerged *T. urticae* female were released in each Petri dish arena. In both assays, preference for oviposition of *T. urticae* was recorded 24 h and 48 h after the female were released.

Data recorded from oviposition preference trials were analyzed for residuals normality and variance homogeneity, and when necessary, were transformed to meet the assumptions of analysis

of variance (ANOVA). Next, data were subjected to analysis of ANOVA, and means were compared by Tukey's test (P<0.05).

RESULTS AND DISCUSSION

There were not significant differences in the oviposition preference of *T. urticae* among the evaluated commom bean genotypes only the in free-choice test (Tabela 1). However, there was a trend of higher number of eggs on the BRS Pérola, and numerically the lowest mean of eggs were observed on IAC Harmonia.

For no-choice test, differences were observed in time of 48 h and total number of eggs. The genotype IPR Campos Gerais highlighted as the least preferred when female of twospotted spider mite were given no-choice for oviposition. On the other hand, the genotype BRS Pérola were the most preferred for oviposition, with 0.51 (48 h) and 1.77 (total) eggs per hour (Table 1). We may infer this cultivar holds traits acting as oviposition stimulant for the twospotted spider mite.

Tabela 1. Mean number of eggs (±SE) of *Tetranychus urticae* in leaf discs on five bean genotypes in free-choice test and no-choice tests. Jaboticabal, SP, Brazil, 2017.

Genotypes	Number of eggs leaf discs					
	Free-choice[1]			No-choice[1]		
	24 h	48 h	Total	24 h	48 h	Total
IPR Campos Gerais	31.30±7.00	29.60.±5.70	60.90±13.16	22.10±3.08	24.70±5.31 b	46.80±7.08 b
IAC Harmonia	24.60±4.56	26.90±4.13	51.50±8.43	34.70±4.20	31.10±3.74 ab	65.80±7.33 ab
BRS Supremo	31.90±6.60	38.50±8.00	70.40±12.71	28.60±3.85	36.00±7.36 ab	64.60±10.22 ab
BRS Talismã	26.10±6.22	33.10±9.62	59.20±14.63	34.30±4.66	37.20±4.35 ab	71.50±6.48 ab
BRS Pérola	43.40±4.78	52.70±8.98	96.10±12.67	33.60±2.57	51.50±6.80 a	85.10±6.40 a
F	1.39^{NS}	1.54^{NS}	1.69^{NS}	2.06^{NS}	3.05*	3.24*
P	0.2577	0.2116	0.1741	0.1020	0.0264	0.0205

[1] Means followed by the letter in column are not significantly difference by Tukey's test, at 5% probability. (±SE) Standard error of mean.

The results of this study demonstrate variation in mite performance on different genotypes of the same crop. An important goal of future research will be to elucidate the possible defense mechanism present in the genotype IPR Campos Gerais to twospotted spider mite, aiming to transfer or increase these traits resistance to other genotypes.

REFERENCES

BOIÇA JÚNIOR, A. L.; SOUZA, B. H. S.; LOPES, G. S.; COSTA, E. N.; MORAES, R. F. O.; EDUARDO, W. I. Atualidades em resistência de plantas a insetos. In: BUSOLI, A. C.; ALENCAR, J. R. D. C. C.; FRAGA, D. F.; SOUZA, L. A.; SOUZA, B. H. S.; GRIGOLLI, J. F. J. (Eds.). Tópicos em entomologia agrícola – VI. Jaboticabal: Gráfica Multipress, 2013. p. 207-224.

ZHANG, Z. 2003. Ecological and agricultural consideration in the management of two-spotted spider mite (*Tetranychus urticae* Koch). *Agricultural Zoology Reviews,* 2:185-236.

ATTRACTIVENESS AND INJURY OF *Phaseolus vulgaris* L. GENOTYPES BY *Anticarsia gemmatalis* HÜBNER (LEPIDOPTERA: EREBIDAE)

J. G. E. A. R. Aiala[1], L. Nogueira[1*], G. A. P. Bernardes[1], C. C. Melville[2], N. T. Oliveira[3], T. L. P. O. Souza[4]

[1] Univ. Estadual de Goiás, UEG, Laboratório de Entomologia Agrícola, Rod. GO 330, Km 241, s/n°, Ipameri, GO, 75780-000, Brazil; [2]Univ. Est. Paulista, UNESP/FCAV, Via de Acesso Prof. Paulo Donato Castellane s/nº, Jaboticabal, SP, 14884-900, Brazil; [3]Univ. Federal de Lavras, Lavras, MG, 37200-000, Brazil; [4]Embrapa Arroz e Feijão, Santo Antônio de Goiás, GO, 75375-000, Brazil; *corresponding author: lucianonogueiraagro@gmail.com

INTRODUCTION

The *Anticarsia gemmatalis* Hübner (Lepidoptera: Erebidae) is the most defoliator pest, causing economic damage to many species of crop plants, including preferentially, soybeans – [*Glycine max* (L.) Merrill] and common bean - *Phaseoulus vulgaris* L. (Herzog; Todd, 1980; Panizzi et al., 2004). These crops are extensively cultivated in Brazil, thus forming very simplified and vulnerable agroecosystems, resulting in problems such as the excessive and indiscriminate use of pesticides, with adverse ecological consequences to the environment. In an attempt to diminish pesticide use in cropping systems, alternative control methods have been investigated, and host plant resistance is one of them (Boiça Júnior et al., 2014). Searching genotypes that express tolerance and/or resistance to insect pest represents an important step for plant breeding.

Thus, the aim of this study we evaluated the attractiveness and the leaf injury caused by *A. gemmatalis* larvae in bean genotypes.

MATERIAL AND METHODS

Assays were conducted in the agricultural entomology Laboratory, UEG, Ipameri, GO, Brazil, under environmentally controlled conditions. The following common bean genotypes were evaluated for resistance to *A. gemmatalis* larvae: BRS Estilo, BRS Requinte, BRS Ametista, BRSMG Realce e BRS FC402.

The experiment consisted of free-choice and no-choice assays setup in completely randomized design with 10 replications each. For the free-choice test, leaf disks (2.2 cm diameter) prepared from the respective genotypes were arranged in glass arenas (14.0 cm diameter) lined with moistened filter paper, where two neonate larvae *A. gemmatalis* was released per genotype, totaling 10 larvae per arena. In the no-choice test, one leaf disk was placed in each Petri dish (9.0 cm diameter) two neonate larvae. In both tests, leaf disk attractiveness was recorded 72 h after the larvae were released. To evaluate leaf injury caused by the larvae, percentage of estimated injury ranging from 0 to 100% was assigned for uninjured leaf discs and fully injured leaf discs, respectively.

Data recorded from leaf disc attractiveness percentage and leaf injury percentage were transformed into arcsine $(x/100)^{1/2}$. Next, data were subjected to analyzed for normality (Levene's test) and variance homogeneity (Shapiro-Wilk test). Data were subjected to analysis of variance (ANOVA), and means were compared by Tukey's test ($P < 0.05$).

RESULTS AND DISCUSSION

The bean genotypes evaluated interfered in attractiveness and injuries caused by *A. gemmatalis* larvae. In the free-choice test, the percentage of *A. gemmatalis* larvae attracted was significantly lower for BRSMG Realce genotype when compared to the others; no difference was found among

other tested genotypes (Fig. 1A). The lowest percentages of injury were verified in the BRSMG Realce and BRS Estilo. The highest injuries were observed in the BRS Requinte genotype (Fig. 1B).

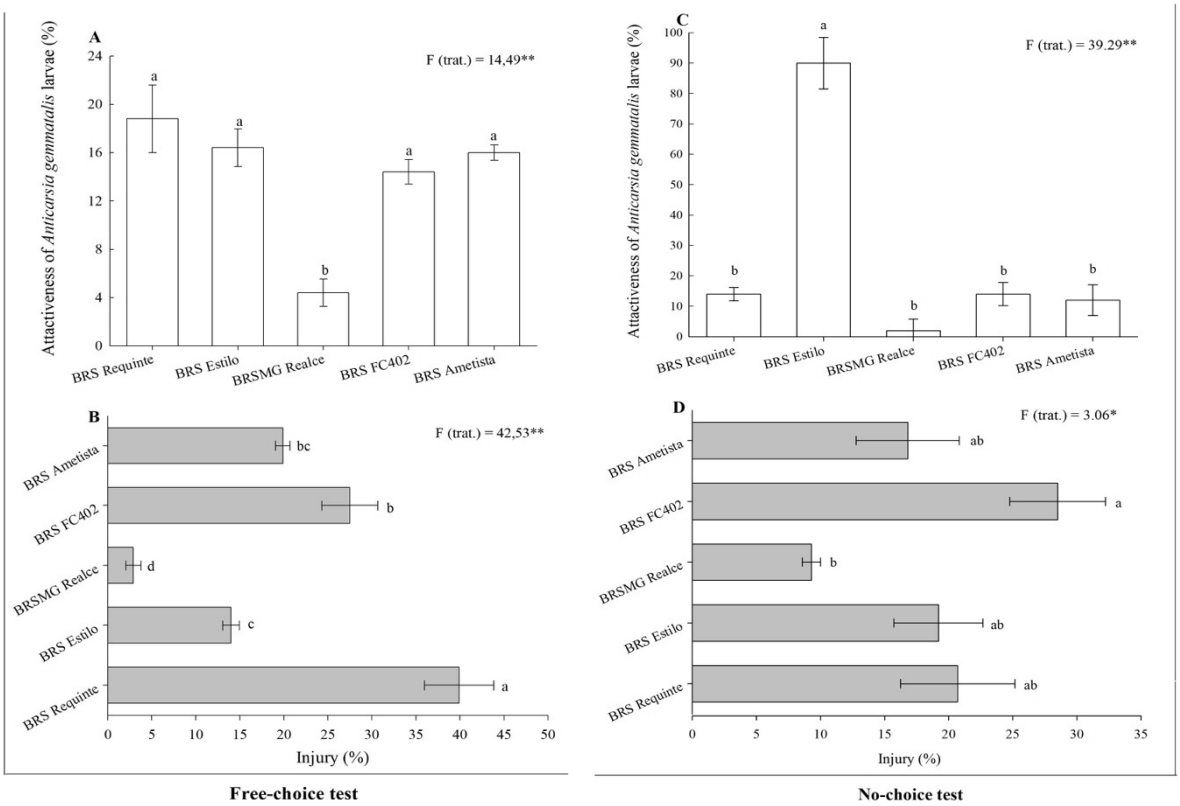


Figure 1 Attractiviness and injury percentage of common bean genotypes to larvae *Anticarsia gemmatalis* in free-choice (**A**, **B**) and no-choice test (**C**, **D**). Bars with diferents letters are significantly different by Tukey test at ** 1% and *5% probability.

In the no-choice test, the percentage of larvae attracted was highest in the BRS Estilo genotype (Fig. 1C). The BRSMG Realce was the least injured of *A. gemmatalis* (Fig. 1D).

The results illustrate that there was a direct relationship between the lower attractiveness of BRSMG Realce to the larvae and the lower injury percentage recorded in this genotype (Fig. 1). This leads us to infer that the genotype BRS Realce has compounds that may have inhibited attractiveness and consequently the feeding of the larvae. In addition, the lower preference for the BRSMG Realce may be linked to physical causes (leaf color). The leaves of this genotype have a light green color, while the others have dark green pattern. Additionally, we cannot rule out morphological factors (presence of trichomes) and the possible effect of chemical substances (volatile) that may have acted as deterrents to larvae feeding preference.

This study shows that BRSMG Realce genotype presents characteristics resistance in the non-preference category for feeding to *A. gemmatalis*.

REFERENCES

BOIÇA JÚNIOR, A. L., et al. Feeding preference and development of *Cerotoma arcuata* (Olivier) (Coleoptera: Chrysomelidae) on soybean. *Pesquisa Agropecuária Tropical*, 44: 238-245. 2014.

HERZOG, D. C.; TODD, J. H. Sampling velvetbean carterpillar on soybean. In: KOGAN, M. et al. *Sampling methods in soybean entomology*. New York: Springer-Verlag, 1980. p. 107-140.

PANIZZI, A. R. et al. Survivorship, larval development and pupal weight of *Anticarsia gemmatalis* (Hübner) (Lepidoptera: Noctuidae) feeding on potential leguminous host plants. *Neotropical Entomology* 33: 563-567. 2004.

FACTORS INFLUENCING EXPRESSION OF ANTIXENOSIS IN *Phaseolus vulgaris* L. TO *Spodoptera frugiperda* (SMITH, 1797) (LEPIDOPTERA: NOCTUIDAE)

C.A. Freitas[1*], M. M. Freitas[1], P. H. S. Barcelos[1], Carbonell, S. A. M.[2], A. L. Boiça Júnior[1]

[1]Universidade Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias (UNESP/FCAV), Via de Acesso Prof. Paulo Donato Castellane s/n, Jaboticabal, SP, 14884-900, Brazil; [2]Instituto Agronômico (IAC), Av. Barão de Itapura, 1.481, CEP 13020-902, Campinas,

INTRODUCTION

Among several control tactics in the integrated pest management, host plant resistance stands out for several advantages that it provides, mainly by acting synergistically with other control methods. Host plant resistance to insects has genetic character, however, the expression of genes conferring resistance can be affected according to the conditions under which the plant is submitted (BOIÇA JÚNIOR et al., 2015).

Factors related to plant, insect and environment can interfere in the resistance levels expression, thus it is relevant standardize the conditions in host-plant resistance bioassays, making necessary to identify and analyze previously possible factors for each insect species and to avoid errors as regard genotype selection with resistance to a pest (SMITH, 2005; BOIÇA JÚNIOR et al., 2015).

Therefore, the aim of this study was to evaluate potential factors related to the resistance expression of common bean to *Spodoptera frugiperda* (J. E. Smith, 1797) (Lepidoptera: Noctuidae), a pest that causes yield losses to several agricultural crops, among them beans.

MATERIAL AND METHODS

Bean plants were grown in 5-L polyethylene pots filled with soil, sand, and organic bovine manure at 2:1:1 ratio, being kept three plants per pot, under a greenhouse covered with anti-aphid mesh netting. When the plants reached v3 phenological stage (first fully expanded trifoliate), leaves were collected and taken to the laboratory, where the experiments were conducted under controlled environmental conditions (25 ± 2 ° C and 60 ± 10% RH).

A randomized block design was used to free-choice test and a completely randomized design for no-choice test, both under sub-subdivided plots scheme, with ten replications. The principal plots consisted by different larval densities (one and two larvae) per cultivar, the subplots by two common bean cultivars (IAC Harmonia and BRS Pérola, pattern of resistance and susceptibility, respectively, according to Souza et al., 2012) and the sub-subplots by two vegetable substrate supply forms (entire leaflet and leaf disc, with 2.5 cm diameter).

The free-choice test was performed on 23 cm diameter glass trays and the no-choice test on 9 cm diameter Petri dishes, both lined with filter paper moistened with deionized water. In free-choice test the vegetal substrates were arranged equidistantly to the center, while in no-choice test were placed individually in Petri dishes. Then second instar *S. frugiperda* larvae were released according to density described previously. After 50 hours of larvae release the leaf area consumed (AFC) by the larvae was determined through the difference between initial and final leaf area, measured by ImageJ software. The data were submitted to normality and homogeneity analysis, later the variance analysis was performed and when significant treatments means were compared by Tukey test ($\alpha = 0.05$).

RESULTS AND DISCUSSION

No significant interactions were observed between the analyzed factors, so regardless of factors evaluated, the resistance expression was not influenced. In relation to larval density, for free-choice and no-choice test the use of two larvae promoted an AFC of 63.0% and 106%, respectively, larger than the use of one larvae (Figure 1).

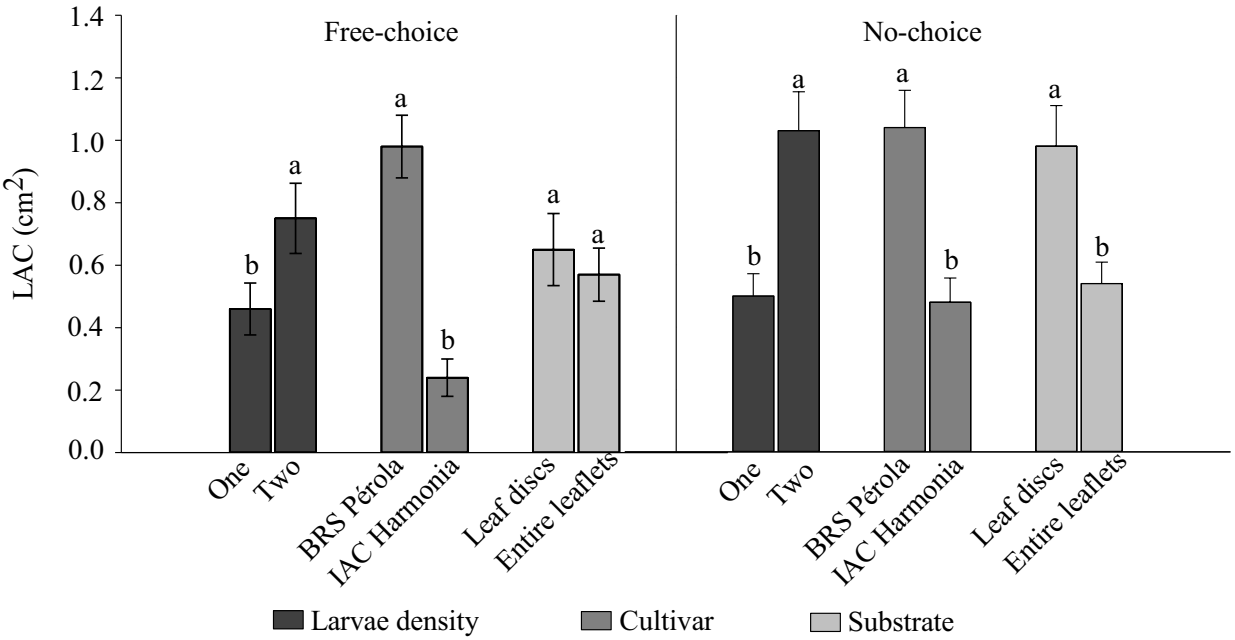


Figure 1. Leaf area consumed (AFC, cm2) by second instar larvae of *Spodoptera frugiperda* evaluating larval density (DL) and plant substrate (SV) factors on resistance expression in two bean cultivars (CV) in free-choice test (DL ($F_{1.9}$ = 17.20; P = 0.0025); CV ($F_{1.18}$ = 45.76; P <0.0001); DL x CV ($F_{1.18}$ = 1.52; P = 0.2342); SV ($F_{1.36}$ = 0.36; P = 0.5538); DL x SV ($F_{1.36}$ = 1.07; P = 0.3068); CV x SV ($F_{1.36}$ = 0.91; P = 0.3458); DL x CV x SV ($F_{1.36}$ = 0.65; P = 0.4256)) and no-choice test (DL ($F_{1.18}$ = 27.24; P < 0.0001); CV ($F_{1.18}$ = 22.21; P = 0.0002); DL x CV ($F_{1.18}$ = 1.21; P = 0.2864); SV ($F_{1.36}$ = 12.86; P = 0.0010); DL x SV ($F_{1.36}$ = 1.91; P = 0.1753); CV x SV ($F_{1.36}$ = 1.53; P = 0.2240); DL x CV x SV ($F_{1.36}$ = 1.57; P = 0.2185)).

According to figure 1, in both tests there was difference in consumption by the larvae among the two cultivars, being evidenced the resistance characteristic of cultivar IAC Harmonia in relation to BRS Pérola. Regarding the vegetal substrate for free-choice test there were no differences in AFC among leaf disc use and entire leaflets, however, in the non-choice test the AFC was 82.5% larger in the leaf discs compared to entire leaflet. Thus, the use of two second-instar larvae on leaf discs better evidence the resistance expression of bean cultivars to *S. frugiperda.*

REFERENCES

BOIÇA JÚNIOR, A. L. et al. A defesa das plantas ao ataque dos insetos. In: BUSOLI, A. C. et al. (Eds.). *Tópicos em entomologia agrícola – VIII*. Jaboticabal: Maria de Lourdes Brandel-ME. 2015. p. 161-179.

SMITH, C. M. *Plant resistance to arthropods: molecular and conventional approaches*. Dordrecht: Springer, 2005. 423 p.

SOUZA, B. H. S. et al. Non-preference for feeding of *Spodoptera frugiperda* (Smith) (Lepidoptera) in bean genotypes. *Bean Improvement Cooperative*, v. 55, p. 211-212, 2012.

ESSENTIAL OILS' EFFECT ON THE OVIPOSITION AND MORTALITY OF *Zabrotes subfasciatus* (Bohemann, 1833) (Coleoptera, Bruchinae)

Castro, I. R.[1]; Silva, F. S.[1]; Bottega, D. B.[1]; Cazal, C. M.[2]

[1]Instituto Federal de Educação, Ciência e Tecnologia Goiano, Zip Code 76200-000 - Iporá, Goiás, Brazil; [2]Instituto Federal de Educação, Ciência e Tecnologia do Sudeste de Minas, Zip Code 36205-018, Barbacena, Minas Gerais, Brazil

INTRODUCTION

Considering beans storage (*Phaseolus vulgaris* L.), the yield may be reduced due to infestations of weevils such as *Zabrotes subfasciatus* (Bohemann, 1833) (Coleoptera: Bruchinae). The attack of this insect directly affects the quality of the grains, besides facilitating the entry of pathogens, making them unviable for consumption and trade (BALDIN & PEREIRA, 2010).

The use of alternative methods such as plant extracts is a viable strategy, since it is less likely to select resistant populations in insects and it leaves no toxic residues in the environment (FERNANDES & FAVERO, 2014).

The chemical substances produced by plants, when isolated, are deadly or repellent for many species of insects (GOMES & FAVERO, 2011). The aim of this paper was to evaluate the insecticidal effect of essential oils of *Xylopia aromatica* (Lam.) Mart (Annonaceae) and *Schinus molle L.* (Anacardiaceae) on *Z. subfasciatus.*

MATERIAL AND METHODS

The experiment was conducted at the Entomology Laboratory of Instituto Federal Goiano - Campus Iporá, Goiás, Brazil. It was employed the completely randomized design with three treatments and eight repetitions constituted by circular plastic containers of approximately 0.45m in height and 0.63m in diameter. These containers were lined with filter paper and it was added 15g of beans.

The treatments were control (T1), 5µL of *X. aromatica'*s fruit essential oil (T2) and 5µL of *S. molle*'s fruit essential oil (T3). They were applied on the filter paper and, subsequently, seven couples of *Z. subfasciatus* were released in each container.

Evaluations were made 03, 06, 12, 24, 48 and 72 hours after the insects' release counting the number of live specimens in the repetition. Seven days after the end of the mortality evaluations, viable and unviable eggs were counted for the evaluation of preference for oviposition. For statistical analysis, the values resulting from the evaluation of mortality and oviposition were transformed into $\sqrt{x + 0,5}$. Data were submitted to analysis of variance and averages comparison by Tukey test at 5% of probability.

RESULTS AND DISCUSSION

The essential oils did not reach a high percentage of mortality. However, when comparing the results, *X. aromatica*'s oil reached the highest averages from the first evaluation time when compared to the control, reaching 18.75% of dead insects within 72 hours after the insects' release (Table 1). These low mortality values compare with those of Magalhães et al. (2015) who observed that the essential oil of *Myracrodruon urundeuva* caused 12.5% mortality of *Tribolium castaneum*'s adults, and mortality increased with the increase of the tested concentrations.

In oviposition's evaluation, treatment with *X. aromatica*'s essential oil showed the lowest averages of *Z. subfasciatus*'s viable eggs, differing statistically from the results obtained in the

control (Table 2). Castro et al. (2010) verified the inhibition of weevil's oviposition *(Callosobruchus maculatus)* in treatments with plant powders of *Piper tuberculatum, Lippia sidoides* and *Sapindus saponaria.*

Table 1. Mortality (%) of *Zabrotes subfasciatus*'s adults in beans treated with essential oils of *Xylopia aromatica* and *Schinus molle*'s at different evaluation times. Iporá-GO, 2017.

Treatment	Time (hours)[1]					
	03	06	12	24	48	72
Control	0 a	0 a	0 a	1,78 a	2,67 a	4,46 a
X. aromatica	11,60 b	14,28 b	15,17 b	16,07 b	17,85 b	18,75 b
S. molle	0 a	1,78 a	4,46 ab	5,35 ab	7,14 ab	7,14 ab
P	0,01*	0,00**	0,00**	0,01*	0,01*	0,02*
F	5,59	9,25	6,57	5,03	5,19	4,22
C.V. (%)	45,17	37,90	41,63	39,93	38,30	38,28

Averages folloed by the same letter in the column do not differ statistically from one another by the Tukey test. ** = significant at 1% * significant at 5%. [1]Data transformed to $\sqrt{x + 0,5}$.

Table 2. Effect of essential oils application on the oviposition of *Zabrotes subfasciatus*'s adults. Iporá-GO, 2017.

Treatment	Eggs[1]	
	Viable	Unviable
Control	138 b	97,50 a
X. aromatica	78,50 a	75,62 a
S. molle	106,75 ab	98,50 a
P	0,00**	0,06ns
F	8,74	3,07
C.V. (%)	14,53	12,32

Averages followed by the same letter in the column do not differ statistically from one another by the Tukey test. ** = significant at 1%. ns = non significant [1]Data transformed to $\sqrt{x + 0,5}$.

CONCLUSION

The dose of 5 µL of the *X. aromatica*'s fruit essential oil presents inhibition on viable eggs' oviposition and insecticidal effect on *Z. subfasciatus* under laboratory conditions.

REFERENCES

Baldin, E.L.L.; Pereira, J.M. Resistência De Genótipos De Feijoeiro A *Zabrotes Subfasciatus* (Bohemann, 1833) (Coleoptera: Bruchidae). Rev. Ciênc. Agrotec., V.34, N.6, P.1507-1513, 2010.

Castro, M.J.P.; Silva, P.H.S.; Santos, J.R.; Silva, J.A.L. Effect Of Vegetal Powders On Oviposition Of *Callosobruchus Maculatus* (Fabr.) (Coleoptera: Bruchidae) On Cowpea. Sociedade Entomológica Do Brasil, V.5, N.4, 2010.

Fernandes, E.T.; Favero, S. Óleo Essencial De *Schinus Molle L.* Para O Controle De *Sitophilus Zeamais* Most.1855 (Coleoptera: Curculionidae) Em Milho. Rev. Bras. De Agroecologia, V.9, N.1, P.225-231, 2014.

Gomes, S.P.; Favero, S. Avaliação De Óleos Essenciais De Plantas Aromáticas Com Atividade Inseticida Em *Triatoma Infestans* (Klug, 1834) (Hemiptera: Reduviidae). Acta Scientiarum. Health Sciences, V.33, N.2, P.147-151, 2011.

Magalhães, C.R.I.; Oliveira, C.R.F.; Matos, C.H.C.; Brito, S.S.S.; Magalhães, T.A.; Ferraz, M.S.S. Potencial Inseticida De Óleos Essenciais Sobre *Tribolium Castaneum* Em Milho Armazenado. Rev. Bras. Pl. Med., V.17, N.4, P.1150-1158, 2015.

FEEDING PREFERENCE OF *Spodoptera cosmioides* (WALKER) (LEPIDOPTERA: NOCTUIDAE) BY BEAN GENOTYPES

L. Nogueira[1*], M. M. Freitas[2], C. A. Freitas[2], P. H. S. Barcelos[2], A. L. Boiça Júnior[2]

[1] Univ. Estadual de Goiás – Câmpus Ipameri, Laboratório de Entomologia Agrícola, Rodovia GO 330, Km 241, s/n°, Ipameri, GO, 75780-000, Brazil; [2]Univ. Estadual Paulista, UNESP/FCAV, Laboratório de Resistência de Plantas à Insetos, Via de Acesso Prof. Paulo Donato Castellane s/nº, Jaboticabal, SP, 14884-900, Brazil; [*]corresponding author: lucianonogueiraagro@gmail.com

INTRODUCTION

The black armyworm, *Spodoptera cosmioides* (Walker) (Lepidoptera: Noctuidae) is a polyphagous pest of crops and spontaneous plants throughout the Americas (Bavaresco et al., 2004). In Brazil, some of its hosts include bean, soybean, pea, cotton and sunflower. Their occurrence is generally related to the use of non-selective insecticides, resulting in imbalances in the agroecosystem and as a consequence, for example, leads to the reduction of natural biological control agents.

However, alternative and sustainable tactics of pest control should be envisaged. The use of resistant cultivars presents desirable characteristics, including specificity for one or more pests, cumulative and persistent effect, diminishes production cost and the possibility of being integrated with other control tactics within the integrated pest management - IPM (Kogan, 1975; Boiça Júnior et al., 2015).

Therewith, the aim of this study was to evaluate the feeding preference of *S. cosmioides* larvae in bean genotypes.

MATERIAL AND METHODS

The following common bean genotypes were evaluated for resistance to *S. cosmioides* larvae: BRS Pérola, IAC Harmonia, BRS Supremo, BRS Talismã, and IPR Campos Gerais. Double and multiple choice tests were performed under a randomized complete block desing, with 10 replications each. In the multiple-choice test, arenas composed (14 cm diamenter), Petri dishes were used. In the double choice test, (8 cm diamenter) Petri dishes were used, confronting the genotypes two by two. Petri dishes were coated at the bottom with moistened filter paper, and 3 cm diameter leaf disc of the respective genotype was distributed equidistantly. In both tests, third-instar of *S. cosmioides* larvae was released per genotype.

The evaluation consisted in counting the average number of larvae present in the leaf discs, after, one, six and 12 hours from the beginning of the experiment. In the double-choice test, the Preference Index (PI) was calculated according to Kogan (1972), using the formula: PI = 2A / (A + T), where A = number of *S. cosmioides* larvae present in the first bean genotype confronted, and T = of the second genotype. The value of the Standard Error of the mean (SE) was added / subtracted to the values of PIs. Values of PI ± SE >1 indicate preference for genotype "A", PI ± SE <1 preference for "T" genotype and PI ± SE = 1 neutraly. For the multiple-choice test the principal component analysis (PCA) (Jackson, 1991) was applied using the Statistica program version 7.0 (Statsoft Inc., 2004). Principal component analysis was used as exploratory analysis, which does not require investigating the assumptions of independence and normality.

RESULTS AND DISCUSSION

BRS Pérola genotype was preferred by *S. cosmioides* when compared with IAC Campos Gerais and BRS Supremo, and neutrality condition was observed when this cultivar was compared to the IAC Harmonia and BRS Talismã (Fig. 1). The same neutrality condition was observed with BRS

Supremo, when compared to BRS Talismã and IPR Campos Gerais. Neutrality was repeated when BRS Talismã was confronted with IPR Campos Gerais. Finally, we can say that IPR Campos Gerais, BRS Talismã and BRS Supremo were more preferred than the genotype IAC Harmonia. Based on the IPs, there is a high susceptibility to the BRS Pérola, and moderate resistance to IAC Harmonia, BRS Talismã and IPR Campos Gerais.

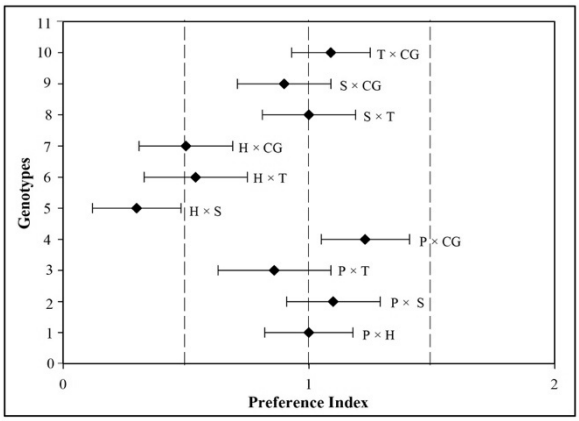


Figure 1 *Spodoptera cosmioides* Preference index (PI) comparing bean genotypes two by two in double change test. P = BRS Pérola; H = IAC Harmonia; S = BRS Supremo; T = BRS Talismã; C = IPR Campos Gerais.

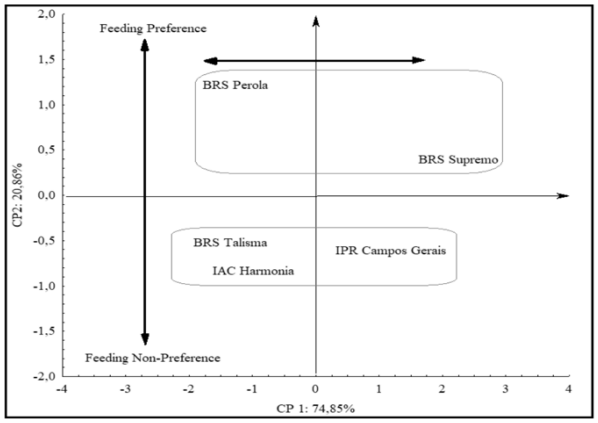


Figure 2 Bi-plot of bean genotypes in the multiple choice test for feeding of *Spodoptera cosmioides* for first two principal's components.

For the multiple-choice test is observed similarity between genotypes IAC Harmonia and BRS Talismã, both were equally preferred by *S. cosmioides*. In contrast, these materials showed a low number of larvae on the discs, which indicates the non-preference for feeding, due to the proximity of these two genotypes in the analysis of principal components and its location at the bottom of the graph. The same occurred for IPR Campos Gerais, wich fell in the group of beans less preferred for feeding (Fig. 2).

On the other hand, it's verified that Pérola was most preferred for feeding followed by BRS Supremo, showing the preference of *S. cosmioides* to both genotypes, which occurs due to the dissimilarity (top and opposite sides of the graph) between them (Fig. 2).

Our results showed high susceptibility of BRS Pérola genotype, moderate susceptibility of BRS Supremo and moderate resistance of IAC Harmonia, IPR Campos Gerais and BRS Talismã to *S. cosmioides*.

REFERENCES

BAVARESCO, A.; GARCIA, M. C.; GRÜTZMACHER, A. D.; RINGENBERG, R.; FORESTI, J. Adequação de uma dieta artificial para a criação de *Spodoptera cosmioides* (Walk.) (Lepidoptera: Noctuidae) em laboratório. *Neotropical Entomology*, v. 33, p. 155-161, 2004.

BOIÇA JÚNIOR, A. L. et al. A defesa das plantas ao ataque dos insetos. In: Busoli, A. C. et al. (Eds.). *Tópicos em entomologia agrícola – VIII*. Jaboticabal: Maria de Lourdes Brandel-ME. 2015. p. 161-179.

JACKSON, J. E. A user's guide to principal components. New York: Wiley, 1991. 569p.

KOGAN, M. Feeding and nutrition of insects associated with soybeans. 2. Soybean resistance and host preferences of Mexican bean beetle, *Epilachna varivestis*. *Annals of Entomological Society of America*, v. 65, p. 675-683, 1972.

KOGAN, M. Plant resistance in pest management. In: Metcalf, R.L.; Luckmann, W.H. (eds) Introduction to insect pest management. New York: Wiley, 1975. p. 103-146.

STATSOFT Inc. Statistica: data analysis software system, version 7. 2004. Tulsa. Disponível em http://www.statsoft.com/

ANTIBIOSIS RESISTANCE IN *Phaseolus vulgaris* L. TO *Spodoptera frugiperda* (SMITH) (LEPIDOPTERA: NOCTUIDAE)

M. M. Freitas[1*], C.A. Freitas[1], P. H. S. Barcelos[1], A. Bianchini[2], A. F. Chiorato[3], A. L. Boiça Júnior[1]

[1]Universidade Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias (UNESP/FCAV), Via de Acesso Prof. Paulo Donato Castellane s/n, Jaboticabal, SP, 14884-900, Brazil; [2]Instituto Agronômico do Paraná (IAPAR), Rodovia Celso Garcia Cid, km 375, CEP 86047-902, Londrina, PR, Brazil; [3]Instituto Agronômico (IAC), Av. Barão de Itapura, 1.481, CEP 13020-902, Campinas, Brazil. [*]E-mail: freitasmm@hotmail.com

INTRODUCTION

Host-plant resistance to insects (HPR) is an important control method and must be included in the Integrated Pest Management (IPM) programs, due to its efficiency and other advantages that it provides to the agroecosystem (BOIÇA JÚNIOR et al., 2017). Thus, selection, identification and characterization of resistance levels of genotypes and/or cultivars is essential for the implementation of HPR in the IPM of a given crop.

As well as the common bean (*Phaseolus vulgaris* L.), which has several pests throughout the development, among them *Spodoptera frugiperda* (Smith, 1797) (Lepidoptera: Noctuidae), a polyphagous insect that presents suitable development in bean, which can cause significant losses to the crop (BOREGAS et al., 2013). Thus, the aim of this study was to evaluate the resistance of bean cultivars to *S. frugiperda* by the antibiosis category.

MATERIAL AND METHODS

In this study were evaluated the cultivars IAC Harmonia, BRS Pérola, BRS Pitanga, BRS Campeiro e IPR Celeiro at vegetative stage V4 (second fully expanded trifoliate). The experiment was carried out under laboratory conditions (26 ± 2 °C and 60 ± 10% RH) and conducted in a completely randomized design with 6 replicates. Each replicate was represented by five newly hatched *S. frugiperda* larvae individualized in plastic containers (145 mL) lined with deionized water moistened filter paper, containing leaves of the respective evaluated cultivar. The biological parameters evaluated were: larval weight at 10 days after hatching, survival and larval period, pupal weight at 24 hours of age, period and pupal survival, total survival, adult longevity and total period (larvae hatching to adult death).

The data were submitted to analysis of normality and homogeneity, after analysis of variance (ANOVA) and when significant, the means of treatments were compared by Tukey's test (α = 0.05). However, data that did not meet the assumptions for ANOVA were analyzed by Kruskal-Wallis test (α = 0.05).

RESULTS AND DISCUSSION

There were no differences regarding pupal survival (Fig. 1B), pupal weight (Fig. 1C), pupal period and adult longevity (Fig. 1D). The cultivar IAC Harmonia caused total mortality of *S. frugiperda* larvae and seven days after hatching only 20% were alive. The cultivars BRS Pitanga and IPR Celeiro also presented low larval survival, 16.67% and 20.83% respectively (Fig. 1A), and lower total survival compared to BRS Pérola and BRS Campeiro (Fig. 1B).

The cultivar IAC Harmonia had lower larval weight, being similar to BRS Pitanga and BRS Campeiro cultivars (Fig. 1C). Regarding to biological stages, it was observed that the cultivar BRS Campeiro causes larger larval and total periods (Fig. 1D).

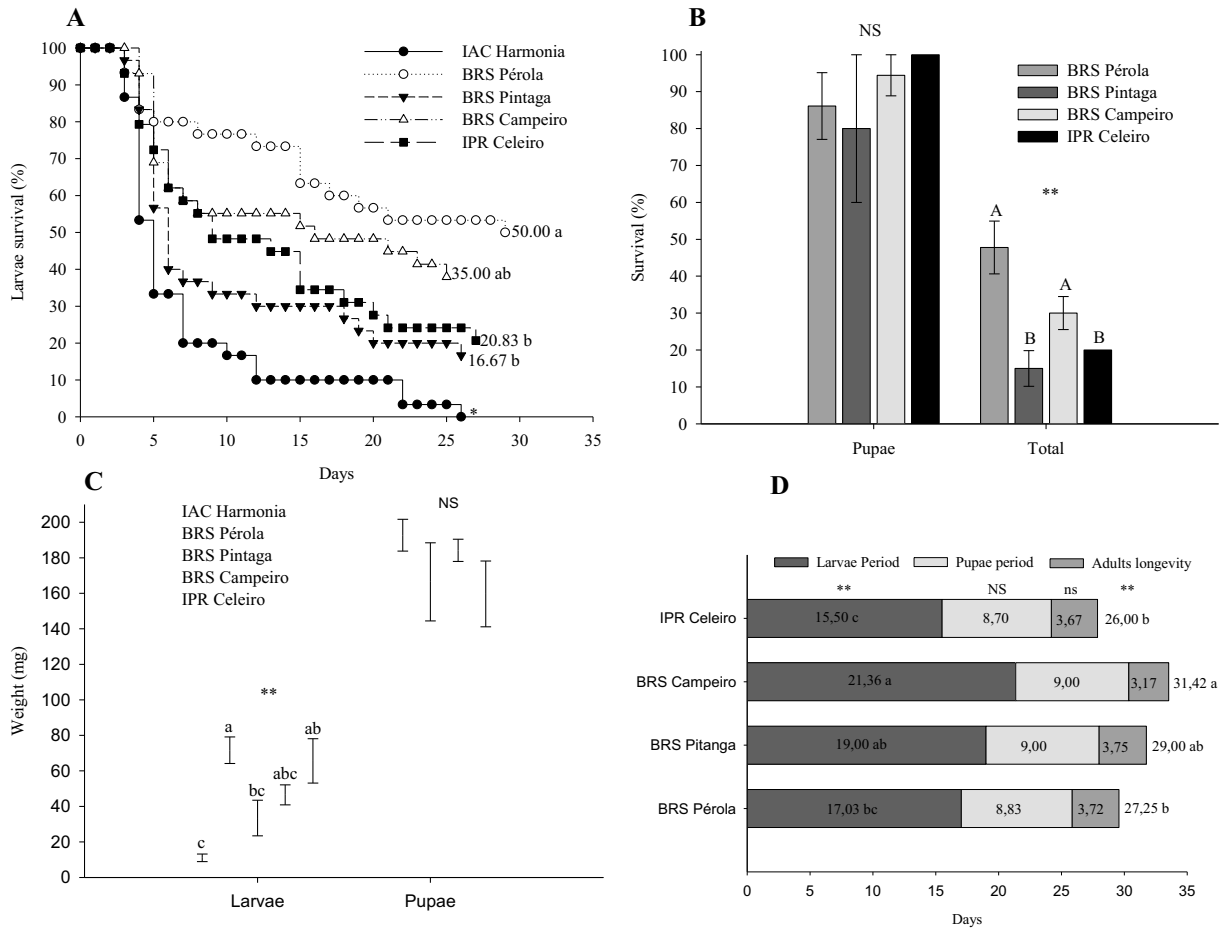


Figure 1. Biological parameters of *Spodoptera frugiperda* in common bean cultivars; (A) larval survival; (B) pupal and total viability; (C) larval and pupal weight; (D) larval period, pupal period, adult longevity and total period. NS – no significant difference by Kruskal-Wallis test (P<0.05). ns – no significant difference by F test (p <0.05). ** significant difference by F test (P< 0.01). * Insufficient data for analysis. Equal lowercase letters do not differ in the same parameter by Tukey's test (p <0.05).

Therefore, it is verified that bean cultivars can negatively affect the *S. frugiperda* development, characterizing the expression of resistance by the antibiosis category, in which the cultivar IAC Harmonia is resistant, while BRS Pitanga, IPR Celeiro and BRS Campeiro are moderate resistant and BRS Pérola is susceptible to *S. frugiperda*.

REFERENCES

BOIÇA JÚNIOR, A. L. et al. Resistência de plantas a insetos em culturas agrícolas. In: CASTILHO R. C. et al. (Eds.). *Tópicos em entomologia agrícola – X*. Jaboticabal: Gráfica Multipress LTDA, 2017. p. 97-122.

BOREGAS, K. G. B. et al. Estádio de adaptação de *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae) em hospedeiros alternativos. *Bragantia*, Campinas, v. 72, n. 1, p. 61-70, 2013.

EFFECT OF BEAN CULTIVARS AND NEEM OIL ON SURVIVAL OF *Spodoptera frugiperda* (J.E. SMITH, 1797) (LEPIDOPTERA: NOCTUIDAE).

M. M. Freitas[1*], C. A. Freitas[1], P. H. S. Barcelos[1], A. Bianchini[2], A. F. Chiorato[3], A. L. Boiça Júnior[1]

[1]Universidade Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias (UNESP/FCAV), Via de Acesso Prof. Paulo Donato Castellane s/n, Jaboticabal, SP, 14884-900, Brazil; [2]Instituto Agronômico do Paraná (IAPAR), Rodovia Celso Garcia Cid, km 375, CEP 86047-902, Londrina, PR, Brazil; [3]Instituto Agronômico (IAC), Av. Barão de Itapura, 1.481, CEP 13020-902, Campinas, Brazil. [*]E-mail: freitasmm@hotmail.com

INTRODUCTION

The fall armyworm *Spodoptera frugiperda* (Smith, 1797) (Lepidoptera: Noctuidae) is a polyphagous pest that causes significant damage to several crops of economic interest, including bean (*Phaseolus vulgaris* L.) (BOREGAS et al., 2013). Among the main control tactics, the use of synthetic insecticides has been the most used tool against *S. frugiperda* (CARVALHO et al., 2013) and, therefore, alternative control methods such as host-plant resistance and the use of natural insecticides are essentials for the fall armyworm management in bean crop. This study investigated the effects of bean cultivars and natural insecticide based on neem oil in the survival of *S. frugiperda*.

MATERIAL E METHODS

The experiments were carried out at Laboratory of Host-Plant Resistance to Insects and under greenhouse conditions at Crop Protection Department – UNESP / FCAV. Bean cultivars used were IAC Harmonia, BRS Pérola, BRS Pitanga, BRS Campeiro, IPR Celeiro. Two independent experiments were conducted in greenhouse and laboratory, and in each experiment ten plants of each cultivar in vegetative stage V3 were used, in which five plants were sprayed with neem oil (spray volume 120 L ha^{-1}; dose of 600 mL ha^{-1}) and five plants were sprayed with water (treatment control). In the first experiment, two hours after application, leaves of each treatment were collected, taken to the laboratory and individualized in plastic containers (140 mL) lined with deionized water moistened filter paper. In each plate, a newly hatched *S. frugiperda* larvae was released, performing thirty replications under a completely randomized design in a 5 x 2 factorial scheme (cultivar x natural insecticide).

In the second experiment, two hours after application, two second-instar *S. frugiperda* larvae were confined in the first trifoliate from the plant in a mesh-bag cage (10 x 15 cm). The experiment was carried out under a completely randomized design in a 5 x 2 factorial scheme (cultivar x natural insecticide). In both experiments larval survival was evaluated 1, 3, 5 and 7 days after the application (DAA) of neem oil. Data were subjected to two-way analysis of variance (ANOVA) for the main effects of cultivar (C) and oil neem (N) on survival of *S. frugiperda*, and when significant, treatment means were separated by Tukey's test ($\alpha = 0.05$).

RESULTS AND DISCUSSION

According to the results there was significant interaction between the main factors only at 7 DAA in laboratory conditions, however, it was verified that the effect of the cultivar and neem significantly influenced the survival of *S. frugiperda* in laboratory and greenhouse (Table 1). In laboratory conditions, lower survival larvae were observed 3 DAA in all treatments that received neem oil (between 35 and 65%) regarding the treatments that did not receive (between 86 and

100%), while at 3 DAA the survival was significantly lower (70%) only in the IPR Celeiro in greenhouse conditions when the neem oil was applied regarding the other treatments (Figure 1).

Table 1. Summary of the ANOVA for the effects of cultivar and application of neem on survival of *Spodoptera frugiperda* larvae fed on bean cultivars.

Effects	1 DAA[1]		3 DAA		5 DAA		7 DAA	
	F	*P*	*F*	*P*	*F*	*P*	*F*	*P*
Laboratory								
Cultivar (C)	-	-	2.22	0.0797	5.11	0.0016**	6.28	0.0004**
Neem (N)	-	-	88.99	0.0001***	98.26	0.0001***	422.35	0.0001***
C x N	-	-	0.98	0.4258	1.36	0.2628	6.28	0.0004**
Greenhouse								
Cultivar (C)	-	-	1.21	0.3238	0.71	0.5884	0.98	0.4283
Neem (N)	-	-	7.17	0.0108*	48.80	0.0001***	108.49	0.0001***
C x N	-	-	0.93	0.4548	0.97	0.4342	0.20	0.9349

DAA – Days after application of neem. [1] Data not analyzed since there was no variation. * P<0.05; ** P<0.01; *** P<0.0001.

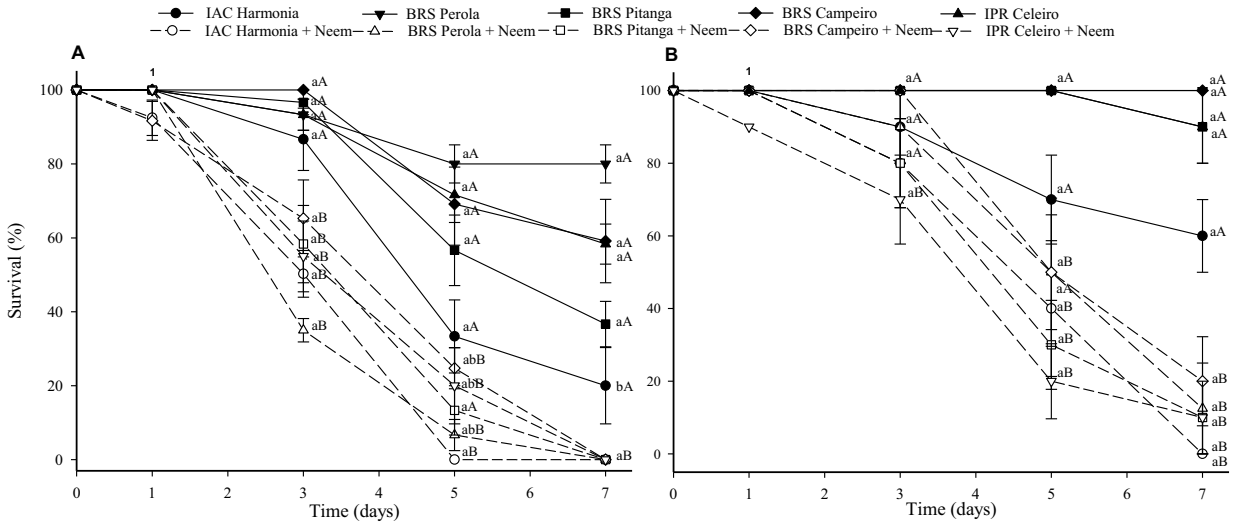


Figure 1. Survival of *Spodoptera cosmioides* larvae fed on bean cultivars with and without application of neem-based natural insecticide in laboratory (A) and greenhouse (B) conditions. Different lowercase letters indicate significant difference between cultivars with or without application and different uppercase letters indicate significant difference in each cultivar with and without neem oil application (P <0.05). [1]Data not analyzed since there was no variation. * P<0.05; ** P<0.01; *** P<0.0001.

At 5 and 7 DAA, in both experiments, negative neem effect on the survival larvae was observed in all cultivars, and at 7 DAA the cultivar IAC Harmonia caused significantly lower survival (20%) compared to the other treatments that did not receive neem oil application in the laboratory (36 to 80%). The results obtained in this study demonstrate that neem negatively affects the survival of *S. frugiperda* in bean cultivars, and the resistance characteristics of the IAC Harmonia cultivar in association with neem increase the control and become an efficient alternative for the management of *S. frugiperda* and can contribute to reduction of losses in bean crop production.

REFERENCES

BOREGAS, K. G. B.; MENDES, S. M.; WAQUIL, J. M.; FERNANDES, G. A. W. Estádio de adaptação de *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae) em hospedeiros alternativos. *Bragantia*, Campinas, v.72, p.61-70, 2013.

CARVALHO R. A.; OMOTO, C.; FIELD, L. M.; WILLIAMSON, M. S.; BASS, C. Investigating the molecular mechanisms of organophosphate and pyrethroid resistance in the fall armyworm *Spodoptera frugiperda*. *PLoS ONE*, San Francisco, v. 8, n. 4, e62268, 2013.

PERFORMANCE AND FITNESS STAGE OF *Helicoverpa armigera* (HÜBNER) (LEPIDOPTERA: NOCTUIDAE) FED ON LEAVES AND PODS OF COMMON BEAN

L. Nogueira[1*], J. G. E. A. R. Aiala[1], G. A. P. Bernardes[1], C. C. Melville[2], A. L. Boiça Júnior[2]

[1] Univ. Estadual de Goiás, UEG, Laboratório de Entomologia Agrícola, Rod. GO 330, Km 241, s/n°, Ipameri, GO, 75780-000, Brazil; [2]Univ. Est. Paulista, UNESP/FCAV, Via de Acesso Prof. Paulo Donato Castellane s/nº, Jaboticabal, SP, 14884-900, Brazil; *corresponding author: lucianonogueiraagro@gmail.com

INTRODUCTION

The cotton bollworm, *H. armigera* Hübner (Lepidoptera: Noctuidae) is a globally distributed polyphagous pest of many crop plants, with high reproductive potential (Sharmad et al., 2005; CABI, 2018). The larvae cause significant losses by attacking the plants with such intensity they that feed on the plants both in the vegetative and reproductive stages (Fitt, 1989), causing losses of up to 80% in the bean crop (Gottems 2013). Depending on the availability of hosts and the performance of *H. armigera* in agroecosystems, significant changes may occur in the population dynamics of this species. Based on the fitness results for *H. armigera* on leaves and bean pods, we evaluated their suitability and potential contribution to population size. Thus, the knowledge of the biological performance of *H. armigera* should contribute to a better understanding of its ecology and improvement of the current integrated pest management – IPM programs. The objective of this study was to evaluate the performance and adaptive stage of *H. armigera* in leaves and common bean pods.

MATERIAL AND METHODS

The study was conducted in the agricultural entomology Laboratory, UEG, Ipameri, GO, Brazil, under controlled conditions. The bean seeds, cultivar BRS Pérola, were sown in 5-L plastic pots and kept in a greenhouse and was irrigated manually every other day or as needed. The experiment consisted of two treatments, plants used in the vegetative stage (V5) and plants with pods used in the reproductive stage (R8). The leaves and pods were collected from the respective plants and taken to the laboratory, washed with a solution of 0.05% sodium hypochlorite, then rinsed in deionized water and subsequently dried with paper towels. The *H. armigera* larvae (<12 h of hatching) were stored in cylindrical plastic containers (8 cm high x 5 cm diam.), containing filter paper moistened with deionized water.

For the study of biological aspects, the following variables were evaluated: (1) larval period: duration, viability and weight of larvae at 10 days age; (2) pupal period: weight at 1 day age (24 h). A completely randomized design with 40 replicates per treatment was used. To estimate the growth index, we used the biological adjustment (*r*L) proposed by (Jallow and Zalucki, 2003). The *r*L is estimated from the larval development period (days), pupal biomass and larval survival, and serves as an indicator of offspring performance. The aptitude index was calculated as $rL = lx.Mx/tl$, where l*x* is the larval survival, m*x* is the average pupae weight, and t*l* is the larval phase duration.

Data were first tested for normality (Levene's test), homogeneity (Shapiro-Wilk's test) and analyzed using Student's pairwise t-test ($P < 0.05$).

RESULTS AND DISCUSSION

In this study, it was possible to observe that both pods and bean leaves interfered negatively in the growth and survival of *Helicoverpa armigera* larvae. The highest weight were observed in the larvae fed with bean pods (Fig. 1A) and the pupae from the treatment in which the larvae fed on

bean pods also had a higher weight (1B). The highest survival was observed in larvae fed with bean leaves (Fig. 3). The growth rate of *H. armigera* was higher when grown in bean pods.

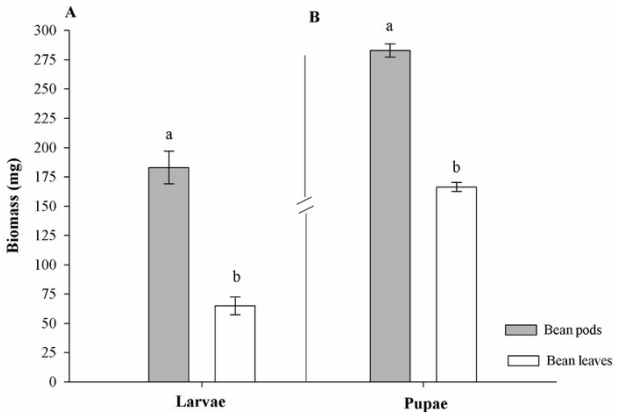


Fig. 1 Average larval biomass (±SE) of *Helicoverpa armigera* fed with bean pods and leaves at larvae (**A**) and Pupae (**B**). Differents letters in the bars indicate significant differences in **A** (Student's t-test; $P < 0.0001$; F = 54.75**) and **B** (Student's t-test; $P < 0.0001$; F = 306.55**).

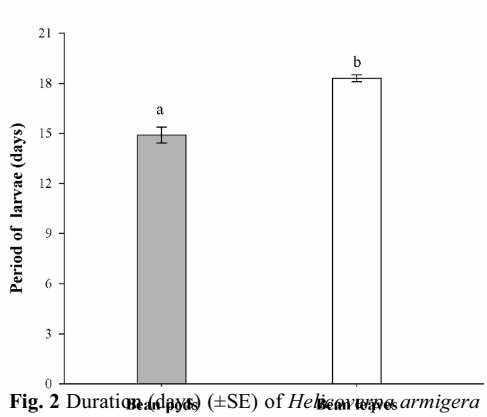


Fig. 2 Duration (days) (±SE) of *Helicoverpa armigera* fed with bean pods and leaves. Differents letters in the bars indicate significant differences (Student's t-test; $P < 0.0001$; F = 43.92**).

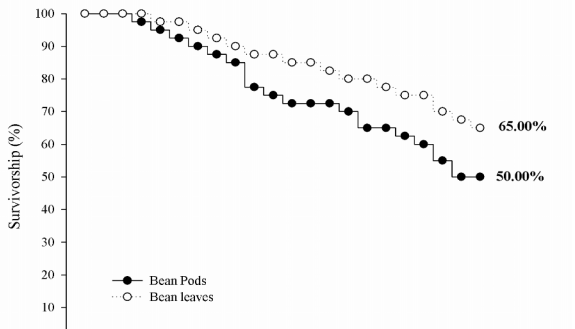


Fig. 3 Age-specific survivorship of larval stages of *Helicoverpa armigera* reared on bean pods and leaves. There is no significant difference between the survivorship curves for larvae through the Log-Rank test ($\chi^2 = 0.1985$; $P = 0.6559$) and Wilcoxon test ($\chi^2 = 0.6733$; $P = 0.7200$).

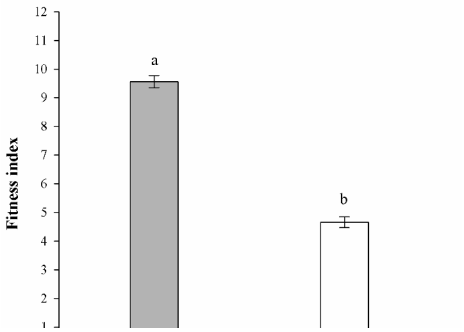


Fig. 4 Fitness index of *Helicoverpa armigera* (±SE) larvae fed bean pods and leaves. Different letters above the bars indicate significant difference (Student's t-test; $P < 0.0001$; F = 303.85**).

Our results show that fresh bean pods are nutritionally more important for growth and development of *H. armigera* larvae than leaves. Larvae have a greater potential to cause damage to the reproductive stage of beans plants.

REFERENCES

SHARMAD, H. C.; PAMPAPATHY, G.; LANKA, S. K.; RIDSDILL-SMITH, T. J. Antibiosis mechanism of resistance to pod borer, *Helicoverpa armigera* in wild varieties of chickpea. *Euphytica*, 142: 107-115. 2005.

CABI. Crop Protection Compendium. *Helicoverpa armigera*. 2018. Avaliable: <http://www.cabi.org/cpc/datasheet/26757>. Acessed: jan., 02, 2018.

JALLOW, M. F. A.; ZALUCKI, M. P. Relationship between oviposition preference and offspring performance in Australian Helicoverpa armigera (Hübner) (Lepidoptera: Noctuidae). *Aust. J. Entomol*. 42: 343–348. 2003.

GOTTEMS, L. 2013. Prejuízos com lagarta *Helicoverpa armigera* chegam a 80% do feijão em SP. Avaliable: https://www.agrolink.com.br/noticias/prejuizos-com-lagarta-helicoverpa-armigera-chegam-a-80--do-feijao-em-sp_168639.html. Acessed: jan., 03, 2018.

PREDADOR AND PARASITOID ARTROPOD'S OCCURRENCE IN COMMON BEAN (*Phaseolus vulgaris* L.), CULTIVATED IN THE STATE OF PARANÁ, BRAZIL

Kawashima[1] AB, Hoshino[1] AT, Menezes Junior[1] AO, Simionato[1] ME, Cantamessa[1] LA, Oliveira[1] LM, Kusdra[2] GRF, Oliveira[2] IJ, Karas[2] F, Matyak[2] N, Teixeira[2] NP, Lima[2] D, Caviglione[3] JH, Androcioli[3] HG

[1]Universidade Estadual de Londrina - UEL, Londrina, PR, Brazil; [2]Instituto Paranaense de Assistência Técnica e Extensão Rural – EMATER, Londrina, PR, Brazil; [3]Instituto Agronômico do Paraná - IAPAR, Londrina, PR, Brazil; handrocioli@iapar.br

INTRODUCTION

Pest occurrence restrain common bean (*Phaseolus vulgaris*, L) production in the second crop ("droughts harvest"), in the state of Paraná (Hohman & Carvalho, 1989), due to a combination of crop succession (beans after soybeans) and pest-favorable climatic conditions. The presence of biological control agents in cultivated areas is fundamental to reduce the pest outbreak, excluding the need of pesticide application in many locations. Thus, it is fundamental to know which natural enemy groups occur in the croplands, so that further research towards strategies to preserve and increase their populations is performed. The aim this study was to evaluate the predator and parasitoid arthropod's occurrence in the common bean, in the southern of Paraná, Brazil.

MATERIALS AND METHODS

The study was performed during the second crop of 2017, in five commercial areas, located in the counties of Pinhão, São João do Triunfo, Cândido de Abreu, Agudos do Sul and Lapa, all in the southern region of Paraná State, Brazil. The bean cultivar used was IPR Tuiuiú, following an Integrated Pest Management (IPM) system. Between the months of February and May, predatory and parasitoid arthropods were collected by Malaise trap (Townes 1972 model). In each commercial area, a trap was placed inside its field, having four weekly sampling during the period. The collected arthropods were sent to the Laboratory of Entomology of the Universidade Estadual de Londrina (UEL), where quantifications and identifications, at least up to the family taxonomic level of the predatory and parasitoid specimens were carried out.

RESULTS AND DISCUSSION

Was collected 3289 specimens of predator and parasitoid insects belonging to 15 and 21 families, respectively, plus the Araneae order (Table 1). The abundance of different natural enemy groups varied among the localities, the county of Lapa had the greater abundance while the county of Candido de Abreu had the smaller one. This difference is probably due to the environmental composition around the cropland. The most abundant predators were the Diptera, Dolichopodidae (43.0%), and the beetles Carabidae (14.8%) and Coccinellidae (14.0%). These groups are important generalist predators, due to their greater adaptability to the agricultural environment, in relation to other more specialist groups (Rand & Tscharntke, 2007). The most abundant Carabidae, *Lebia concinna* (Brullé, 1838), represented over 80% in three areas and about 50% in the rest, this predator along Dolichopodidae and Coccinellidae contribute to reducing the main bean pest's population, like whitefly, thrips, mites (Lundgren et al., 2014) caterpillars in the first instar, eggs, nymphs and aphids (Bueno et al., 2012).

Table 1 - Predator and parasitoid arthropod's abundance captured by the malaise trap, in common bean cropped area located in the state of Paraná. Data referring to four sampling during the second crop of 2017.

	Taxa (Family)	County					Sum	RF (%)[1]
		Pinhão	São João do Triunfo	Cândido de Abreu	Agudos do Sul	Lapa		
Predators	Dolichopodidae	103	12	27	6	246	394	42.97
	Carabidae	9	21	2	12	92	136	14.83
	Coccinellidae	20	1	45	35	27	128	13.96
	Vespidae	1	2	1	1	58	63	6.87
	Hemerobiidae	22	0	1	14	17	54	5.89
	Araneae [2]	8	4	5	9	20	46	5.02
	Syrphidae	3	1	1	14	10	29	3.16
	Another 9 families [3]	13	10	6	14	24	67	7.31
Parasitoids	Encyrtidae	40	313	14	37	762	1166	49.16
	Tachinidae	129	39	40	109	343	660	27.82
	Braconidae	20	11	57	21	101	210	8.85
	Ichneumonidae	22	5	24	25	97	173	7.29
	Another 17 families [3]	14	4	6	8	131	163	6.87

[1] Relative frequency, expressed in percent; [2] Taxon at the order level; [3] Families that together accounted less than 8% of the collected.

The most abundant parasitoids were the Diptera Tachinidae (27.8%), and the Hymenoptera Encyrtidae (49.2%) and Braconidae (8.9%). Among the Encyrtidae family, a lot of individuals from the genus *Copidosoma* (Ratzeburg, 1844) were collected, known as an important parasitoid of Plusiinae caterpillars. Tachinidae flies have certain representatives that parasite some caterpillars, true bugs and *Diabrotica* beetles (Bueno et al., 2012); while Braconidae wasps have representatives that are parasitoids of caterpillars and aphids (Moraes et al., 1991; Starý et al., 2007). The predator and parasitoid arthropods occur in common bean crops in the southern of Paraná State, being important bio-control agents responsible for balancing the agroecosystem and maintaining a low pest density population.

REFERENCES

Bueno, A. de F. Inimigos naturais das pragas da soja. In: Soja: manejo integrado de insetos e outros artrópodes-praga. Hoffmann-Campo, C. B.; Corrêa-Ferreira, B. S.; Moscardi, F. (Ed). Brasília: Embrapa, p.493-629, 2012.

Hohmann, C. L.; Carvalho, S. M. Pragas e seu controle. In: O feijão no Paraná. Londrina: Fundação Instituto Agronômico do Paraná, (IAPAR, Circular 63), p.217-246, 1989.

Lundgren, J. G. et al. Molecular determination of predator community of a cassava whitefly in Colombia: pestspecific primer development and field validation. Journal of Pest Science, v.87, p.125-131, 2014.

Moraes, R. R. et al. Inimigos naturais de *Rachiplusia nu* (Guenée, 1952) e de *Pseudoplusia includens* (Walker, 1857) (Lepidoptera: Noctuidae) em soja no Rio Grande do Sul. Pesqui. Agropecu. Bras.,v.26, n.1, p.57-64, 1991.

Rand, T.; Tscharntke, T. Contrasting effects of natural habitat loss on generalist and specialist aphid natural enemies. Oikos, v.116, p.1353-1362, 2007.

Starý, P. et al. Aphid parasitoids (Hymenoptera, Braconidae, Aphidiinae) and their associations related to biological control in Brazil. Revista Brasileira de Entomologia, v.51, n.1, p.107-118, 2007.

GENETIC DIVERSITY IN *Phaseolus lunatus* L. BASED ON MORPHOLOGICAL SEED CHARACTERS

Wilson Vitorino de Assunção Neto[1], Rafael da Costa Almeida[2], Antônia Maria de Cássia Batista[1], Hildete Leal do Nascimento[1], Ângela Celis de Almeida Lopes[1], Leonardo Castelo Branco[1], Regina Lucia Ferreira Gomes[1]

[1]Dep. de Fitotecnia, Universidade Federal do Piauí, Teresina, PI, Brazil; [2]Instituto Federal do Piauí, Pedro II, PI, Brazil

INTRODUCTION

Phaseolus lunatus L. is a tropical legume, characterized by high genetic diversity, which allows its adaptation to the most different environmental conditions. This species has great socioeconomic importance in the Northeast of Brazil, due to its rusticity, which makes it possible to prolong the harvest in the dry period. Thus, this study aimed to study the genetic diversity of the lima bean accessions of the Germplasm Active Bank of the Federal University of Piauí (UFPI), by the morphological characterization of the seeds.

MATERIALS AND METHODS

The genetic material used consisted of 219 lima bean accessions from the Germplasm Active Bank of UFPI, registered in the period from 2013 to 2017. In the physical characterization of the seeds, quantitative descriptors were evaluated, according to Bioversity International (IPGRI, 2001), in ten seeds taken at random from each accession (Tanabata et al., 2012), as well as the thickness with a digital caliper, and the weight of one hundred seeds (P100S) in g, with an average of three replicates. The seeds were classified as small (less than 30 g), medium (31 to 40 g), normal (41 to 59 g), and large (greater than 60 g). The shape as a function of the index J, obtained by the relation between length and width, were classified as spherical, elliptical and oblong/short reniform. The classification of the seeds according to the profile as a function of the H index, based on the thickness/width ratio, was: flattened, semi-flattened and full (Vilhordo et al., 1996). In the clustering analysis, the hierarchical UPGMA method was adopted, and we use the mean Euclidean distance as a measure of dissimilarity.

RESULTS AND DISCUSSION

According to the Mackie (1943) classification, the accessions of the present study are predominantly belonging to the Big Lima Group, representing the set of Andean genes, whose domestication occurred in South America, with a presence of large and flat seeds (Table 1).

Table 1 - Classification of bean accessions of the Germplasm Active Bank of the Federal University of Piauí, Brazil, according to the size, the shape (function of index J) and the profile (function of the index H).

Descritor	Avaliação			
Size	Small (7.30%)	Medium (18.18%)	Normal (28.31%)	Big (45.20%)
Shape	Spherical (50.68%)	Elliptical (48.85%)	Oblong/short reniform (0.45%)	
Profile	Flattened (96.80%)	Semi-flattened (2.74%)	Full (0.45%)	

In the dendrogram generated by the UPGMA hierarchical method (Figure 1) there were seven groups in which the most divergent and complementary pairs of accessions are in different groups. The group 1 had accesses with large seeds of flattened profile. In the group 2, the seeds were flattened and of small size. The group 3 had access with semi-flat and medium seeds. The group 4 was composed of normal size seeds. Group 5 had access with large and flat seeds. The group 6 with large seeds was formed by four accessions that presented as higher averages for the evaluated characteristics, and group 7 brought together accessions of large seed.
The characteristics that most contribute to the genetic divergence between the accessions were the seed area and the weight of one hundred seeds.

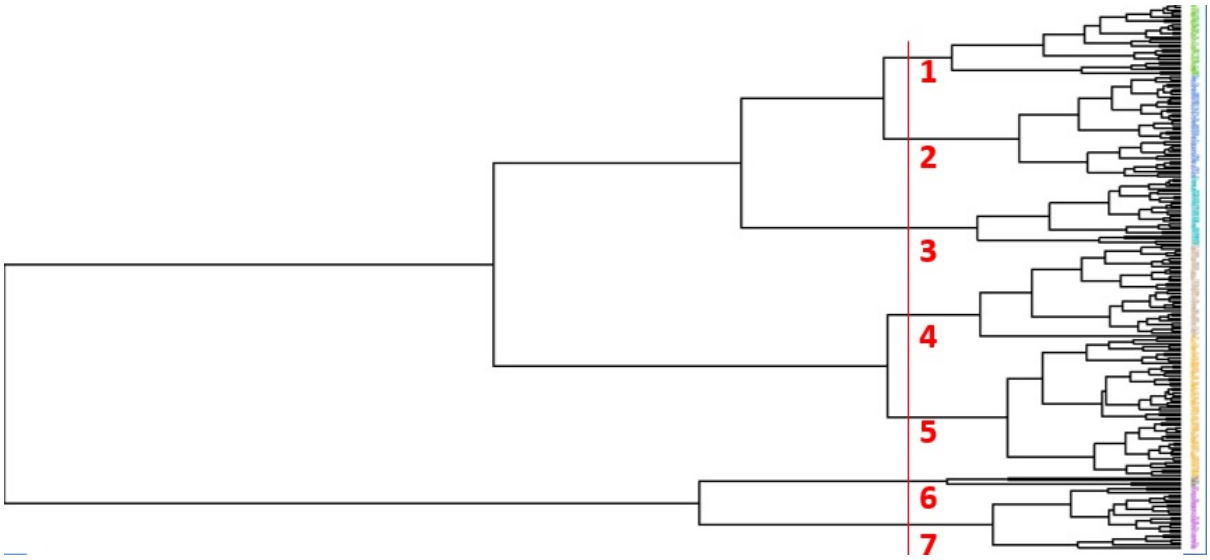


Figure 1 - Dendrogram of dissimilarity of 219 accessions of bean bean generated by the UPGMA grouping method, based on the mean Euclidean distance for quantitative characters. Teresina, PI, 2017

ACKNOWLEDGEMENTS

This research was financially supported by CNPq and CAPES.

REFERENCES

IPGRI. 2001. *Descritores para Phaseolus lunatus (feijão-espadinho)*. International Plant Genetic Resources Institute, Rome. pp. 51.

Mackie, W.W. 1943. Origin dispersal and variability of the lima bean (*Phaseolus lunatus*). *Hilgardia*, 15: 1-29.

Tanabata, T., Shibaya, T., Hori, K., Ebana, K., Yano, M. 2012. SmartGrain: high-throughput phenotyping software for measuring seed shape through image analysis. *Plant Physiology*, 160: 1871–1880.

Vilhordo, B.W., Mikusinski, O.M.F., Burin, M.E., Gandolfi, V.H. 1996. Morfologia. In Cultura do feijoeiro comum no Brasil. (Araújo, R.S., Rava, C.A., Stone, L.F., Zimmermann, M.J.O., Coord.). Piracicaba: Potafos, pp. 71-99.

SELECTION OF REPRESENTATIVE GENOTYPES OF LIMA BEAN (*Phaseolus lunatus* L.) FROM BRAZIL

Hildete Leal do Nascimento[1], Regina Lucia Ferreira Gomes[1], Ângela Celis de Almeida Lopes[1], Rafael da Costa Almeida[2], Leonardo Castelo Branco Carvalho[1]

[1]Dep. de Fitotecnia, Universidade Federal do Piauí, Teresina, PI, Brazil; [2]Instituto Federal do Piauí, Pedro II, PI, Brazil

INTRODUCTION

Ex-situ plant germplasm collections have increased enormously in number and size over the last decades as a result of global efforts to conserve genetic resources for food and agriculture. Faced with the availability of lima bean germplasm presenting variability for plant architecture and yield related traits, a selection of genotypes with high crop representativeness is of great importance for the conservation and development of improved cultivars to be inserted into the production system path of crop development and breeding. Definition of criteria for assemble and evaluation of the quality of core collections is a prerequisite for selecting high-quality cores (Odong et al., 2013). In this sense, the objective of this work was to select lima bean genotypes (*Phaseolus lunatus* L.) from Brazil, with high representativity.

MATERIALS AND METHODS

We selected 30 accessions of different thematic groups (distinct geographical origin and variation in color and shape of the seed) conserved in Germplasm Bank of the Federal University of Piauí. The experiment was conducted in the Teresina-PI, Brazil, and the experiment design was DBC with four replicates. The following traits were evaluated: number of days for flowering (NDF), number of days to maturity (NDM), grain color (GC), number of pods per plant (NPP), pod length (PL) , number of seeds per pod (NSP), weight of one hundred seeds (W100G), grain yield per plant (GYP) and grain yield expressed in kilograms per hectare (GY). The distance matrix was obtained through the method of Gower (1971) and it was used for a grouping of the most similar accessions by UPGMA (Unweighted Pair Group Method with Arithmetic Mean) and for selection of the most divergent by means of the Tocher method (Rao, 1952), adapted with inverse agglomerative criteria, as proposed by Vasconcelos et al. (2007), with a cutoff of 20%.

RESULTS AND DISCUSSION

The UPGMA method divided the accessions into 6 groups: Group 1 - UFPI 969, UFPI 933, and UFPI 950; Group 2 - UFPI 968, UFPI 948, UFPI 953, and UFPI 961; Group 3 - UFPI 956, UFPI 1004, UFPI 951, UFPI 959, UFPI 1017, and UFPI 942; Group 4 - UFPI 972, UFPI 1018, UFPI 666, UFPI 949, UFPI 999, UFPI 985, UFPI 965, UFPI 962, and UFPI 954; Group 5 - UFPI 960, and UFPI 967; Group 6 - UFPI 945, UFPI 958, UFPI 934, UFPI 1011, UFPI 978, and UFPI 971 (Figure 1a). Among them, was selected by Tocher method with inverse agglomerative criteria the accesses: UFPI 948, UFPI 967, UFPI 971, UFPI 950, UFPI 972, and UFPI 949; being therefore the most dissimilar accesses among them (Figure 1b). Both the means and variances between the thematic sample (30 accessions) and the selected genotypes (6 accessions) did not present significant differences by t-Student and F tests, respectively. This indicates that the selection strategy adopted was effective to obtain accesses representative of the genetic diversity of the original sample. The similarity coefficient (S) between the two collections was 87%, which shows the high efficiency of the Tocher method with inverse agglomerative criterion as an alternative for the assembly of core collections with a high degree of representativeness (Vasconcelos et al.,

2007). According to the results, the traits and strategy addressed in the present study can be used to obtain and evaluate suitable core collections of lima bean.

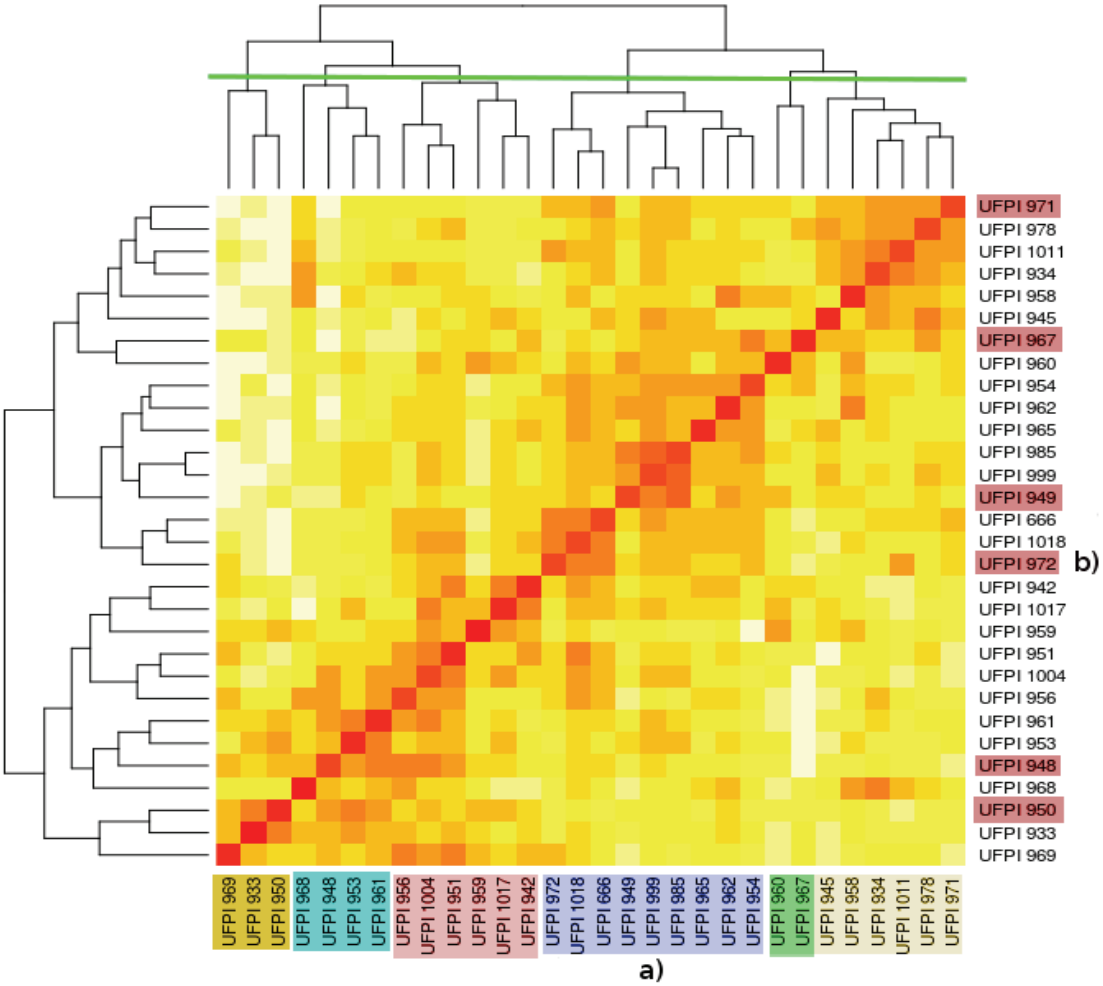


Figure 1. Heatmap representing the dissimilarity matrix obtained from the Gower method. Red regions represent high similarity between accessions and white regions represent low similarity. **a)** Groups formed according to UPGMA algorithm. **b)** Accessions selected as representative of the initial sample, according to the Tocher optimization method with inverse agglomerative criterion.

ACKNOWLEDGEMENTS

This research was financially supported by CNPq and CAPES.

REFERENCES

Gower, J. C. (1971) *Biometrics,* 27**:** 857–874.

Odong, T. L.; et al. (2013) Theor Appl Genet, 126:289–305.

Rao C.R. (1952). New York: John Wiley & Sons, 390p.

Vasconcelos, E. S.; et al. (2007) Pesquisa Agropecuária Brasileira, 42: 507-514.

GENETIC VARIABILITY FOR ALUMINUM TOLERANCE IN COMMON BEAN FROM DIFFERENT CENTERS OF ORIGIN

Douglas Mariani Zeffa[1,2], Sebastião Soares de Oliveira Neto[3], Guilherme Renato Gomes[4], Felipe Fadel Baptista[1] and Vânia Moda-Cirino[1]

[1]Instituto Agronômico do Paraná, Londrina, Paraná, Brazil.
[2]Universidade Estadual de Maringá, Paraná, Brazil
[3]Universidade Estadual de São Paulo, Botucatu, Brazil.
[4]Universidade Estadual de Londrina, Londrina, Paraná, Brazil.

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is produced in the tropics and sub-tropics mainly by small farmers where abiotic factors limit the yield potential. Among abiotic factors, aluminum toxicity is a serious environmental problem that limits crop production at commercial level (Singh et al., 2017). Thus, the aim of this study was to evaluated genotypes from different centers of origin for aluminum tolerance.

MATERIAL AND METHODS

Experiments were carried out in a greenhouse at Instituto Agronômico do Paraná (IAPAR) in Londrina, Paraná, Brazil. A total five genotypes common bean genotypes were evaluated, being three andeans (IPR Garça, BRS Radiante and DRK 2756) and two mesoamericans (IPR Tangará and IPR Uirapuru). Experimental design was a completely randomized design with four repetitions per treatment. Two Al conditions for distinguishing tolerant and sensible genotypes were applied: with 10 mg L^{-1} Al (+Al) and without Al (-Al) considered is a control, using nutrient solution proposed by Hoagland and Arnon (1950).

Seedlings were transplanted to polyethylene pots with five liters of nutrient solution and one plant per pot. Nutrition solution was aerated permanently with pH maintained around to 4.0±0.2. After 21 days, the following traits were analyzed shoot length (SL, cm), mean root length (MRL, cm), shoot dry biomass (SDB, g) and root dry biomass (RDM, g). The deviances analysis were calculated for each variable ($P \leq 0.05$). The heatmap and principal component analysis (PCA) were performed using standard Euclidean distances. The genotypes were classified for aluminum tolerance using the selection index under +Al conditions, which calculates the distances of each genotype to an ideal genotype (Wricke and Weber, 1986).

RESULTS AND DISCUSSION

The deviance values differed significantly by the chi-square test ($P < 0.05$) to all traits under +Al and –Al (data not shown), indicating high genetic variability among the genotypes. The visual representation by heatmap allowed to distinguishing clearly the behavior of the genotypes in both Al conditions (Figure 1), indicating that under +Al the average of all traits were lower when compared to –Al and that this condition affected the plants physiology and morphology. PCA revealed difference in the distribution of genotypes on the plot and showed a higher distribution of genotypes under +Al than -Al, indicating that the common bean genotypes can be better distinguished in stress conditions (Figure 2). The most tolerant aluminum genotypes were DRK 2756, BRS Radiante, IPR Garça, IPR Tangará and IPR Uirapuru, respectively.

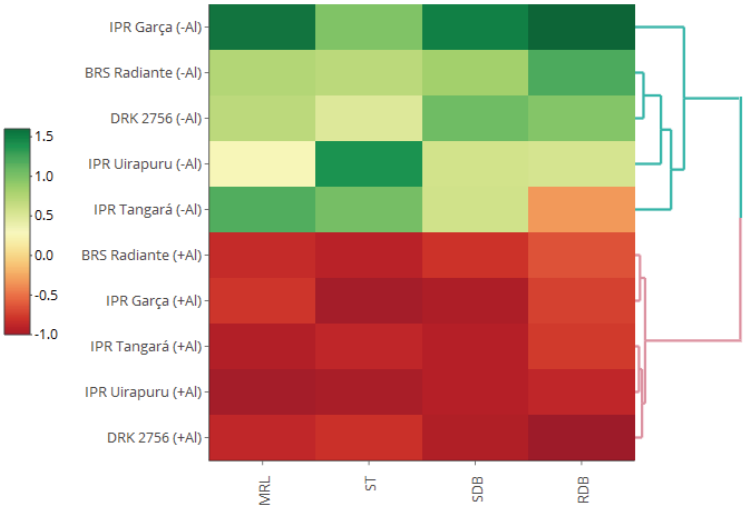


Figure 1. Heatmap of relationships among shoot length (SL), mean root length (MRL), shoot dry biomass (SDM) and root dry biomass (RDM), evaluated in five common bean genotypes in the presence (+ Al) and absence (- Al) of aluminum.

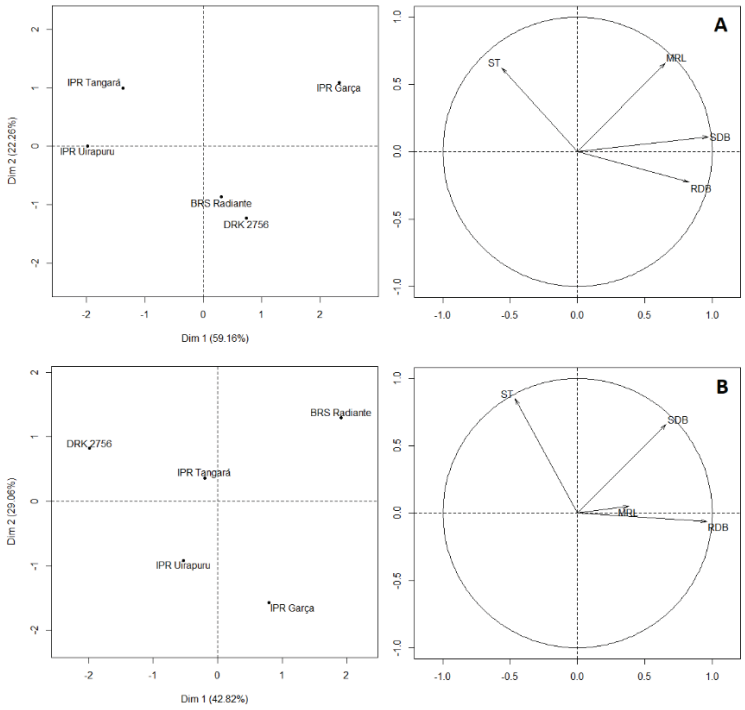


Figure 2. Principal components analysis (PCA) of the genetic relationships among five common genotypes bean in the absence (A) and presence of aluminum (B).

REFERENCES

Hoagland DR and Arnon DI (1950). The water culture method for growing plants without soil. *Circular. California Agricultural Experimental Station*, Berkeley. 32 p.

Singh, S., Tripathi, D. K., Singh, S., Sharma, S., Dubey, N. K., Chauhan, D. K., & Vaculík, M. (2017). Toxicity of aluminium on various levels of plant cells and organism: a review. *Environmental and Experimental Botany*, *137*, 177-193.

Wricke G and Weber WE (1986). *Quantitative genetics and selection in plant breeding.* New York: Walter de Gruyter, 406 p.

RESPONSE OF RED COMMON BEAN GENOTYPES TO ALUMINUM TOXICITY

João Marcos Novais[1]; Débora Jumes[1]; Guilherme Vinícius Pierolo Amorin[1], Giovana dos Santos[1], Leonardo Miguel dos Santos[1]; José dos Santos Neto[1]; Vânia Moda-Cirino[1]

[1]Agronomic Institute of Paraná State - IAPAR, Celso Garcia Cid Road, Km 375, 86057-970, Londrina, PR, Brazil.

INTRODUCTION

Brazil is a large common bean producer and consumer, however the average productivity of the country is far below the productive potential of the cultivars. Many adverse biotic and abiotic factors are responsible for the low yield and instability of production. Among the abiotic factors aluminum (Al) toxicity is one of the most limiting factors for plant growth in acidic soils. Approximately one third of areas cultivated with common bean in Brazil, have high Al concentration and low fertility in soils (GIANNAKOULA, 2008). The existence of considerable genetic variability for reaction to aluminum toxicity allows the development of cultivars with desirable agronomic characteristics and more adapted to these stress conditions, contributing in an effective way to increase productivity and stability of bean production. Therefore, the identification and characterization of Al tolerant in bean genotypes represents a viable solution for bean production on the acid soils that comprise such a large fraction of the world's lands (KOCHIAN et al., 2005). The aim of this study was to evaluate common bean red commercial group genotypes reaction to aluminum toxicity.

MATERIAL AND METHODS

The experiment was carried out in the greenhouse at Agronomic Institute of Paraná State (IAPAR), Londrina - Paraná – Brazil, in November and December 2017. The experimental design was a randomized block design with eight replications and the treatments were arranged in a factorial scheme with 14 treatments, consisting of a combination of seven genotypes and two aluminum concentrations (0 and 4 ppm). The bean seedlings with height between 6 and 8 cm were selected and transplanted into polyethylene pots of 3.35 liters containing nutrient solution of Hoagland and Arnon (1950), modified by Pavan and Binghan (1982). The pH of the solution was maintained at 4.0 ± 0.2 by adjusting every two days with HCl_3 or 1N NaOH. At the V4 stage of development the plants were collected and evaluated: Maximum root length (RL), root volume (RV), plant height (PH), shoot dry matter (SDM) and root dry matter (RDM). We also calculated the Reduction Index (RI) for RL, using the expression RI (%) = {[(CSA - CCA) / CSA] x100} (MOLINA et al., 2001), where: CSA: measurement with concentration of 0 ppm of Al; CCA: measurement with concentration of 10 ppm of Al. The data were submitted to analysis of variance and Scott-Knott cluster test ($p \leq 0.05$).

RESULTS AND DISCUSSION

The RL was the variable that suffered more interference of aluminum toxicity, which can be visualized in the first days of cultivation in nutrient solution. The roots of plants grown in solution containing 4 ppm of Al did not develop normally, becoming short and thick. There was interaction between the genotype and aluminum factors only for the variable root length (Table 1). For the aluminum factor the analysis of variance revealed significant effects for RL, PH and SDM, showing that when the genotypes were submitted to the concentration of 4 ppm of Al there was a reduction in these variables, except for genotype R Line 2, which did not show significant reduction difference in root length and height when cultivated in solution with Al. For the variables

RV and RDM, all genotypes did not present a significant reduction in the presence of Al. This can be explained by the fact that despite the reduction of root length, the roots became thicker.

Table 1 - Average results of root length (RL), root volume (RV), plant height (PH), shoot dry matter (SDM), root dry matter (RDM) of common bean red commercial group genotypes grown in solution with and without the presence of aluminum.

Genotypes	RL		RV		PH		SDM		RDM	
	without Al	with Al	without Al	with Al	without Al	with Al	without Al	with Al	without Al	with Al
BRS Embaixador	36.47 Aa	22.66 Ba	4.50 Ab	3.75 Aa	82.75 Aa	63.41 Ba	2.02 Aa	1.24 Ba	0.27 Aa	0.24 Aa
DRK 15	26.81 Ab	20.13 Ba	5.12 Aa	4.12 Aa	90.77 Aa	49.35 Ba	1.90 Aa	1.39 Ba	0.25 Aa	0.23 Aa
R Line 1	28.53 Ab	22.52 Aa	4.62 Ab	4.00 Aa	89.51 Aa	66.66 Ba	2.00 Aa	1.42 Ba	0.23 Aa	0.24 Aa
R Line 2	25.93 Ab	19.73 Aa	5.00 Aa	4.50 Aa	79.93 Aa	64.23 Aa	2.15 Aa	1.28 Ba	0.26 Aa	0.22 Aa
R Line 3	35.30 Aa	22.13 Ba	5.12 Aa	4.25 Aa	80.77 Aa	67.02 Aa	2.20 Aa	1.42 Ba	0.24 Aa	0.22 Aa
DRK 18	28.93 Ab	19.35 Ba	5.87 Aa	5.00 Aa	76.63 Aa	55.35 Ba	1.99 Aa	1.33 Ba	0.27 Aa	0.24 Aa
KID 44	41.80 Aa	21.80 Ba	5.50 Aa	5.00 Aa	91.05 Aa	66.12 Ba	2.08 Aa	1.61 Ba	0.25 Aa	0.25 Aa

Means followed by different lowercase letters in columns and uppercase letter in the rows differ by F test ($p \leq 0.05$) and the Scott and Knott averages group test ($p \leq 0.05$), respectively.

The Figure 1 shows that the genotypes DRK 18, DRK 15, R Line 1 and R Line 2 were characterized by lower average reduction rates, but low root length without the presence of aluminum. On the other hand, the genotypes BRS Embaixador, R Line 3 and KID 44 showed good root growth when cultivated without aluminum, but above average reduction rates. The genotypes that more than approached the ideal quadrant (below-average reduction index and above-average root length) were R Line 3 and BRS Embaixador.

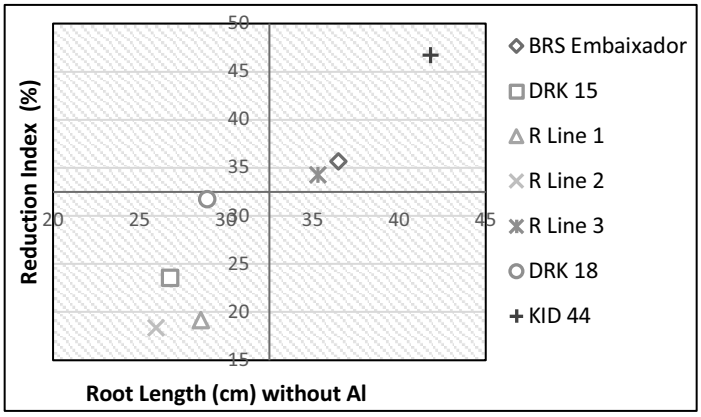


Figure 1 - Relationship between root length (cm) in condition without aluminum (0 ppm) and root length reduction index (%) when submitted to stress by Al (4 ppm) of seven red bean genotypes.

REFERENCES

GIANNAKOULA, A. et al. Aluminum tolerance in maize is correlated with increased levels of mineral nutrients, carbohydrates and proline, and decreased levels of lipid peroxidation and Al accumulation. Journal of Plant Physiology, v.165, p.385-396, 2008.

HOAGLAND, D. R.; ARNON, D. I. The water culture method for growing plants without soil. California Agricultural Experimental Station, 1950. (Circular, 374).

KOCHIAN, L. V.; PIÑEROS, M.; HOEKENGA, O. A. The physiology, genetics and molecular biology of plant aluminum resistence and toxixity. Plant and Soil, v.274, p.175-195, 2005.

PAVAN M. A.; BINGHAM F. T. Toxicity of aluminum to coffee seedlings grown in nutrient solution. Journal of Soil Science Society of América, v. 46, p.993-997, 1982.

MOLINA, J. C. et al. Response of common bean cultivars and lines to water stress. Crop Breeding and Applied Biotechnology, v.1, n.4, p.363-372, 2001.

IMPROVED OPAQUE BLACK COMMON BEAN GENTOPYES WITH ADAPTATION TO ACID SOILS

Oscar Hugo Tosquy-Valle, Rigoberto Zetina-Lezama, Francisco Javier Ibarra-Pérez and Ernesto López-Salinas

INIFAP, Campo Experimental Cotaxtla. Km 34.5 carr. Federal Veracruz-Córdoba, Medellín de Bravo, Ver., México tosquy.oscar@inifap.gob.mx

INTRODUCTION

Common bean production in tropical climates of southeastern Mexico takes place predominantly in acid soils. In the state of Veracruz, the vast majority of these acid soils are found in the area known as "Llanos de Isla and Juan Rodríguez Clara", where around 100,000 hectares have an extreme acid soil reaction, which limits plant growth and development thus reducing productivity of the bean crop (Zetina *et al*., 2002). In this production area, approximately 7,000 hectares are annually planted with tropical black bean cultivars, which include landraces, improved cultivars such as Negro Jamapa and cultivars of unknown origin of poor adaptation to acid soils (Tosquy *et al*., 2008). The objective of this research was to identify outstanding black bean genotypes for their adaptation to acidic soils and high efficiency to produce seed yield, with and without application of dolomite, as well as to determine yield components that associates the most with seed yield under such abiotic stress.

MATERIALS AND METHODS

In the fall-winter (October-January) 2016-17 season, two trials were carried out in Juan Rodríguez Clara, Veracruz, México under a very acid soil (pH=4.67) condition. One trial was conducted under stress conditions imposed by soil acidity; the other received dolomite (2.58 Mg ha^{-1}) incorporated 27 days before sowing, so that the soil pH was closed to 6.0 at harvest. Twelve bred bean lines were field evaluated and compared to improved commercial cultivars Negro Grijalva and Negro Comapa, in a RCBD with three replications each with experimental plots composed of three 5.0 m long rows 0.80 m apart. The variables measured included dry matter production without grain, 100-seed weight, number of pods per plant and grain yield. Analysis of variance was undertaken and LSD test (α=0.05) used for separation of means, simple correlations between the measured variables were performed as well. The geometric mean (GMi) (Samper and Adams, 1985) and the relative efficiency index (REIi) (Graham, 1984) were also estimated to determine the effect of soil acidity on seed yield of each bean genotype.

RESULTS AND DISCUSSION

Number of pods per plant and seed yield had the strongest effect of acid soil stress, with average reductions greater than 27% (data not shown). Result also indicated that number of pods per plant was the only trait positively correlated with seed yield, r = 0.91** and 0.87**, with and without dolomite, respectively. According to the combined analysis of variance, seed yield varied significantly between soil acidity conditions (with and without dolomite) and genotypes ($P \leq 0.01$), but not for the interaction of both factors. Table 1 shows that average seed yield obtained with the application of dolomite (667.6 kg ha^{-1}) was significantly higher than without adding dolomite (480.9 kg ha^{-1}), that corresponds to 38.8% seed yield difference. These results corroborate the susceptibility of the bean crop to soil acidity, which limits the development of plants and reduces grain yield (Zetina *et al*., 2002). Negro Citlali / XRAV-187-3-1-8 was identified as the bred line with the best adaptation to acid soils of southern Veracruz; such bred line had average seed yields,

with (911.0 kg ha^{-1}) and without (706.3 kg ha^{-1}) application of dolomite, significantly higher than the commercial cultivars used as checks. Negro Citlali / XRAV-187-3-1-8 also showed the highest index values of both, GMi (802.1) and REIi (2.0), thus indicating that this genotype has outstanding productive efficiency with and without acid soil stress. Jamapa Plus / XRAV-187-3-4-4 also showed significantly higher average seed yield and greater productive efficiency than commercial controls (Table 1).

Table 1. Average seed yield and product ivy indexes estimated under acid soils, with and without dolomite, in Juan Rodríguez Clara, Ver., México, during the fall-winter crop season yr 2016-17.

sGenotype	Seed yield (kg ha^{-1})			GMi‡	REIi††
	With dolomite	Without dolomite	Average†		
Papaloapan/SEN 46-3-7	598.0	481.7	539.8 cde	536.7	0.90
Papaloapan/SEN 46-6-6	692.7	505.3	599.0 bc	591.6	1.09
Papaloapan/SEN 46-7-7	652.7	394.3	523.5 cde	507.3	0.80
Papaloapan/SEN 46-7-11	581.7	396.3	489.0 de	480.1	0.72
Negro Citlali/XRAV-187-3-1-6	633.3	454.7	544.0 cde	536.6	0.90
Negro Citlali/XRAV-187-3-1-8	911.0*	706.3*	808.7 a	802.1	2.00
Negro Citlali/XRAV-187-3-14-6	626.7	442.3	534.5 cde	526.5	0.86
Negro Citlali/XRAV-187-3-14-7	689.0	479.7	584.3 c	574.9	1.03
Negro Citlali/XRAV-187-3-16-7	576.0	368.7	472.3 e	460.8	0.66
Jamapa Plus/XRAV-187-3-1-8	603.0	480.7	541.8 cde	538.4	0.90
Jamapa Plus/XRAV-187-3-1-2	714.7	475.0	594.8 c	582.6	1.06
Jamapa Plus/XRAV-187-3-4-4	767.0	576.0	671.5 b	664.7	1.38
Negro Comapa (check)	661.0	512.0	586.5 c	581.7	1.05
Negro Grijalva (check)	639.0	459.0	549.0 cd	541.6	0.91
Average§	667.6 *	480.9	574.2	566.6	1.02
LSD (0.05)	124.2	95.0	76.3		

†Average genotype seed yield. §Average seed yield with and without dolomite soil condition. ‡Geometric mean. ††Relative efficiency index. Genotypes with same letter(s) are statistically similar according to LSD test (0.05).

CONCLUSIONS

Negro Citlali / XRAV-187-3-1-8 was the genotype with the best adaptation to acid soil conditions in southern Veracruz, which showed the highest relative efficiency with and without application of dolomite. In both soil conditions of edaphic acidity, the number of pods per plant was the only trait that was highly and positively correlated with seed yield.

REFERENCES

Graham, R. D. 1984. Adv. Plant Nutr. 1:57-102.

Samper, C. and Adams, M. W. 1985. Ann. Rep. Bean Improv. Coop. 28: 53-54.

Tosquy, V. O. H., López, S. E., Zetina, L. R., Ugalde, A. F. J., Villar, S. B. y Cumpián, G. J. 2008. TERRA Latinoamericana 26(3):227-233.

Zetina, L. R., Pastrana, A. L., Romero, M. J. y Jiménez, Ch. J. A. 2002. Libro Técnico Núm. 10. INIFAP. CIRGOC. Campo Experimental Papaloapan y Huimanguillo. Veracruz, México. 170 p.

PHYSIOLOGICAL RESPONSES OF BLACK BEANS GENOTYPES TO ALUMINUM TOXICITY

João Marcos Novais[1]; Daniel Soares Alves[1]; José dos Santos Neto[1]; Vânia Moda-Cirino[1]

Agronomic Institute of Parana State - IAPAR, Celso Garcia Cid Road, Km 375, 86047-902, Londrina, PR, Brazil

INTRODUCTION

One of the main limitations of agricultural crops in the tropics is related to the occurrence of acid soils associated with the presence of aluminum. Approximately one third of the bean production areas in Brazil are located in regions that present soils with high aluminum concentration and low fertility, causing a reduction in the development of the plants and a decrease in productivity. The first consequence of aluminum toxicity is inhibition of root growth, which prevents the roots from exploring deeper layers of the soil, reducing the absorption of water and nutrients, especially under drought (YANG et al., 2012). The deficiency of water and nutrients causes changes in the physiological processes that are associated with bean productivity, such as stomatal movement, photosynthesis, respiration, cell division and cytoplasmic movements (TAIZ; ZEIGER, 2013). The objective of this study was to characterize the physiological responses of seven bean genotypes from the commercial black group to aluminum toxicity.

MATERIAL AND METHODS

The experiment was carried out in the greenhouse at Agronomic Institute of Paraná (IAPAR), Londrina - Paraná – Brazil, in October to November 2017. The experimental design was a randomized complete block design in a 2 x 7 factorial scheme with four replications. The first factor was composed by absence and presence of aluminum (Al) in the nutrient solution, concentration of 4 ppm, and the second factor by seven genotypes, three cultivars (BRS Esteio, IPR Inhambu and IPR Urutau) and four lines (Line 1, Line 2, Line 3 and Line 4) of the IAPAR bean breeding program. Uniform seedlings approximately 6 to 8 cm high were transplanted into 3.35 liter polyethylene pots containing nutrient solution from Hoagland and Arnon (1950), modified by Pavan and Binghan (1982), and the pH maintained at 4.0 ± 0.2. The variables analyzed were photosynthesis, transpiration and internal carbon concentration, which were measured in the morning by means of an infrared gas analyzer (ADC Bioscience ®) using photon flux density equal to 1000 μmol m^{-2} s^{-1} (NINOU et al., 2012). The evaluations were carried out at the V4 development stage, 20 days after the transplanting of the seedlings, in fully developed young leaves exposed to the sun. The data were submitted to analysis of variance and Scott-Knott cluster test ($p \leq 0.05$).

RESULTS AND DISCUSSION

Related to photosynthesis, BRS Esteio, IPR Inhambu, IPR Urutau and Line 2 did not present statistical difference among themselves and values close to the average of the control treatment, without aluminum. The genotype Line1 was below the average of the control treatment, demonstrating the effect of Al in the net assimilation of carbon (Figure 1), which can cause productivity losses during the development cycle. However, for Line 3 and Line 4 there was greater net carbon assimilation, which may indicate promising materials in relation to Al tolerance.

The highest values of internal carbon concentration observed for IPR Urutau and Line 2 genotypes may indicate aluminum effect in the development of these genotypes. The transpiration did not present significant difference in relation to the concentration of Al in solution, being close to the average of the control treatment. The levels of transpiration were ideal for a good

development of the plant, not representing physiological damage to the evaluated genotypes. This fact indicates that only the evaluation of this variable should not be used to select Al tolerant genotypes.

It is expected that the cultivation of plants in the presence of aluminum causes a decrease in photosynthesis and transpiration, however, these physiological changes may be associated with other environmental factors, such as drought (YANG; RAO; HORST, 2013).

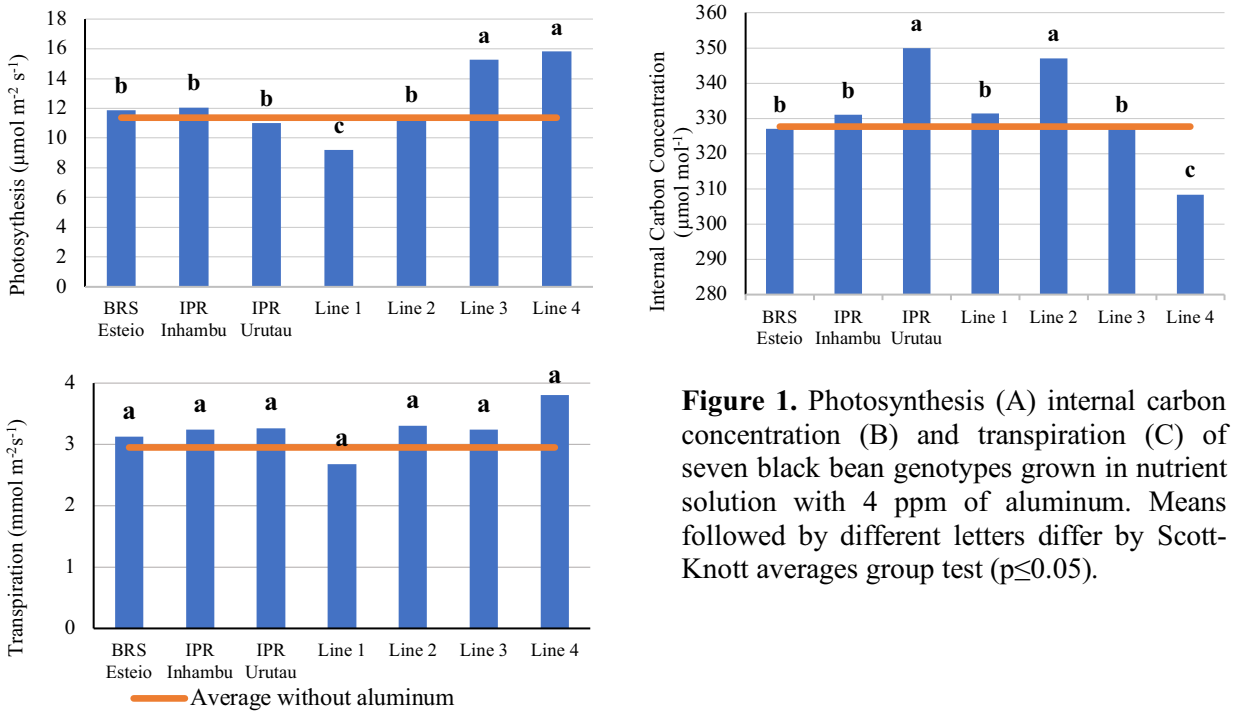


Figure 1. Photosynthesis (A) internal carbon concentration (B) and transpiration (C) of seven black bean genotypes grown in nutrient solution with 4 ppm of aluminum. Means followed by different letters differ by Scott-Knott averages group test ($p \leq 0.05$).

REFERENCES

HOAGLAND, D. R.; ARNON, D. I. The water culture method for growing plants without soil. California Agricultural Experimental Station, 1950. (Circular, 374).

NINOU, E.; TSIALTAS, J.T.; DORDAS, C. A.; PAPAKOSTA, D. K. Effect of irrigation on the relationship between leaf gas Exchange related traits and yield in dwarf dry bean grown under Mediterranean conditions. Agricultural Water Management, Amsterdam, v. 116, n. 1, p. 1-7, 2012.

PAVAN M. A.; BINGHAM F. T. Toxicity of aluminum to coffee seedlings grown in nutrient solution. Journal of Soil Science Society of América 46. 1982. 993-997.

TAIZ, L.; ZEIGER, E. Plant Physiology. 5. ed. Porto Alegre: Artmed, 2013. 954 p.

YANG, Z. B.; ETICHA, D.; ALBACETE, A.; RAO, I. M.; ROITSCH, T.; HORST, W. J. Physiological and molecular analysis of the interaction between aluminium toxicity and drought stress in common bean (*Phaseolus vulgaris*). Journal of Experimental Botany, Vol. 63, No. 8, pp. 3109–3125, 2012.

YANG, Z. B.; RAO, I. M.; HORST, W. J. Interaction of aluminium and drought stress on root growth and crop yield on acid soils. Plant Soil, v.372, n.1, p.3–25, 2013.

ECOTOXICOLOGY OF COMMON BEANS GROWN IN SOIL CONTAMINATED WITH COPPER

Engelhardt, M.M.[1], Lima, F.R.D.[1], Martins, G.C.[1], Dos Reis, R.H.C.L.[1], Cândido, G.S.[1], I.C.F.[1], Vasques, Marques, J.J.[1]

[1]Federal University of Lavras, Lavras, Minas Gerais, Brazil. email: mateusme1@hotmail.com

INTRODUCTION – The use of metals in several human activities has led to overspread and accumulation in different environmental compartments, mainly in soil. As a consequence, adverse effects on terrestrial species, ecosystem functions, and services may occur. Copper is a common metal pollutant. As also an essential metal, it is by definition necessary for various functions of organisms, animals and plants, but it is also potentially toxic.Uncontaminated soils generally have less 20 mg Cu kg^{-1}, but depending on the parent rock as much contents as high as 100 mg Cu kg^{-1} are found.

The yearly brazilian production of beans reached approximately 3.3 millions tons. The plants of *Phaseolus vulgaris* L. cv. BRSMG Madrepérola stand out mainly due to the quality of the grains, which maintain the coloration for a longer period in relation to other cultivars. Moreover, such cultivar presents high production potential and resistance to the diseases that occur in this crop.

Some plant species have been used as indicators of soil quality. The species selected for assessing soil quality should be reasonably widespread and be a important food supply. Therefore, *P. vulgaris* is a good species for such evaluation. The objective of this work was to evaluate the emergence and early development of a common bean cultivar under different doses of Cu contamination in two typical tropical soils.

MATERIALS AND METHODS – The experiment was carried out in a greenhouse following the recommendations of ISO 11.269-2. The soils used were a Dystroferric Red Latosol (LVdf) and Dystrophic Yellow Red Latosol (LVAd), which were collected in the 0-20 cm soil layer. In order create good conditions to plant growth, base saturation was increased to 70% and pH stabilized at 6. Fertilizers were applied using current recommendations. The common bean (*Phaseolus vulgaris*) variety BRSMG Madreperola was selected. Pots were filled with 500 cm^3 of dry soil contaminated with $Cu(NO_3)_2$ in the soil concentrations of 0; 75; 150; 300; 600; 800; 1000 and 1200 mg kg^{-1}. Treatments were arranged in a completely randomized design with eight treatments and four replications. In each pot, 10 seeds were planted 24 hours after the $Cu(NO_3)_2$ application. After plant emergence, thinning reduced the number of plants per pot to 5. The experiment was conducted for of 21 days, starting after 50% of germination of the control pot.

At the end of the experimental period, shoot dry weight, emergence (counted at the beginning of the test), plant hight and stem diameter were evaluated. Data normality and homogeneity were tested. The EC_{50}, concentrations that reduce endpoints by 50% when compared with control, were estimated through non-linear and linear models. All analyses were performed using STATISTICA v. 7.

RESULTS AND DISCUSSION – The ecotoxicological variables tested indicated sensitivity to Cu, be the emergency parameters followed by shoot dry weight those with lower values of EC_{50} (Table 1). Plant emergence was considered the most affected variable, possibly because it is the first target of heavy metal toxicity, and the seedling has less defense mechanisms than the adult plant. Shoot dry weight was affected by Cu due to a decrease in the photosynthetic efficiency. Copper interferes in the electron transport chain of photosystem I, causing depression in the growth

of sensitive plants. Stem diameter and plant height presented higher values of EC_{50} (Table 1) and were less sensitive to Cu contamination.

The LVdf presented lower values of EC_{50} compared to LVAd, probably due to higher clay and organic matter contents (Table 2), soil properties that most influence Cu availability. Copper is cationic and prone to covalent bonding especially to negative sites in organic compounds.

Table 1. EC_{50}, effect concentration (with 95% confidence intervals in parentheses) for Cu toxicity Cu to *Phaseolus vulgaris* L. cv. BRSMG Madrepérola grown in different soils.

Endpoint	LVdf			LVAd		
	EC_{50}	R^2	Model	EC_{50}	R^2	Model
	mg kg^{-1}			mg kg^{-1}		
Emergence	335 (444 - 226)	0,92	Gompertz	651 (785 - 518)	0,84	Gompertz
Shoot dry weight	481 (571 - 391)	0,94	Hormesis	676 (724 - 627)	0,96	Hormesis
Stem diameter	701 (815 - 585)	0,84	Linear	949 (1134 - 764)	0,83	Logistic
Shoot length	949 (1134 - 764)	0,76	Hormesis	760 (904 - 617)	0,81	Linear

Dystroferric Red Latosol (LVdf); Dystrophic Yellow Red Latosol (LVAd)

Table 2. Physical atributes and organic matter of Dystroferric Red Latosol (LVdf) and Dystrophic Yellow Red Latosol (LVAd) used in the laboratory test.

Properties	LVAd	LVdf
Organic Matter (dag kg^{-1})	2,11	3,99
Clay (dag kg^{-1})	31	68
Silt (dag kg^{-1})	11	13
Sand (dag kg^{-1})	58	19

CONCLUSIONS – The plants grown in LVdf were less affected by Cu than the ones grown in LVAd. Seedling emergence and shoot dry weight were the variables most sensitive to Cu pollution. Total Cu contents above 335 mg kg^{-1} are likely to reduce plant growth in at least 50 %.

ACKNOWLEDGMENTS – The authors thank the CAPES, CNPq, and FAPEMIG for financial support.

PHOTOSYNTHETIC EFFICIENCY THROUGH THE IRGA LI-6400XT OF COMMON BEANS cv. BRSMG MADREPÉROLA IN SOIL CONTAMINATED WITH COPPER

Engelhardt, M.M.[1], Lima, F.R.D.[1], Vasques, I.C.F.[1], Oliveira, C. [1], Martins, G.C.[1], Dos Reis, R.H.C.L.[1], Cândido, G.S.[1], Castro, E.M. [1], Marques, J.J.[1]

[1]Federal University of Lavras, Lavras, Minas Gerais, Brazil. email: mateusme1@hotmail.com

INTRODUCTION – Copper is a micronutrient in plants and its function is related to photosynthesis rate, respiration, metabolism of C and N, and also protection against oxidative stress. The major role of Cu functionality is based on enzymatically bound Cu working as a catalyzer in redox reactions. However, in greater Cu concentrations, it is reported to be phytotoxic. Therefore, it is important to establish relations between soil chemical characteristics and Cu uptake by plants.

Common beans are an important food supply, especially in Brazil, where it represents a great part of the daily Brazilian diet. Many cultivars have been developed in order to increase quality and productivity. BRSMG Madrepérola appears to be a good cultivar, with high productive potential and great resistance to diseases and insects. For these reasons, cv. BRSMG Madrepérola was chosen to evaluate photosynthetic rate, transpiration rate, stomatal conductance to H_2O, and intercellular CO_2 concentration and their alterations with increasing concentrations of Cu in soil.

MATERIALS AND METHODS – The plants were cultivated in a greenhouse following the recommendations of ISO 11.269-2. The soils used were a Dystroferric Red Latosol (LVdf) and a Dystrophic Yellow-Red Latosol (LVAd), which were collected in the 0-20 cm soil layer. Fertilizers were applied using current recommendations. The common beans (*Phaseolus vulgaris* L.) BRSMG Madrepérola cultivar was selected. Pots were filled with 500 cm^3 of dry soil contaminated with $Cu(NO_3)_2$ in the soil concentrations of 0; 75; 150; 300; 600; 800; 1000 mg kg^{-1}. Treatments were arranged in completely randomized design with eight treatments and four replicates. In each pot, 10 seeds were planted 24 hours after $Cu(NO_3)_2$ application. The experiment was conducted for 21 days, starting after 50% of seedling emergence of the control pot. Evaluated variables were: photosynthetic rate (A), transpiration rate (E), stomatal conductance to H_2O (g_{sw}), and intercellular CO_2 concentration (C_i). Measurements were conducted with an infrared gas analyzer (IRGA) model LI-6400 XT at 10:00 AM and the photon flux of photosynthetic radiation was standardized at 800 µmol m^{-2} s^{-1} in the equipment chamber. The evaluations were made in young leaves completely expanded and pathogen-free. Data were submitted to variance analysis (ANOVA, $p < 0.05$) and also submitted to regression analysis ($p < 0.05$).

RESULTS AND DISCUSSION – The variables related to gas exchange, A, E, and g_{sw}, did not present significant differences with increasing Cu concentration in common beans, in both soil (Figure 1). Significant differences were observed only for C_i, when plants were grown in LVAd, in the highest Cu concentration (1000 mg kg^{-1}). In this case, there was a 30% increase in C_i when compared to the control treatment (Figure 2).

The significant increase in C_i in LVAd showed carbon assimilation potential under these conditions. However, this potential was not enough to allow the greatest concentration of CO_2 to result in a higher photosynthetic rate. However, higher carbon assimilation is one component of the photosynthetic rate, not the only one, that directly increases the photosynthetic rate. The increase of C_i in higher concentrations of Cu may be due to structural differences caused by the presence of Cu, for example, increase in density or stomata size, being responsible for a higher concentration of CO_2 within the leaves at these concentrations. However, since there was no significant difference for stomatal conductance, it is likely that another type of structural modification would explain this greater accumulation of internal CO_2 in common bean leaves at higher concentrations of Cu, such as lower diffusion resistance CO_2 into the common bean leaves at these concentrations of Cu, caused by the deposition of cuticle components. Another structural modification that would result in a higher CO_2 concentration in this case would be a smaller number of chlorophyllic parenchyma cells, especially the spongy one, which, due to its smaller amount, could represent a lower

resistance to diffusion and accumulation of this higher concentration of CO_2 collected and accumulated in common leaf bean tissue. Copper can directly affect the process of cell division, and may, at high concentrations, be responsible for decreasing the number of cells in plant tissues.

That reason for this behavior has been verified only in the LVAd could be related to the fact that Cu would be more available to the plant in this class of soil; and a greater concentration absorbed would result in these effects being observed, which was not possible in a situation where Cu would be less available to plants as in the LVdf.

Figure 1. Photosynthetic rate, transpiration rate and stomatal conductance to H_2O of common beans cv. BRSMG Madrepérola grown in LVAd (A) and LVdf (B) with increasing Cu concentrations. Mean standard deviation is represented by bars (n = 4).

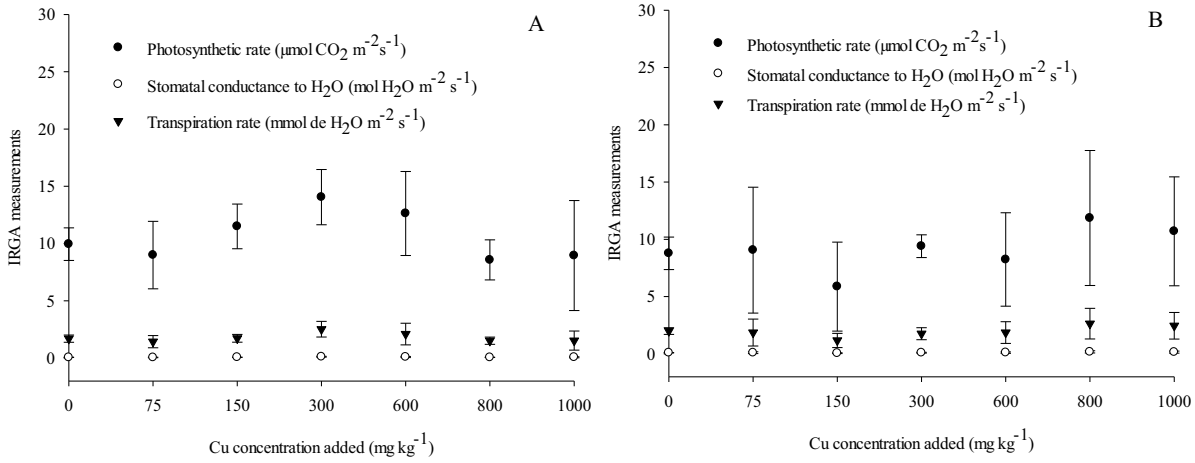
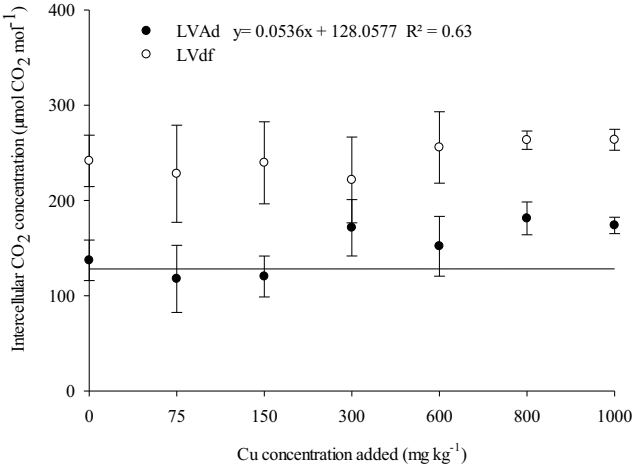


Figure 2. Intercellular CO_2 concentration of common beans cv. BRSMG Madrepérola cultivated in LVAd and LVdf with increasing Cu concentrations. Mean standard deviation is represented by bars (n = 4).



CONCLUSIONS – Copper concentration up to 1000 mg kg^{-1} does not interfere in photosynthetic rate, transpiration rate, and stomatal conductance to H_2O in common beans variety BRSMG Madrepérola cultivated in the soil classes LVAd e LVdf. However, the intercellular CO_2 concentration is increased when grown in the soil with lower Cu adsorption capacity, the LVAd.

MACRONUTRIENTS ACCUMULATION IN COMMON BEAN cv."BRSMG MADREPÉROLA" ON SOIL CONTAMINATED WITH MERCURY

Lima, F.R.D.[1], Engelhardt, M.M.[1], Vasques, I.C.F.[1], Silva, A.O. [1], Martins, G.C.[1], Dos Reis, R.H.C.L.[1], Pereira, P. [1], Cândido, G.S.[1], Oliveira, J.R., Marques, J.J.[1]

[1]Federal University of Lavras, Lavras, Minas Gerais, Brazil. email: frandislim@gmail.com

INTRODUCTION – Mercury in environment can pose many adverse effects upon entire ecosystems. In plants, Hg may cause reduced growth due to many metabolic pathways that are affected. Mercury in plants can reduce photosynthetic rate and water absorption, nutrient absorption, and inhibit protein synthesis.

Common beans are cultivated throughout Brazil under a variety of environmental conditions. "Carioca" beans are preferred among consumers, representing 79% of the beans consumed in Brazil. The cv. BRSMG Madrepérola produces high quality "Carioca" beans. The BRSMG Madrepérola cultivar is semi-precocious, taking 85 days from seedling emergence to physiological maturation. It also exhibits resistance to the main diseases, including common mosaic virus and various anthracnose breeds. This study aimed to verify the effects of Hg on macronutrients accumulation (P, K, Ca, Mg, and S) in common beans cv. BRSMG Madrepérola cultivated in two remarkably different soils.

MATERIALS AND METHODS – The experiment was carried out in a greenhouse. A Dystroferric Red Latosol (LVdf) and a Dystrophic Red-Yellow Latosol (LVAd) were used. Samples were collected from the 0-20 cm soil layer. Base saturation was increased to 60% and pH stabilized at 6. It was chosen the common beans (*Phaseolus vulgaris* L.) variety BRSMG Madrepérola. Mercury concentrations of 0; 2.5; 5.0; 10.0; 20.0; 40.0 and 80.0 mg kg^{-1} in the soil were applied to 500 cm^3 of dry soil in the form $HgCl_2$ solutions. A completely randomized design was set, with seven treatments and four replications. Fertilization was made according to recommendations. Ten seeds were sown in each pot 24 hours after the $HgCl_2$ application. After emergence, and the number of plants per pot was thinned to six. The experiment lasted 21 days after 50 % of seedling emergence in the control pot.

After the experiment, the plants were harvested, dried and weighted. Macronutrients in the biomass were analyzed according to standard procedures. Nutrient accumulation was obtained by multiplying concentration by biomass dry weight. Data were submitted to analysis of variance (ANOVA, $p < 0.05$) and those that presented a significant difference, also submitted to the regression analysis ($p < 0.05$).

RESULTS AND DISCUSSION – As the plants did not produce enough dry matter, it was decided not to carry out the N determination because it would require a different, sample-consuming analytical route. Accumulation of Ca and Mg in plants decreased with increasing Hg concentrations in both soils, whereas P and K decreased only when grown in the LVAd. However, sulfur accumulation was not sensible to Hg doses in both soils (Table 1).

Those results can be ascribed to interference of Hg in macronutrient accumulation. Mercury can compete with nutrients during transportation, occupying the transmembrane nutrient transporters. Affinity between Hg and S has been reported in previous studies. Mercury can be absorbed and carried in plants by complexes between Hg and S, through sulfate transporters. It is suggested that S is absorbed alongside Hg.

In the LVdf, P and K accumulation was not affected by Hg concentrations, possibly due to soil features that favor Hg retention, such as higher iron oxides contents. However, when grown in LVAd, it was observed a linear decreasing on P and K accumulation by plants (Table 2). Thus, the accumulation of Ca and Mg shows to be more influenced by soil Hg contents than P and K.

Table 1. Variance analysis results for macronutrients accumulation P, K, Ca, Mg and S ($p < 0.05$) in cv. BRSMG Madrepérola grown in LVAd and LVdf contaminated with Hg, (± mean standard deviation, n = 4).

Hg concentration added	P	K	Ca	Mg	S
mg kg^{-1}	---------- mg pot^{-1} ----------				
	Common beans in LVAd				
0	6.1 ± 0.66	35.87 ± 6.69	14.75 ± 4.53	6.05 ± 0.86	3.45 ± 2.43
2.5	4.41 ± 0.94	30.5 ± 6.48	10.03 ± 1.95	3.85 ± 0.65	3.12 ± 0.45
5	3.85 ± 0.35	29.54 ± 6.02	8.36 ± 1.52	3.8 ± 0.74	4.02 ± 0.55
10	4.09 ± 1.02	27.53 ± 6.04	9.9 ± 0.46	4.16 ± 0.25	1.94 ± 0.6
20	4.99 ± 1.25	29.96 ± 8.74	9.79 ± 2.2	4.74 ± 0.74	3.83 ± 1.83
40	2.18 ± 1.05	20.32 ± 11.05	2.73 ± 0.88	1.88 ± 0.78	3.42 ± 1.28
80	1.78 ± 0.69	11.9 ± 2.58	1.73 ± 1.15	1.19 ± 0.61	2.39 ± 0.55
ANOVA	*	*	*	*	**ns**
	Common beans in LVdf				
0	5.77 ± 3.06	32.22 ± 18.41	23.87 ± 14.62	9.06 ± 4.89	3.42 ± 1.12
2.5	3.09 ± 0.93	15.5 ± 2.85	11.46 ± 2.22	4.55 ± 0.84	1.99 ± 0.35
5	3.83 ± 1.17	15.79 ± 3.1	10.6 ± 3.59	3.99 ± 1.08	1.64 ± 0.47
10	3.14 ± 0.51	17.94 ± 1.39	11.86 ± 0.4	4.7 ± 0.11	1.78 ± 0.17
20	2.78 ± 0.59	14.65 ± 2.16	10.14 ± 2.69	3.96 ± 0.62	2.38 ± 0.57
40	2.87 ± 0.69	22.39 ± 8.18	9.84 ± 2.24	4.19 ± 1.25	3.32 ± 1.42
80	2.76 ± 1.55	19.34 ± 7.04	5.96 ± 1.29	3.53 ± 1.15	2.87 ± 0.63
ANOVA	**ns**	**ns**	*	*	**ns**

Table 2. Estimation of adjusted regression models for the macronutrients accumulation that showed significant differences between treatments ($p < 0.05$).

Macronutrients	Regression equation	R^2
	Common beans in LVAd	
P	$y = - 0.0433x + 4.8897$	0.6786
K	$y = - 0.2633x + 32.4423$	0.916
Ca	$y = 0.0019x^2 - 0.2869x + 12.3135$	0.8185
Mg	$y = - 0.0485x + 4.7609$	0.7132
	Common beans in LVdf	
Ca	$y = - 0.1224x + 14.7157$	0.3982
Mg	$y = - 0.0315x + 5.5623$	0.2298

CONCLUSIONS – Increasing Hg concentration in soil provides lower K, P, Ca, and Mg accumulation in plants. Mercury in soils did not influence S accumulation in common beans cv. BRSMG Madrepérola. In soils with high Hg adsorption capacity, accumulation of K and P by common beans was not affected by increasing Hg doses.

ACKNOWLEDGMENTS - The authors thank the CAPES, CNPq, and FAPEMIG for financial support.

MICRO-NUTRIENT CONTENT IN CRUDE SEEDS OF BEAN LANDRACES BEANS GROWN IN TWO REGIONS

Carla Xavier Alves[1]; Gilberto A Peripolli Bevilaqua[2], Irajá Ferreira Antunes[2]; Luis Osmar Braga Schuch[1], Paulo Eduardo Rocha Eberhardt[1]

[1]Departamento de Fitotecnia, Faculdade de Agronomia, Universidade Federal de Pelotas, Pelotas, RS; [2]Embrapa Clima Temperado, Pelotas, RS

INTRODUCTION

The common bean plant is very demanding in nutrients due to the small and shallow root system and its short cycle. In addition, nutrients need to be readily available to the crop in times of demand so as not to limit productivity. The seeds, similar to the other organs of plants, have a very variable chemical composition because it is an organ formed at the end of the plant cycle. The concentration of mineral micronutrients in the seeds can also affect the biological fixation of atmospheric nitrogen, especially in the case of legumes (JACOB NETO E FRANCO, 1989).

Micronutrients, such as iron, manganese and zinc, are determinants or components of several processes, such as protein synthesis, membrane permeability, ion absorption, respiration, and starch synthesis and hormone control. Pereira et al. (2009) and Piegas et al. (2011) evaluating the content of nutrients in landraces beans in the State of Santa Catarina and Rio Grande do Sul, respectively, observed genotypes that stood out for the micronutrient contents. The goal of this study was to evaluate the iron, manganese and zinc content in native cultivars from Rio Grande do Sul and their interaction with two planting environments.

MATERIAL AND METHODS

The seeds were obtained from the evaluation of cultivars of Embrapa Clima Temperado cultivars implanted in two cultivation sites: Sobradinho and São Luiz Gonzaga, in Rio Grande do Sul. 12 genotypes were evaluated, mostly black beans, except Amarelinho of yellow grain, TB 0226 (red) e Vinho 141 (purple). The controls used were BRS Intrépido (Black check) and Carioca (color check). The iron, manganese and zinc contents were evaluated in whole seeds according to Silva (1999). Variance analysis was performed for cultivation site, genotypes, and a comparison test of means to evaluate the genotypes at each culture site.

RESULTS AND DISCUSSION

The analysis of variance indicated that there was a significant interaction between genotypes and culture environment for the nutrient content analyzed. Fe and Zn contents were lower in São Luiz Gonzaga, unlike Mn. It is known that both water stress and high temperature during the period of grain filling may be possible explanations for the variations in nutrient concentration fact observed mainly in São Luiz Gonzaga.

Fe levels in Sobradinho and São Luiz Gonzaga (Table 1) ranged from 0.08 to 0.12 $g.kg^{-1}$ and 0.06 to 0.10 $g.kg^{-1}$ respectively. These values are similar to those found by Pereira et al., (2011) evaluating genotypes in two environments. However, higher than those found in Colombia (BEEBE et al., 2000). The AM-10 genotype showed the highest levels at both sites, although not differing from other genotypes. In Sobradinho, the TB 02-20 genotype presented a similar value to AM-10. In São Luiz Gonzaga, all genotypes were statistically similar and inferior to AM-10.

The genotypes ZL-1 and Preto Ibérico showed the highest levels for zinc (Zn) and manganese (Mn) in seeds from Sobradinho, as well as the AM-10 genotype was the highest in São Luiz Gonzaga.

According to Lemos et al. (2004), the nutritional characteristics are influenced by both genotype and environmental conditions in plant and seed development. Pereira et al. (2009) observed a significant reduction in the accumulation of some nutrients in the environment that presented high precipitation, in the pod formation phase, similar to what happened in São Luiz Gonzaga, where the grains presented lower levels for most nutrients.

Table 1 - Levels of zinc (Zn), manganese (Mn) and iron (Fe) in seeds of bean landraces cultivated at two sites, Sobradinho (SOB) and São Luiz Gonzaga (SLB), RS, Brazil.

	Fe (g\kg)		Zn (mg\kg)		Mn (mg\kg)	
Genotypes	SOB	SLB	SOB	SLB	SOB	SLB
AM-10	0.11 A ab	0.10 A a	28.27 B defg	32.58 A a	16.88 B bc	20.90 A a
Amarelinho	0.09 A cdef	0.06 B b	30.57 A bcde	24.30 B fg	16.70 A bcd	16.32 A de
AS-7	0.10 A bcde	0.08 B b	26.27 B fg	29,53 A b	15.68 B cde	17.91 A c
BRS Intrépido(C)	0.09 A cde	0.07 B b	29.20 A cdef	26.00 B def	14.68 B e	19.34 A b
Carioca (C)	0.09 A def	0.08 A b	30.76 A bcde	26.55 B bcdef	16.37 B bcd	19.43 A b
CK-4	0.08 A ef	0.06 B b	31.10 A bcd	24.74 B efg	14.65 A e	15.22 A ef
Guabiju	0.08 A f	0.08 A b	27.14 A fg	27.45 A bcde	13.31 B f	19.73 A b
Preto Ibérico	0.10 A abc	0.07 B b	33.30 A b	29.33 B BC	17.03 A b	16.88 A cd
TB 02-20	0.12 A a	0.07 B b	31.67 A bc	26.44 B cdef	16.90 A bc	13.98 B fg
TB 02-21	0.10 A bcde	0.08 B b	27.97 A efg	22.57 B g	15.56 A de	13.93 B g
TB 02-25	0.10 A bcd	0.07 B b	26.00 A g	28.47 A bcd	14.71 A e	15.06 A fg
TB 02-26	0.09 A def	0.06 B b	28.75 A cdefg	28.58 A bcd	14.97 A e	14.43 A fg
Vinho 141	0.10 A bcd	0.07 B b	32.25 A b	27.23 B bcdef	16.66 A bcd	17.63 A c
ZL-1	0.10 A bcd	0.07 B b	37.22 A a	28.65 B bcd	18.61 A a	19.55 A b
Average	0.10	0.07	30.03	27.32	15.90	17.17
CV%	10.3		5.6		4.2	

Averages followed by the same capital letter in the row and lower case letters in the column do not differ by Duncan's test at the level of 5%.

CONCLUSIONS

There is interaction between genotype and environment for nutrient content in whole grains. In Sobradinho, the ZL-1 genotypes are distinguished for high levels of Zn and Mn and TB 02-20 for Fe content. In São Luiz Gonzaga, the AM-10 genotype stands out for Fe, Zn and Mn.

REFERENCES

BEEBE, S.; GONZALEZ, A.V.; RENGIFO, J. Research on trace minerals in the common bean. Food and Nutrition Bulletin, Boston, v.21, n.4, p.387-391, 2000.

JACOB-NETO, J.; FRANCO, A.A. Determinação do nível críticos do Mo nos nódulos do feijoeiro (*Phaseolus vulgaris* L.). Turrialba, vol.39 n.2, p. 215-223, 1989.

LEMOS, L.B.; OLIVEIRA, R.S.; PALOMINO, E.C. et al. Características agronômicas e tecnológicas de genótipos de feijão do grupo comercial Carioca. Pesquisa Agropecuária Brasileira, v. 39 319-326, 2004.

PEREIRA, T.; COELHO, C.M.M.; BOGO, A. et al. Diversity in common bean landraces from South-Brazil. Acta Botanica Croatica, v.68, n.1, p. 79-92, 2009.

PIEGAS, B.N.; BEVILÁQUA, G.A.P.; ANTUNES, I.F. et al. Oligoelement levels in common bean breeding lines and landraces from Rio Grande do Sul, Brazil. BIC 54, 2011.

SILVA, F. C., Manual de análises químicas de solos, plantas e fertilizantes / Embrapa Solos, Embrapa Informática Agropecuária – Brasília: Embrapa Comunicação para transferência de tecnologia, 1999. 370 p.

ACCUMULATION OF NUTRIENTS IN SNAP BEAN

Luiz H.C. Almeida1, Renan R. Barzan1, Natalia F. Carr1, Guilherme R. Gomes1, Eli C. Oliveira1, Claudemir Zucareli1, Gustavo A. F. Fregonesi1

[1]State University of Londrina Londrina - UEL, Brazil

INTRODUCTION

The nutrient uptake gait during the bean pod (*Phaseolus vulgaris* L.) cycle provides important information related to the amount of nutrients absorbed and the intensity of absorption of each nutrient. It is possible to apply fertilizers at the most appropriate time. Due to the importance and the scarcity of research on the nutrient absorption requirement of the bean pod, this work had the objective of evaluating the nutrient uptake during the crop cycle, in potted planting with fertirrigation.

MATERIAL AND METHODS

The experiment was carried out in a greenhouse with transparent polyethylene cover, from the State University of Londrina, Brazil (23 ° 23'S, 51 ° 10'W), between September and December 2017. Conducted in 5-liter pots of sand with five seeds sown and after the emergency the thinning was carried out for two plants. The spacing used was 0.50 meters between rows, with seven rows and 12 pots in each row, with a total of 84 pots. Cultivars were of determined habit: UEL-1 and Alessa. Fertilizers were used: MAP (200g $1000L^{-1}$); Ca(NO3)2 (800 g $1000L^{-1}$); CaCl2 (300g $1000L^{-1}$); MgSO4 (300g $1000L^{-1}$) e KNO3 (400g $1000L^{-1}$). The micronutrients were supplied by Rexolin BRA® 30 g $1000L^{-1}$ and Rexolin M48® with the 30 g L^{-1} solution, complementing the Fe content. The drip system consisted of pumps, model BCR 2000 from SCHNEIDER, connected to a timer programmed to irrigate for 40 seconds. Each dripper was set at a maximum flow rate of 300 mL min^{-1}. The irrigation shift was defined based on climatic characteristics - temperature, relative humidity, and crop characteristics, ranging from one to five times a day. The plants were collected for analysis every 10 days. All the collected tissues, including the pods, were placed in a forced air circulation oven at 55 ºC until reaching a constant dry mass(DM). For laboratory analysis of nutrient uptake the plants were collected and separated into leaves, stems, roots and pods and performed according to Malavolta et al. (1997). With the data of the total accumulation of nutrients in each collection in each part of the plant, the relative accumulation and the productivity of the fresh pods in kg ha^{-1} were obtained.

RESULTS AND DISCUSSION

The yield of the cultivars UEL 1 and Alessa were, respectively, 20,702 kg ha^{-1} and 15,217 kg ha^{-1}, a value within the yield range obtained in bean pod production cultivated in the soil in a period of mild temperatures and high precipitation by Oliveira et. al (2007) from 13,000 to 25,000 kg ha^{-1}. Table 1 shows the accumulation in kg ha^{-1} of the macronutrients. The accumulation of DM was slow until 20 days after emergence (DAE) when plants were in the V4 stage, UEL 1 and Alessa accumulated respectively 12 and 13% of the maximum DM value, which was mainly allocated in the leaves. The significant relative accumulation was from 40 DAE, the plants were at the R7 stage, the appearance of the pods and more egalitarian distribution between leaf, stem, and root. As stated by Brito (1992), the accumulation of DM increases significantly at each stage of the cultural cycle, being greater during the period of pod filling and physiological maturation.

Table 1: Accumulation of nutrients in the different organs of the plant during the cultural cycle, UEL 1 and Alessa, respectively.

	UEL 1						ALESSA					
	N	**P**	**K**	**Ca**	**Mg**	**S**	**N**	**P**	**K**	**Ca**	**Mg**	**S**
	Kg ha^{-1}						**Kg ha^{-1}**					
Roots	1,11	0,32	0,95	0,57	0,09	0,26	1,54	0,33	0,89	0,33	0,08	0,22
Leaves	7,19	0,54	4,37	5,02	0,39	0,27	6,67	0,62	3,95	5,55	0,5	0,42
Stems	5,71	1,06	8,5	3,85	0,27	0,67	7,78	0,89	10,68	4,79	0,31	0,75
Pods	12,74	1,09	9,69	1,05	0,52	0,36	10,78	1,45	5,62	0,87	0,42	0,42
Total	26,75	3,01	23,51	10,5	1,26	1,56	26,77	3,28	21,15	11,54	1,31	1,81

According to El-Husny (1992), macronutrient extraction by common bean follows the decreasing order: N, K, Ca, Mg, S, and P. In this work the order changes in the last three nutrients: N, K, Ca, P, S, Mg. The order of extraction of the nutrients was equal in the two cultivars used in the experiment. The N was the nutrient most required by the bean pod during the whole cycle, followed by K accumulating in greater quantity in the stem and pod UEL 1 and in the cultivar Alessa, stem, and pod. Thirdly, in the extraction, Ca had a greater accumulation in the leaf in cultivar UEL 1 and Alessa. The P had an accumulation in the cultivar UEL 1 of 3.01 kg ha^{-1} and in the cultivar Alessa of 3.28 kg ha^{-1}. The S accumulated in UEL 1 was approximately 1.56 kg ha^{-1}, the part of the plant where the largest amount was in the stems, comparing with the cultivar Alessa that had a total accumulation of 1.81 kg ha^{-1} with higher concentration in the stems too. Finally, Mg had a total accumulation of 1.26 kg ha^{-1} in UEL 1, with a higher leaf allocation (0.39 kg ha^{-1}) and in Alessa of 1.31 kg ha^{-1}, with the highest accumulation between leaf and pod with 0.5 kg ha^{-1}. Malavolta et al. (1997) stated that the filling of the bean pods must occur in part by the redistribution of the N, P and K content in the leaves, the content of which decreases. According to Marschner (2012), the low redistribution of Ca occurs due to the small mobilization capacity in the phloem, leading to the increase of its contents in vegetative organs, especially in those with a higher transpiration rate. The Ca remains until the end of the cycle well present in the leaf and stem, where a small amount can reach the pod.

CONCLUSION

The decreasing order of extraction of the macronutrients of the bean pod in the two cultivars analyzed, UEL 1 and Alessa, was N, K, Ca, P, S, Mg. Take into account the longer absorption period for adequate fertilization, especially N, K and Ca, which are the most required by the plant.

REFERENCES

BRITO, M.M.P. Marcha de absorção de nitrogênio do solo, do fertilizante e da fixação biológica em caupi (*Vigna uniculata* (L.) Walp.) e feijão comum (*Phaseolus vulgaris* L.) determinada usando 15N. 1992. 197 p. Dissertação (Mestrado em Fitotecnia)-Escola Superior de Agricultura Luiz de Queiroz, Piracicaba, SP.

EL-HUSNY, J.C. Limitações nutricionais para a cultura do feijoeiro (*Phaseolus vulgaris* L.) em um solo do Norte de Minas Gerais. 1992. 151 p. Dissertação (Mestrado em Fitotecnia)-Universidade Federal de Lavras, Lavras, MG.

MALAVOLTA, E.; VITTI, G.C.; OLIVEIRA, A.S. Avaliação do estado nutricional das plantas: princípios e aplicações. Piracicaba, ed. Potafós, 1997.

MARSCHNER, H. Mineral nutrition of higher plants. 3.ed. London: Academic Press, 2012.

LEAF AREA INDEX OF THE SNAP BEAN WITH DETERMINED GROWTH HABIT ACCORDING TO THE PLANTS ARRANGEMENT

Guilherme Renato Gomes; Gustavo Henrique Freiria; Luiz Henrique Campos de Almeida; Douglas Mariani Zeffa; Eli Carlos de Oliveira and Lúcia Sadayo Assari Takahashi

State University of Londrina (UEL)

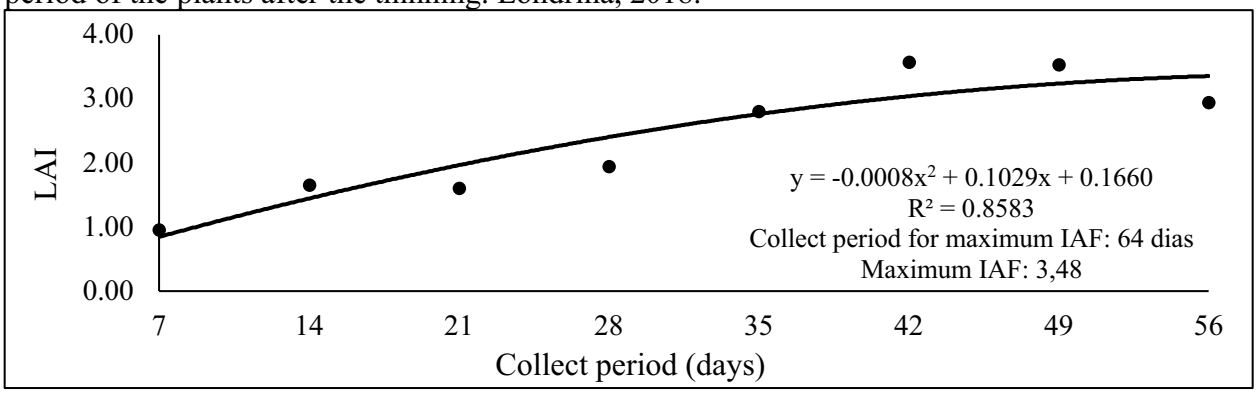
INTRODUCTION: The variation of the spacing between lines and the number of plants per hectare determines the arrangement of the plants by area. One of the objectives of the alteration of the plant arrangement is maximize the interception of the incident radiation, aiming the greater use of the energy absorbed for photosynthesis. One of the factors that affect the interception of the incident radiation is the leaf area index, which deals with the relation between m² of leaf per m² of soil. As higher this ratio, higher will be the intercepted radiation and, consequently, the leaf area index. For the snap bean with determined growth habit there are few the recommendations of spacing and number of plants by area to be used. Thus, the objective was to verify if leaf area index of the determined bean-pod of growth is modified according to different plant arrangements.

MATERIAL AND METHODS: The experiment was conducted in Londrina, State of Paraná, Brazil, with sowing in March 2017. The cultivar Feltrin Macarrão Napoli® was used. Two weeks after sowing the plants were thinned in order to reach the desired populations. Populations of 133333, 177778, 222222, 266667 and 311111 plants per hectare were tested associated with the 0.45 and 0.90 meter spacings between rows. The experimental units were five meters long. The experimental design was a randomized complete block design, in a factorial scheme 5 (plant populations) x 2 (spacings between rows), with three replications. The leaf area index (LAI) was indirectly estimated by the LA-2000 canopy analyzer model LAI-2000 (LI-COR, 1992). After the thinning, and in each experimental unit, the measurements were performed weekly until the pods were harvested (56 days after the thinning), always at dusk, and consisted of a reading above the canopy (reference reading) and three readings performed in the sowing line, below the canopy (soil level), totaling 4 readings per experimental unit. The apparatus estimated the leaf area index based on mathematical equations, considering the proportion of the radiation that reached the soil in relation to which was intercepted by the canopy of the snap bean plants. Statistical analysis was performed by applying the F test, and the plant populations, as well as the collect periods (considered source of variation) of the plants submitted to regression analysis ($p < 0.05$).

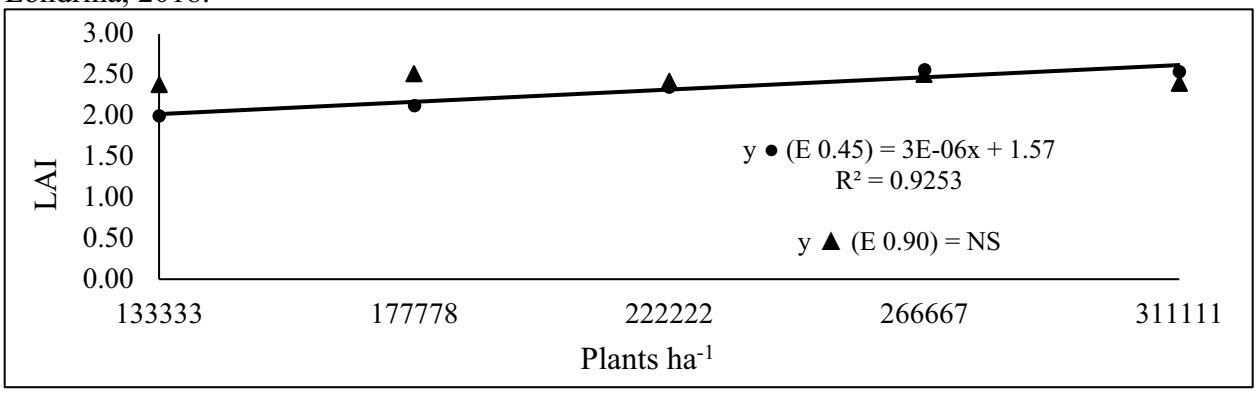
RESULTS AND DISCUSSION. There was isolated significance for the collect period, as well as significant interaction between number of plants per hectare and spacing between lines. There was a polynomial adjustment of the leaf area index according to the increment in the collect period of the snap bean plants, with estimated maximum LAI of 3.48 in the estimated maximum collect period at 64 days after the thinning (Graph 1). The results showed that the harvesting of the pods did not correspond to the senescence period of the plants, showing that the significant loss of their leaf area occurs after 64 days. Graph 2 showed that the LAI increased linearly with the increment of the number of plants per hectare associated only to the spacing of 0.45 meters between rows. The results of Table 1 showed a significant difference for the LAI means in the spacing of 0.90 meters between rows in the populations of 133333.0 and 177777.0 plants per hectare, evidencing the effect of compensation of the snap bean plants.

CONCLUSION: Regardless of the row spacing, the maximum leaf area index for the bushing snap bean, cv. Feltrin Macarrão Napoli® occurs at 64 days post thinning. The increment of the number of plants per hectare associated only to the spacing of 0.45 meters between rows increase linearly the leaf area index of bushing snap bean, cv. Feltrin Macarrão Napoli®.

Graph 1. Leaf area index of the snap bean with determined growth habit according to collect period of the plants after the thinning. Londrina, 2018.



Graph 2. Deployment of the interaction of factor number of plants per hectare as a function of the factor between lines for the index of leaf area of the snap bean with determined growth habit, Londrina, 2018.



*NS: not significant

Table 1. Split of the interaction of the factor spacing between lines in function of the factor number of plants per hectare for the index of leaf area of the snap bean with determined growth habit, Londrina, 2018.

Spacing between lines (m)	Plants ha^{-1}				
	133333,0	177777,0	222222,0	266666,0	311111,0
0,45	2.01 b	2.13 b	2.36 a	2.56 a	2.54 a
0,90	2.38 a	2.52 a	2.42 a	2.51 a	2.39 a

REFERENCES

LI-COR. Plant canopy analyzer: LAI-2000; operating manual. Lincoln: 1992. 179 p.

PRODUCTIVITY AND NUMBER OF NODULES IN BEANS CO-INOCULATED WITH AZOSPIRILLUM BRASILIENSE AND RHIZOBIUM IN AGROECOLOGICAL CULTIVATION SYSTEM

Luiz H.C. Almeida1, Eli C. Oliveira1, Felipe Hugo Mossini1, Mauricio Ursi Ventura1

[1]State University of Londrina - UEL, Brazil

INTRODUCTION:

Beans need high nitrogen (N) demand, because this element is essential for the production of nucleic acids and proteins, the main constituents of the molecules that are fundamental to all biological processes of the plant. Like other leguminous plants, the common bean has the ability to fix the atmospheric nitrogen when in symbiosis with fixing bacteria, which can contribute to the reduction of the use of nitrogen fertilizers. The objective of this study was to evaluate the efficiency of the Biological Nitrogen Fixation (BFN) by bacteria of the genus *Rhizobium* and *Azospirillum*, inoculated separately and combined, in the common bean variety IPR Tangará, identifying which inoculation this variety has the best behavior in agroecological production system.

MATERIAL AND METHODS:

The experiment was carried out on the farm of the State University of Londrina, Londrina, north of Paraná State on 23 ° 55'46" South Latitude and 50 ° 52'23 " west Longitude with an altitude of 508 meters, average annual of temperature of 20 ° C and 1588 mm of rainfall. Before the installation of the experiment, soil samples were collected at depth 0-20 cm for the chemical analysis, showing characteristics of good fertility and balanced soil, requiring no correction or addition of fertilizers. The experimental design was a randomized block with four treatments, with 1- Untreated control, without inoculation, 2-Inoculated only with *Rhizobium tropici* , 3-Inoculated only with *Azospirillum* and 4- Co-inoculation with *Rhizobium tropicium* and *Azospirillum*, using the common bean genotype, IPR TANGARÁ. There were five replicates, five blocks, being twenty experimental plots, each consisting of eigth lines of five meters(m) in length, spaced at 0.45 m, with ten plants per linear meter. The useful area was formed by the four central lines, eliminating one meter of the ends of each line. Sowing was performed using the amount of seeds sufficient to obtain a density of 10 to 12 plants per linear meter, on March 19, 2016. The emergence occurred seven days after sowing. In the full flowering stage (R6), of all plots, the roots of five plants were collected in the useful area of each plot, at depth of 0 to 20 cm. The roots were washed with running water using sieves, and then the number of nodules and the dry matter in grams were determined. Harvesting of the useful area was done 100 days after sowing (DAS) and yield was obtained with humidity corrected to 13%. The data were evaluated through analysis of variance by the F test. When the F value was significant at the 5% probability level, Tukey's test was used to compare the averages.

RESULTS AND DISCUSSION:

There was no significant difference in the number and dry mass of the nodules (Table 1), probably due to the low specificity of the plant to the symbiosis with the inoculated bacteria (HUNGRIA et al., 2013). There is also the limitation due to the high population of native rhizobia present in the soil, that may be more efficient than the ones inoculated artificially, where there is competition with the site of nodular infection, (HUNGRIA & VARGAS, 2000), since the experiment was installed in an agro-ecologically stable area with a high content of organic matter where five years

ago was adopted the organic cultivation, with rotation of cultures, and had already received experiments with bacteria fixing of nitrogen.

Table 1 - Number and dry mass of nodules, number of pods per plant and bean yield as a function of inoculation of *Rhizobium tropici* and *Azospirillum brasiliens*e, alone, and co-inoculation

Treatments	Nodules Plant^{-1}	Dry mass of Nodule	Pods Plant^{-1}	Yield
		---- g ----		---- Kg ha^{-1}---
1- Untreated control	14,720	0,043	10,958	1436,593
2-*Rhizobium tropici*	13,043	0,057	12,292	1386,963
3-*Azospirillum brasiliense*	10,333	0,063	12,190	1634,074
4-*Rhizobium* + *Azospirillum*	14,095	0,065	12,833	1361,926
Média	13,048	0,057	12,068	1454,889
C.V. (%)	22,05	1,91	11,26	45,4
Fc	2,3760	1,3290	1,9860	0,1750
Pr>Fc	0,9060	0,3109	0,1699	0,9116

Averages among treatments do not differ according to the F test at 5% significance.

Regarding the productivity, there was no significant difference with the treatments. This is mainly due to the high incidence of anthracnose disease, caused by the pathogen *Colletotrichum lindemuthianum*, at the reproductive stage. Gitti et al. (2012) did not obtain differences between inoculation and non-inoculation of the bean seeds with *Azospirillum brasilense* on the number of pods. Hungary et al. (2013) also did not verify differences between non-inoculation, nitrogen fertilization, inoculation without nitrogen fertilization, inoculation with *A. brasilense*, inoculation with *R. tropici* and co-inoculation in common bean cultivated in the rainy season of 2009/10 in Londrina .

CONCLUSION:

The inoculation of *Rhizobium tropici* and *Azospirillum brasiliense*, alone, and co-inoculation did not provide differences in number and mass of nodules and yeild in relation to the control.

REFERENCES

GITTI, D. C.; ARF, O.; KANEKO, F. H.; RODRIGUES, R. A. F.; BUZETTI, S.; PORTUGAL, J. R.; CORSINI, D. C. D. C. Inoculação de Azospirillum brasilense em cultivares de feijões cultivados no inverno. Revista Agrarian, Dourados, v. 5, n.15, p. 36-46, 2012.

HUNGRIA, M; VARGAS, M. A. T. Enviromenmental factors impacting N_2 fixation in legumes grow in the tropics, with an emphasis on Brazil. Field Crops Research, v. 65, p. 151-164, 2000.

HUNGRIA, M.; MENDES, I. C.; MERCANTE, F. M. Tecnologia de fixação biológica do nitrogênio em pequenas propriedades familiares e em propriedades tecnificadas. Londrina: Embrapa Soja, 2013 (Documentos, 338).

HUNGRIA, M.; NOGUEIRA, M. A.; ARAUJO, R. S. Co-inoculation of soybeans and common beans with rhizobia and azospirilla: strategies to improve sustainability. Biology Fertility of Soils, Firenze, v. 49, n. 7, p. 791-801, 2013.

EVALUATION OF PHYSICAL PARAMETERS OF CREOLE BEANS

Guilherme. E. P. X.; Seles, G. M.; Machado, M. A. M.; Sanglard, D. A.*; Batista, F. E. R.; Barbosa, M. H. C. and Nunes, C. F.

Biotechnology Lab, Agriculture Science Institute (ICA), Universidade Federal de Minas Gerais (UFMG), 39.404-547, Montes Claros, MG, Brazil. *E-mail: demerson.ufmg@gmail.com

Common beans plant breeding programs aim to obtain high yielding varieties, combined with disease resistance and seed production with shape, size, color and brightness acceptable by the market. In addition, bean grains must have desirable culinary and nutritional characteristics, such as ease of cooking, good palatability, soft tegument texture, and ability to produce a light and dense broth after cooking. The identification, in the creole cultivars, of the characters lost in the modern cultivars, allows the reinsertion of characters of interest in the genotypes that meet the demands of the consumer market nowadays. The objective of this study was to measure the shapes and sizes of grains (sphericity and volume), as well as their densities, of creole beans grown in the mesoregion of North of Minas Gerais, Brazil.

The measurements were performed at the Biotechnology Laboratory of the Universidade Federal de Minas Gerais, *campus* Montes Claros. It was studied tem accessions of creole beans (*Phaseolus vulgaris* L.), manually collected, with moisture content of approximately 0.92 (d. b.). The drying was carried out at the constant temperature of 40 ºC. The reduction of the moisture content during drying was accompanied by the gravimetric method (mass loss), using an analytical balance of 0.01 g. The drying was performed until the final water content reached 0.13 (d. b.). The water contents of the product were determined by the oven method, 105 ± 1 °C, until it reaches constant weight. The shape and size of the grains were analyzed by sphericity and volume, from the measurements of 300 grains of each accession, according to Moshsenin (1986). The values of the characteristic dimensions and orthogonal axes (Figure 1) were obtained with the aid of a digital caliper with an accuracy of 0.01 mm.

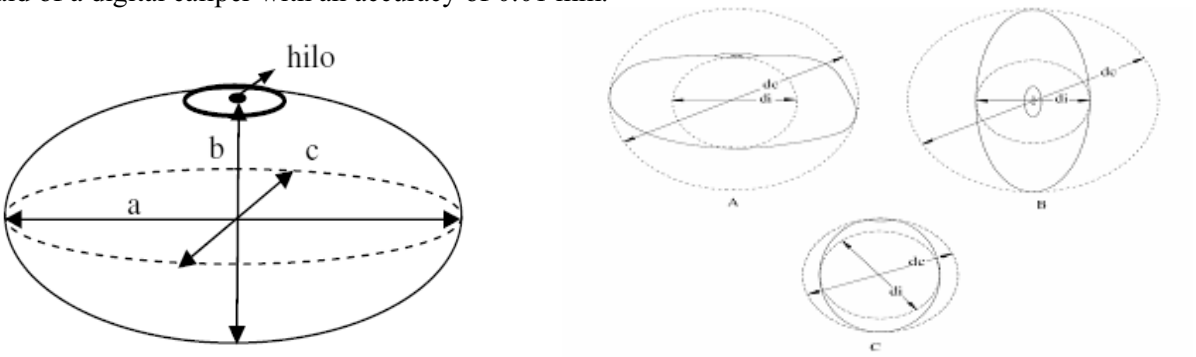


Figure 1. Schematic design of the bean grain, considering the oblate spheroid shape with its characteristic dimensions: (a) largest grain axis in mm; (B) mean grain axis in mm; (C) smaller grain axis in mm; (Cr) circularity in%; (Di) diameter of the largest circle inscribed in mm; (Dc) diameter of the smallest circumscribed circle in mm; (A) major axis, Cr_1; (B) mean axis, Cr_2; (C) smaller axis, Cr_3.

The density (g/cm^3) was determined by dividing the mass of the sample by its volume (Ferreira et al. 2002). The statistical analyses were performed in completely randomized design (CRD), where the mean (μ), the variances (σ^2) and the analysis of variance (ANOVA) were estimated by using the GENES program (Cruz, 2001).

Variance analysis of the physical properties sphericity, volume and density were all significant (Table 1). Thus, it is verified that the analyzed bean grains present variations of their characteristic dimensions (Table 2), as observed for most biological products, which, during drying, irregularly contract in different directions, as already observed by Corrêa et al. (2002). The volumetric changes of the products, due to dehydration, are reported to be the main causes of changes in the physical properties of agricultural grains (Sokhansanj & Lang, 1996). Zogzas et al. (1994) observed that the volumetric contraction of vegetable

products during drying is not due only to water content, but it also depends on the process conditions and product geometry. In this study, it can be verified that the characteristic dimensions of the grains reduce with the water content decrease. The sphericity of the grains also reduced during the drying process, while the circularity did not show a definite trend in its values with the reduction of the water content. In addition, the surface/volume ratio of the grains increased with the reduction of the water content during the drying process (Table 2). The theoretical bases for knowing the process of volumetric contraction involve complex laws of mechanical and material deformation (Towner, 1987). Although, there is an increasing tendency for genetic bean enhancement programs to intensify their researches on physical properties of the beans.

Table 1. Summary of sphericity, volume and density variance analyses in grains of creole beans accessions grown in the mesoregion of North of Minas Gerais, Brazil.

ANOVA		DF	SS	MS	F	F critic
Sphericity	**Accession**	9	33,640.4616	3,737.8290	159.0674	1.9122
	Residue	290	6,814.5415	23.4984		
	Total	299	40,455.0031			
Volume	**Accession**	9	318,877.6000	35,430.8444	59.7782	1.9123
	Residue	290	171,884.5430	592.7053		
	Total	299	490,762.1430			
Density	**Accession**	9	0.3840	0.0426	142.000	2.2106
	Residue	290	0.0945	0.0003		
	Total	299	0.4785			

ANOVA: Variance analysis; DF: Degree of Freedom; SS: Sum of Squares; MS: Mean of Squares.

Table 2. Means (μ) and variances (σ^2) of physical parameters in creole bean accessions grown in the mesoregion of the North of Minas Gerais, Brazil.

Accessions of creole beans	Measurements					
	Sphericity (%)		Volume (mm^3)		Density (g/cm^3)	
	μ	σ^2	μ	σ^2	μ	σ^2
Andu Indiano	96.2681	2.4694	154.8063	448.2148	1.2200	0.0004
Andu Manteiga	95.2533	11.1957	105.4824	374.4491	1.1899	0.0009
Andu Preto Precoce	103.0484	17.8583	295.6497	953.0872	1.0861	0.0028
Azuk Claro	113.8209	9.8515	53.2552	975.0596	1.1568	0.0011
Azuk Roxo	110.6989	25.7984	281.7536	267.0881	1.2881	0.0005
Bonina	104.3245	19.7602	134.5898	696.2885	1.2412	0.0005
Branco Mineiro	105.2441	36.1243	141.8934	138.5960	1.3508	0.0011
Campeiro Preto	601.0814	63.1603	601.0814	631.6034	1.3343	0.0001
Carioquinha Precoce	114.3910	41.2260	132.9935	604.5292	1.2843	0.0001
Catador Sisquim	65.7498	10.5395	55.2506	52.5374	1.3422	0.0110

REFERENCES

Cruz, CD. Programa Genes: aplicativo computacional em genética e estatística. Viçosa: UFV, 2001.

Ferreira, SMR et al. Controle de qualidade em sistemas de alimentação coletiva I. São Paulo: Varela; 2002.

Moshenin, NN. Physical properties of plant and animal materials. New York: Gordon and Breach Publishers, 1986. 841p.

Resende, O et al. Forma, tamanho e contraçao volumétrica do feijao (*Phaseolus vulgaris* L.) durante a secagem. Revista Brasileira de Produtos Agroindustriais, 7, 15-24, 2005.

Sokhansanj, S; Lang, W. Prediction of kernel and bulk volume of wheat and canola during adsorpsion and desorption. Journal Agricultural Engineering Research, New York, 63(2), 129-136, 1996.

Towner, GD. The tensile stress generated in clay through drying. Journal Agricultural Engineering Research, New York, 37(4), 279-289, 1987.

Zogzas, NP et al. Densities, shrinkage and porosity of some vegetables during air drying. Drying Technology, New York, 12(7), 1653-1666, 1994.

VIGOR TESTS IN THE EVALUATION OF THE PHYSIOLOGICAL QUALITY OF SEEDS OF BLACK BEAN GENOTYPES

E.V.C. Jorge[1], A.M.S. de S. David[1], L.V. de S. Cangussú[1], J.C. Figueiredo[1], R.A.Alves[1], F.H.B. Machado[1], R.A.N. Silva[1], C.D.da Silva[1], J.L.R. Barbosa[1], M.B.O. Silva[1*], A.J. de Carvalho[1], I. Aspiazú[1]

[1]Universidade Estadual de Montes Claros
*Corresponding author: mariunim@yahoo.com.br

INTRODUCTION: In Brazil, amongst the different types of common bean grains, the black one is the second most consumed. In this way, its seeds received greater attention by the breeding programs and, therefore, presented more cultivars registered at and protected by the Ministry of Agriculture, Livestock and Supply (MAPA, 2012). Thus, to ensure the recommendation of new cultivars, the genotypes should present, besides a higher productive potential, high physiological quality (germination and vigor) in the seeds produced. Thus, the objective of this work was to evaluate vigor tests on the physiological quality of seeds of common bean genotypes of the black commercial group.

MATERIAL AND METHODS: The experiment was carried out at the Laboratory of Seed Analysis, State University of Montes Claros, Campus de Janaúba-MG. The seeds used were from the planting at the Experimental Farm of the Agricultural Research Company of Minas Gerais - EPAMIG, located in the district of Mocambinho, Jaíba-MG. The trial consisted of eight elite lines and three commercial cultivars, components of the value for cultivation and use (VCU) test of common bean of the black commercial group in the 2013 autumn-winter. The experimental design was completely randomized with four replications of 50 seeds per treatment. The tests used were mass of one thousand seeds (M1000), plantlets emergence (PE), plantlets length (LS) and dry mass of plantlets (BRASIL, 2009). The data were submitted to analysis of variance and, when significant, means were compared by the Scott-Knott test at 5% significance.

RESULTS AND DISCUSSION: There was a significant effect of the genotypes for all variables studied. Seeds of the CNFP 15359, CNFP 15290, CNFP 15289, CNFP 15361, CNFP 15304, IPR Uirapuru and BRS Campeiro genotypes presented the highest mass values of one thousand seeds (Table 1). Seeds of larger size and or density have more reserves and are potentially more vigorous (GASPAR and NAKAGAWA, 2002). Only seven of the genotypes evaluated showed greater emergence of seedlings with values between 90 and 94% (Table 1). However, all genotypes evaluated presented an emergence percentage above 80%. It should be noted that the minimum percentage of germination required for the commercialization of bean seeds is 80% (BRASIL, 2005). The length of plantlets was higher for the CNFP 15304 line, while the greater accumulation of dry mass was obtained by that line and by the BRS Campeiro cultivar (Table 1). The dry mass of plantlets aims to determine the transfer of reserves to the embryo (LUDWING *et al*., 2011). Thus, genotypes that have high-vigor seeds have higher shoot, dry mass production and higher growth rates (KOLCHINSKI *et al*., 2006).

Table 1: Average values of a thousand seeds (M1000), plantlets emergence (PE), length (PL) and dry mass (DM) of black bean genotypes grown in the 2013 autumn-winter crop, Jaíba-MG.

GENOTYPES	M1000 (g)	PE (%)	PL (cm)	DM (g)
CNFP 153012	20,37 B[1]	92 A	41,64 B	6,68 B
CNFP 15359	21,66 A	85 B	39,33 C	6,54 B
BRS Esplendor	16,77 B	90 A	37,20 D	4,92 C
CNFP 15310	18,51 B	91 A	37,21 D	5,16 C
CNFP 15290	23,08 A	88 B	38,22 D	7,00 B
CNFP 15289	23,18 A	83 B	37,75 D	5,57 C
CNFP 15361	21,74 A	86 B	42,63 B	5,78 C
CNFP 15292	17,36 B	94 A	40,15 C	6,15 C
IPR Uirapuru	23,19 A	94 A	42,07 B	7,01 B
CNFP 15304	24,40 A	93 A	44,60 A	7,74 A
BRS Campeiro	24,69 A	90 A	42,18 B	7,94 A
CV (%)	9,19	5,98	3,39	12,16

[1]Media followed by the same letter in the column belong to the same group by the Scott-Knott test at 5% significance.

CONCLUSIONS: There is variation among the evaluated genotypes in relation to the mass of one thousand seeds, emergence, length and dry matter of plantlets. The CNFP 15304 line stands out among the genotypes studied, because it presents the highest physiological seed quality.

REFERENCES:

BRASIL. MINISTÉRIO DA AGRICULTURA, PECUÁRIA E ABASTECIMENTO. Regras para análise de sementes. Brasília: DNDV/CLAV, 2009. 365 p.

BRASIL. MINISTÉRIO DA AGRICULTURA, PECUÁRIA E ABASTECIMENTO. Instrução Normativa 25/2005. Ministério da Agricultura, Pecuária e Abastecimento (Anexo V - Padrões para produção e comercialização de sementes de feijão). Brasília, DF: SNAD/DNDN/CLAV: D.O.U, Brasília, DF, p. 18, Seção 1, 20/12/2005.

GASPAR, C. M.; NAKAGAWA, J. Influência do tamanho na germinação e no vigor de sementes de milheto (*Pennisetum americanum* (L.) Leeke). Revista Brasileira de Sementes, Londrina, PR, v. 24, n. 1, p. 339-344, 2002.

KOLCHINSKI, E. M.; SCHUCH L. O. B.; PESKE S. T. Crescimento inicial de soja em função do vigor das sementes. Revista Brasileira Agrociência, Pelotas, v. 12, n. 2, p. 163-166. 2006.

LUDWIG, M. P *et al*. Qualidade de sementes de soja armazenadas após recobrimento com aminoácido, polímero, fungicida e inseticida. Revista brasileira de sementes, Londrina v. 33, n. 3. 2011.

MAPA. MINISTÉRIO DA AGRICULTURA, PECUÁRIA E ABASTECIMENTO. Zoneamento agrícola de risco climático: cultivares de feijão - ano - safra 2011/2012. Brasília, 2012. Disponível em: http://sistemasweb.agricultura.gov.br/sislegis/action/detalhaAto.do?method=visualizarAtoPortalMapa&chave=1425726611. Acesso em : 08 Jan. 2018.

BENEFITS OF ROCK POWDER COATING OF COMMON BEAN SEEDS

Gilberto A Peripolli Bevilaqua[1], Paulo Eduardo Rocha Eberhardt[2], Regis Araujo Pinheiro[2], Carla Xavier Alves[2], Irajá Ferreira Antunes[1]

[1]Embrapa Clima Temperado, gilberto.bevilaqua@embrapa.br; [2]Faculdade de Agronomia Eliseu Maciel/Universidade Federal de Pelotas

INTRODUCCION

Common bean (*Phaseolus vulgaris* L.), the most cultivated legume for feeding, is one of the most important sources of protein in human's diet. Brazil is one of the main producers and consumers of the grain, which drives a very promising seed market.

Common bean seeds present problems of storability and pest attack, especially in mild climate regions and there are several treatment options to preserve their quality. The definition of methodologies for the conservation of seeds from the ecological point of view is of great importance, since Brazil occupies the second position in Latin America in terms of organically managed area, and about 70% of Brazilian organic production is located in the Southern and Southeastern States (DAROLT, 2002).

The emergency phase is crucial to achieve an adequate stand of seedlings which must present high vigor which will ensure a good crop yield potential. The objective of this work was to analyze and quantify the advantages of the bean seed coating with different rock powder types on the potential storage and vigor of the seedlings.

MATERIAL AND METHODS

Common bean seeds, cv. Expedito, from Embrapa Clima Temperado were used. The seeds were treated with following rocks powder: Basalto from São Mateus do Sul-PR, Natural phosphate from Catalão-GO and Granodiorito, from Pelotas - RS and the combination of Phosphate with Basalt. The adhesive slurry used to fix the powder to the seed was PVA glue, diluted in water, in the proportion of 70 ml of glue to 30 ml of water. The amount of adhesive syrup added to the seed was 3% in all cases. The seeds and gum were placed in a plastic bag which was inflated and hermetically sealed and stirred, then the rock powder was added and again inflated and agitated until complete fixing of the powder to the seeds. The doses of rock powder used in this work was 12% weight gain. The seeds were analyzed by the sand emergency test after 180 days of storage in brown paper packages under favorable conditions of humidity and temperature.

RESULTS AND DISCUSSION

The amount of adhesive solution added directly affects the amount of powder adhered to the seeds and the PVA glue showed good results, but other formulations can be used. Different doses of rock dust were used during the development of the work and the dose of 10% increase in seed mass was the one that presented the best response and was used here in this work.

It was observed a significant effect of the coating on the water content of the seeds and the natural phosphate was the one that provided the lowest water content, significantly higher than the control (Table 1). The reduction was of the order of 1.5 percentual point (pp) and according to Harrington (1972), the reduction of 1pp in water content, in the range of 5 to 14%, doubles the seed storage potential. With the reduction obtained of the water content of the seeds, the storage is guaranteed for a longer period favoring the conservation of the physiological and sanitary quality of the seeds.

Table 1. Seed water content, emergence, emergency speed index (ESI), dry mass of seedlings (DMS) in bean seeds treated with different types of rock dust and harvested at 21 days after sowing.

Rock powder types	Water content (%)	Emergence (%)	ESI	DMS (g)
Natural phosphate	9.0 a	51 a*	11.6 a	5.7 ns
Basalt	10.2ab	47 ab	11.3 ab	5.9
Granodiorite	11.2ab	47 ab	10.3 ab	6.1
Basalt + natural phosfhate	11.0 ab	43 b	9.7 b	5.9
Check	12.3 b	42 b	9.0 b	6.0
CV%	16	14.6	22.4	20

*Values followed by the same letter in the column do not differ by Duncan's test at the 5% level of significance.

Rock powder affected significantly the emergence of seedlings. This fact is due to the amount of time the seeds have been kept under storage, 180 days, which caused loss of vigor due to the environmental conditions of Pelotas, RS, region of mild temperature and high relative humidity. Bevilaqua et al (2015) state that the treatment effect tends to increase throughout storage. The natural phosphate was the one that presented the best results followed by basalt, granodiorite and the mixture of basalt and natural phosphate, but these did not differ from the control. As for the speed of emergence, the beneficial effect of the natural phosphate can again be observed. According to Ching (1973) the P plays an essential role in the germination and vigor of the seeds and its increase seems to positively affect these variables.

For the production of dry mass it is observed that there was no difference between the treatments. The long storage time, short evaluation period and testing season may have affected seed response. The treatment of seeds presented conditions to control the attack of pests, mainly by weevils, that were present in the control.

CONCLUSION

The seed coating with basalt, natural phosphate and granodiorite rock dust present specific effects on seed mass, water content increases seed mass, emergence and emergence velocity but no effect on dry mass production of seedlings.

REFERENCES

BEVILAQUA, G.A.P.; JOB, R.B.; EBERHARDT, P.E.R.; ALVES, C.X.; PINHEIRO, R.A. 2015. Metodologia de recobrimento de sementes de feijão com pó de rocha. Pelotas: Embrapa Clima Temperado, 20 p. (Embrapa Clima Temperado. Boletim de pesquisa e desenvolvimento, 229)

CHING, T. M.; Adenosine triphosphate content and seed vigor. Plant Physiology, Lancaster, v. 51, p. 400-402, 1973.

DAROLT, M.R. Agricultura orgânica: inventando o futuro. Londrina: IAPAR, 2002. 250 p.

HARRINGTON, J.F. Seed storage and longevity. In: KOZLOWSKI, T.T. (Ed.). Seed Biology. New York: Academic Press, 1972. v.3, p.145-245.

VAN STRAATEN, P. Farming with rocks and minerals: challenges and opportunities. Anais Acadêmia Brasileira de Ciências, Rio de Janeiro, v. 78, n. 4, 2006. http://dx.doi.org/ 10.1590/S0001-37652006000400009

ACCELERATED AGING TESTS IN SNAP BEAN

L.H.C. Almeida1; G.R. Gomes1; L.S.A. Takahashi1; E.C. Oliveira1

[1]Agronomy Department, "Universidade Estadual de Londrina" (Londrina State University) - UEL, Brazil

INTRODUCTION

Vigor tests provide additional information on the physiological potential of the seeds. Accelerated aging is one of the tests, which presents satisfactory results to determine the viability of vegetable seeds when submitted to high temperature and relative humidity. The test principle is based on the increase in the rate of deterioration due to its exposure to high temperature and relative humidity, considered the most influential environmental factors in the intensity and speed of deterioration (MARCOS FILHO, 1999). Seeds that deteriorate more slowly after being subjected to such conditions are considered more vigorous. However, little is known about the performance of bean-pod seeds submitted to the conventional test methodology, as well as its variations. The objective of this study was to evaluate the efficiency of different methodologies of the accelerated aging test to determine the physiological potential of pod bean cultivars.

MATERIAL AND METHODS

The experiment was carried out in the Laboratory of Seed Analysis of the State University of Londrina, state of Paraná, Brazil. The cultivars of selected seed bean cultivars Top Seed and Feltrin were used. The accelerated aging test was performed using the gerbox method, with 40 mL of distilled water per plastic box (gerbox), at temperatures of 41, 43 and 45 ° C for periods of 48, 72 and 96 hours, adapted from Marcos Filho (1999). After the exposure period, the seeds were submitted to the germination test (BRAZIL, 2009) with four replicates of 50 seeds per treatment. The values obtained were expressed as a percentage. The experimental design was completely randomized, with 4 replicates. The data were submitted to ANOVA by the F test and the means compared by Scott-Knott's test ($p < 0.05$).

RESULTS AND DISCUSSION

The ANOVA found significance for the number of normal seedlings of bean-pod, with the significant effect of the interaction between cultivar, temperature and period of exposure. Figure 1 showed higher germination loss when the bean seeds were submitted to a temperature of 45ºC in both cultivars, with a more pronounced reduction as the exposure period in the accelerated aging test increased. When unfolding the cultivar factor within temperature and period of exposure, Feltrin inferiority was observed in all temperatures and greater sensitivity to the increase of the exposure period that drastically reduced germination. The temperature was deployed at each cultivar level and exposure period confirmed that the increase in temperature up to 45ºC was damaging to the germinative performance, though as the longer the exposure period, the lower the quality of the snap bean seed.

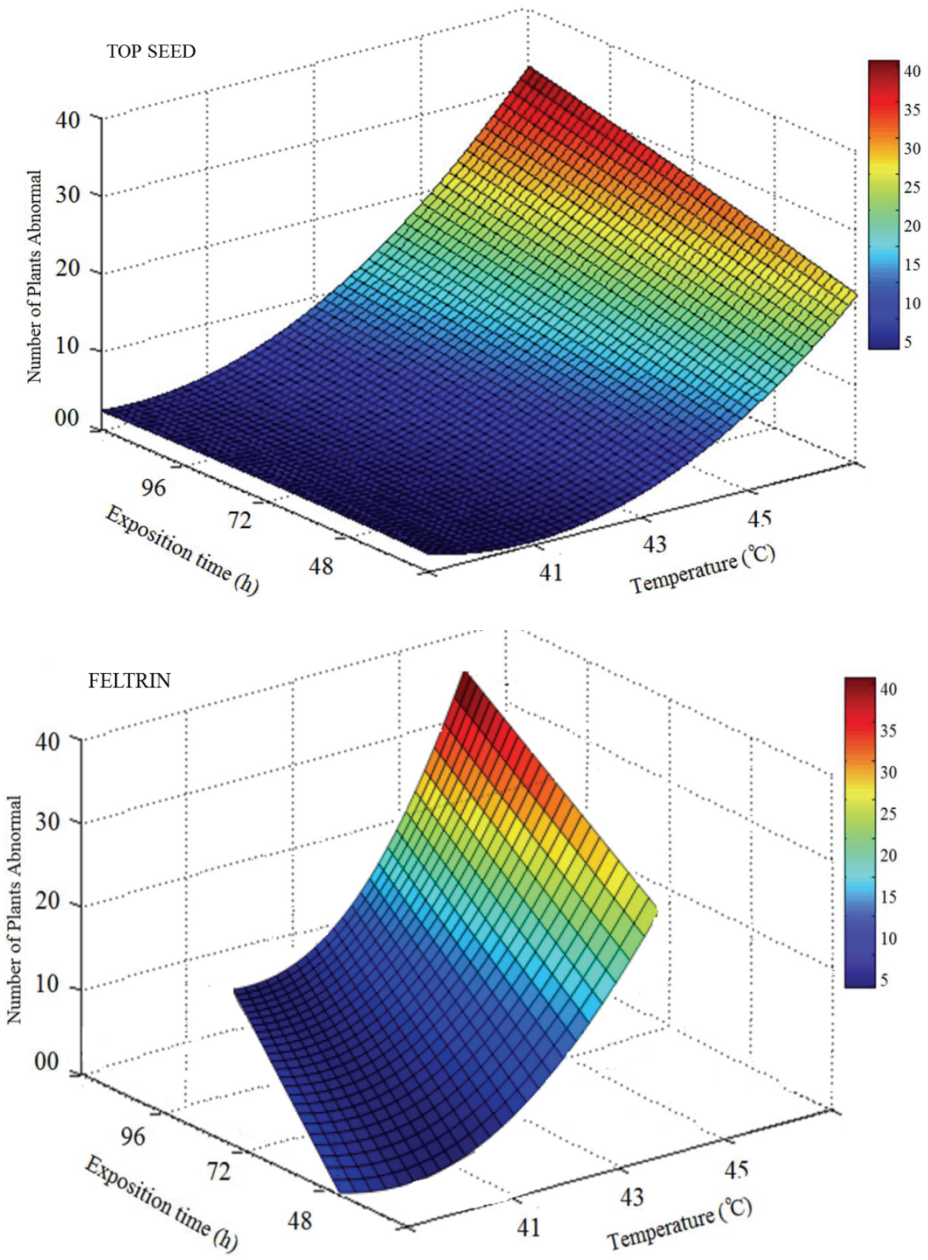


Figure 1. Number of plants abnormal in function of temperature and exposition time.

REFERENCES

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. Regras para análise de sementes. Ministério da Agricultura, Pecuária e Abastecimento. Secretaria de Defesa Agropecuária. Brasília, DF: Mapa/ ACS, 2009. 395p.

MARCOS FILHO, J. Teste de envelhecimento acelerado. In: KRZYZANOWSKI, F.C.; VIEIRA, R.D.; FRANÇA NETO, J.B. (ed.). Vigor de sementes: conceitos e testes. Londrina: ABRATES, 1999. p.3.1-3.24.

EFFECT OF FOLIAR APPLICATION OF SEAWEED EXTRACT IN COMMON BEAN YIELD

Eli C. Oliveira1, Luiz H.C. Almeida1, Claudemir Zucareli1; L.S.A. Takahashi1

[1]State University of Londrina Londrina - UEL, Brazil

INTRODUCTION

Brazil is the world's largest producer of beans (*Phaeoseolus vulgaris* L.), with three distinct seasons (Oliveira et al., 2013),low productivity can be attributed to environmental limitations, since the cultivation occurs often in regions of water restriction, prejudicing the germination process and, consequently, affecting the production. Studies have shown that the use of seaweed extracts increases the rate of germination, rooting, initial development and production of crops such as lettuce, tomato, corn and common beans (MÓGOR et al., 2008). The algae present in their composition hormones and nutrients which can be an alternative to aid in the development and final yield of the bean. Therefore, the objective of this work was to evaluate the effect of foliar applications of the *Ascophyllum nodosum* seaweed extract on the development and productivity in the bean

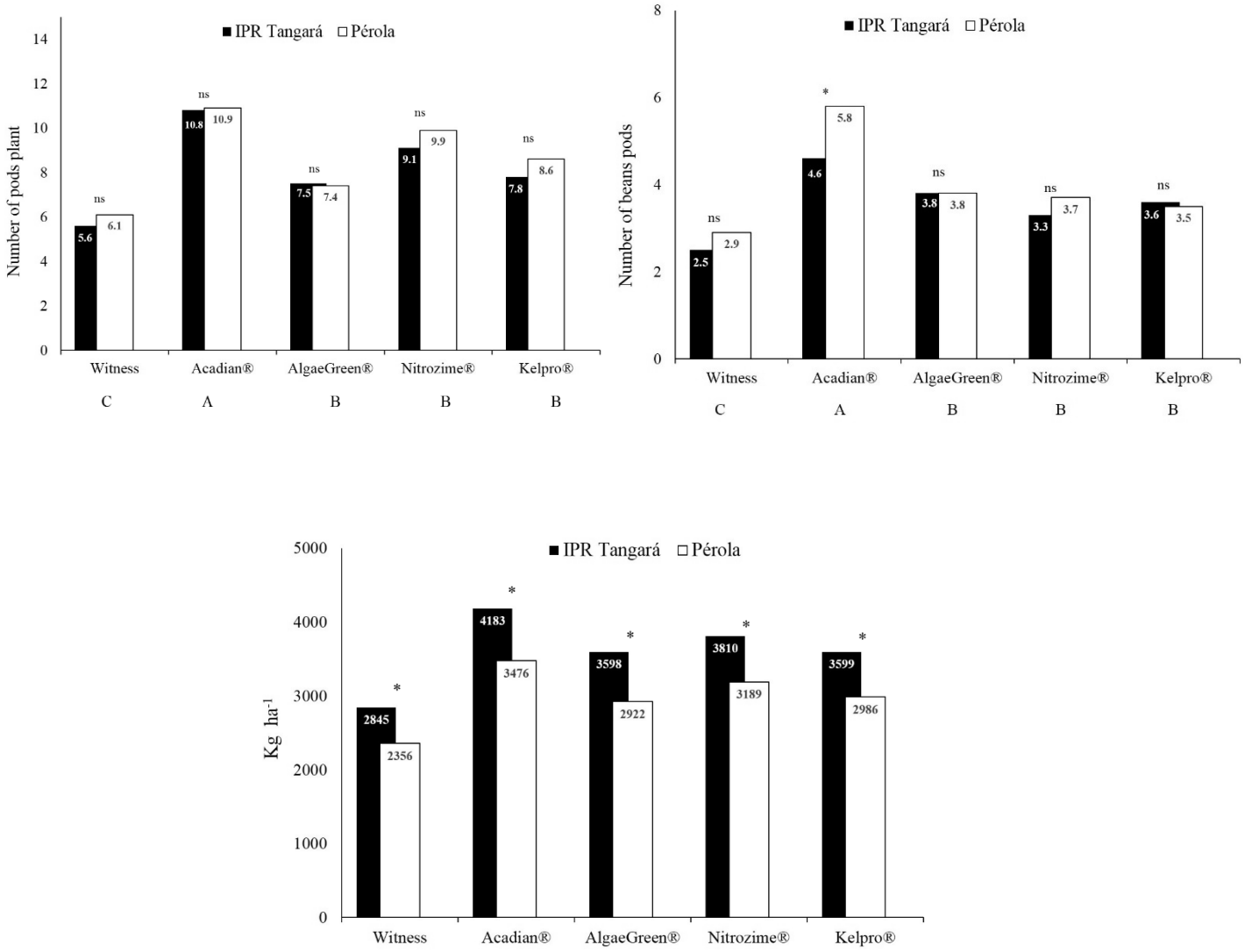
MATERIAL AND METHODS

The experiment was carried out in the experimental area ($23^0$37'42 " S and $51^0$06'15 " O and 586 m), cultivated in soil classified as clayey Cambisol hb Eutrophic (CXbe), 64% clay and 15% sand. The experimental design was used in four randomized blocks in a 2x5 factorial scheme consisting of two cultivars (IPR Tangará and Pérola) and the application of four commercial products based on seaweed extract *Ascophyllum nodosum* (Acadian®, AlgaeGreen®, Nitrozime® and Kelpro®) and a control with no application, in the vegetative stage V_3. The plots were constituted by 4 rows of 6 meters in length consisting of 40 plots. The average number of pods per plant, average number of grains per pod and productivity were measured by mass measurement, and the data were transformed in kg ha^{-1} (13% wet basis). The data were submitted to analysis of variance by the global F test to test the hypotheses of the main effects and the interaction between the factors. When the interaction effect was significant, the degrees of freedom were deployed.

RESULTS AND DISCUSSION

The variance of the experimental error of the culture environments was homogeneous ($p \leq 0.01$), allowing the accomplishment of the analysis of joint variance for all evaluated characters. In the analysis of joint variance, significant effects were observed for the two varieties and seaweed extracts, thus, there was a differentiated response between levels within each factor studied. The results of the parameters evaluated are presented in figure 1. The treatment containing the Acadian® seaweed extract presented the highest values, followed by the other treatments, all of which differed from the untreated control. The Pérola variety showed the difference in relation to the Tangará IPR, in response to the extract, thus demonstrating a greater viability of these vegetables, with the emission of a greater number of branches, as well as a greater number of sites for the formation of reproductive structures, resulting in a larger number of grains per pod. In the present study, the differences observed in the increase in the number of pods per plant and the numbers of grains per plant were efficiently related to increase grain yield due to the magnitude of the estimates of correlations obtained. The increase in the responses studied was influenced by the

foliar application of seaweed extract, however, there were differences among bean varieties in yield.



Means not followed by the same capital letter differ from each other, Tukey ($p \leq 0.01$). [ns]Not significant.

Figure 1. Mean of number of pods per plant, number of grains per pod and yield of two varieties of bean after application of seaweed extracts.

REFERENCES

OLIVEIRA, E. C.; ANDRADE, F. A.; MIGLIORANZA, E. The Three Annual Seasons of Dry Beans in Brazil. American-Eurasian Journal of Agricultural & Environmental Sciences, v. 13, p. 1004-1006, 2013.

MÓGOR, A. F.; ONO, E. O.; RODRIGUES, J. D.; MOGOR, G. Aplicação foliar de extrato de alga, ácido L-glutâmico e cálcio em feijoeiro. Scientia Agraria, v. 9, n. 4, p. 12-15, 2008.

PRODUCTIVE VALUE OF IMPROVED OPAQUE BLACK COMMON BEAN CULTIVARS IN CENTRAL VERACRUZ, MEXICO

Ernesto López-Salinas, Oscar Hugo Tosquy-Valle and Francisco Javier Ibarra-Pérez

INIFAP, Campo Experimental Cotaxtla. Km 34.5 carr. Federal Veracruz-Córdoba, Medellín de Bravo, Ver., México lopez.ernesto@inifap.gob.mx

INTRODUCTION

Cotaxtla Experimental Field Station common bean improvement Program of INIFAP, has been working since 1954 in the development of black, small and opaque common bean cultivars. To date, 13 cultivars have been developed, of which 10 have been the most commercially used, Negro Jamapa, released in 1958, standing out for its wide adaptation in favorable environments. Negro Huasteco-81 and Negro Tacaná are highly tolerant to BGYMV, Negro INIFAP is well adapted to acid soils, Negro Comapa displays wide adaptation and tolerance to angular leaf spot and Verdín, released in 2015, is well adapted to terminal drought conditions. The objective of the study was to evaluate the productive validity of improved common bean cultivars, released in the last 60 years, in comparison with a commercial control.

MATERIALS AND METHODS

A yield trial, which consisted of 10 improved black common bean cultivars and T 39 considered as a check cultivar, was carried out during three crop seasons, two trials in 2015 under irrigation during the winter-spring season; the other one was undertaken under rainfed conditions during the summer season of year 2016. Cultivars were planted at a plant density of 250,000 plants ha^{-1}, in a RCBD with three replications with experimental plots consisted of three rows 5.0 m long and 0.80 m apart. Furrow irrigation was applied six times (around 50 cm each) during the entire crop cycle for the winter-spring field trial. The bean crop under rainfed conditions received 706 mm of rain during the crop cycle. The disease incidence in all three field trials did not significantly affect seed yield, therefore grain yield (kg ha^{-1}) was the only variable statistically analyzed for each individual field trial; combined analysis of variance was also conducted to test genotype x environment interaction, and the LSD ($\alpha = 0.05$) test was used for separation of cultivar means.

RESULTS AND DISCUSSION

Statistical significance was found in each of the three environments. Seed yield of cultivar Negro Tacaná was significantly outstanding across environments; Negro Papaloapan together with Negro Tropical, Verdín and Negro Jamapa performed well in two conditions, either under irrigation or under rainfed conditions. Negro Huasteco-81 was the cultivar with the lowest field performance in all evaluation sites Table 1). According to the combined analysis of variance, Negro Tacaná was the most productive genotype (1576 kg ha^{-1}), whose yield was statistically similar to that of Negro Papaloapan and higher than the rest of the bean cultivars. Check cultivar T 39 (1288 kg ha^{-1}) and Negro Huasteco-81 (1235 kg ha^{-1}), produced the lowest average grain yields. Previous studies carried out in southeastern Mexico have shown the outstanding yield performance under irrigation, rainfed and residual soil moisture conditions of the four most outstanding cultivars here reported. Negro Tacaná has stood out its higher level of resistance to BGYMV compared to Negro Huasteco-81 (López *et al*., 1997). Negro Papaloapan has seed yield stability across the tropical environments of southeastern regions of Mexico (López *et al*., 2015); Negro Tropical is highly tolerant to fungal diseases such as rust and angular leaf spot (López *et al*., 1999) and Verdín is an early season cultivar with excellent adaptation to terminal drought (Tosquy *et al*., 2016). In turn, Negro Jamapa,

although it has good productive performance in favorable environments, has shown poor adaptation to stress conditions due to drought and acid soils, as well as susceptibility to viral and fungal diseases. It is important to continue with these field evaluations in environments with presence of biotic and abiotic stresses, in order to specify the productive validity of the cultivars under study.

Table 1. Average seed yield (kg ha^{-1}) of improved black common bean cultivars tested over two yr (2015 and 2016) under irrigation and rainfed conditions in three locations of central Veracruz, Mexico.

Cultivar	La Candelaria, Medellín (W-S 2015)	Rincón Grande, Orizaba (S 2015)	El Rubí, Medellín (W-S 2016)	Average[†]
Negro Tacaná	1503.3 *	1979.0 *	1246.7 *	1576.3 a
Negro Papaloapan	1223.3	1898.0 *	1466.7 *	1529.3 ab
Negro Tropical	1401.7 *	1344.3	1405.0 *	1383.7 bc
Verdín	1303.3 *	1566.3	1250.0 *	1373.2 bc
Negro Jamapa	1423.3 *	1847.7 *	819.0	1363.3 c
Negro Cotaxtla-91	1261.0 *	1586.0	1126.7	1324.5 c
Negro Comapa	1131.7	1538.7	1286.7 *	1319.0 c
Negro Veracruz	1423.3 *	1418.7	1078.3	1306.8 c
Negro INIFAP	1388.3 *	1389.0	1098.3	1291.9 c
T 39 (check)	957.3	1535.7	1372.7 *	1288.5 c
Negro Huasteco-81	1195.0	1393.7	1115.3	1234.7 c
ANOVA	*	**	**	*
CV (%)	11.88	12.37	14.14	12.80
LSD (0.05)	261.31	335.11	290.52	164.4

W-S = winter-spring crop season, S = summer season. [†]Cultivars with same letters are statistically similar according to LSD test (0.05).

CONCLUSIONS

Negro Tacaná and Negro Papaloapan were the improved bean cultivars with the highest productive performance across the test environments. These cultivars produced significantly higher yield than the check cultivar T 39. Negro Tropical and Verdín cultivars excelled under irrigated conditions, while Negro Jamapa produced high yields in two environments, one irrigated the other under rainfed conditions.

REFERENCES

López, S. E., et al. 1997. Crop Sci: 37(3):1022.
López, S. E., et al. 1999. Agron. Mesoam. 10(2):69-74.
López, S. E. et al. 2015. Rev. Fitotec. Mex. 38(2):173-181.
Tosquy, V. O. H., López, S. E., Villar, S. B., Acosta, G. J. A. y Rodríguez, R. J. R. 2016. Rev. Mex. Cienc. Agríc. 7(7):1775-1780.
Ugalde, A. F. J. et al. 2014. Agron. Mesoam. 25(1):35-44.

BRS INTRÉPIDO – A BLACK COMMON BEAN CULTIVAR RELEASED FOR CULTIVATION IN SOUTH BRAZIL

Irajá Ferreira Antunes[1], Gilberto A. P. Bevilaqua[1], Beatriz Marti Emygdio[1]

Embrapa Clima Temperado, Caixa Postal 403, CEP 96001-970, Pelotas, RS, Brazil[1]; e-mail: iraja.antunes@embrapa.br

Black seeded common bean (*Phaseolus vulgaris* L.) cultivars are the preferred ones by consumers in Rio Grande do Sul State, and Western Santa Catarina and Parana States. These three States form the South Region of Brazil, the greater common bean producer region of the country.

Due to importance of common bean as source of proteins and carbohydrates for the Brazilian people, a significant number of breeding programs are under development in the South Region, both public and private.

In order to permit farmers to have quick access to new released cultivars, public research institutions of the South Region designed a common bean trial where each institution is given the possibility to place two lines from own choice, besides the presence of check cultivars chosen according to research teams of each state. At each two-year cycle, a common statistical analyses at each state is performed, from which the decision on the release of the respective line as a new cultivar is under judgment by each institution. The common bean trial has been called Ensaio Sul-brasileiro de Feijão (Southern Brazil Common Bean Trial), which attends Ministry of Agriculture requirements for cultivar registration, that permits commercial use of this new cultivar.

Embrapa Clima Temperado (Temperate Climate Embrapa), a research center located in Pelotas, Rio Grande do Sul State, Brazil, is one of the public research institutions that develops a common bean breeding program. The last black bean cultivar released by this research center, was BRS Expedito, in 2003. In 2015, two new black seeded common bean cultivars have been registered at the Brazilian Ministry of Agriculture: BRS Paisano and BRS Intrépido.

The present paper describes the work that has been conducted up to the registration of BRS Intrépido as well as reveals cultivar characteristics.

BRS Intrépido results from the cross TB 97-13 x TB 98-26 that was performed in 1999. TB 97-13 is a breeding line selected at Embrapa Clima Temperado from the cross FT Tarumã x BR-Ipagro 2 - Pampa, while TB 98-26 is also a line selected at Embrapa from the cross Iapar 31 x Guapo Brilhante, both commecial cultivars. At Embrapa Clima Temperado's experimental fields, were performed the F_2 to F_4 generation advances by single pod discent. In 2004, at the F_4 generation, was selected the plant that resulted, later on, in the line identified as TB 02-02. From 2004 to 2006 crop years, TB 02-02 has shown a favorable behavior both in preliminary as in advanced State trials. Based on these results, in 2006, the line was included in the Brazilian Southern Region Common Bean Research Network, covering the Southern States of Rio Grande do Sul, Santa Catarina and Parana. From 2005/06 to 2007/08, from a total of 41 experiments scattered through the mentioned States, BRS Intrépido showed a mean seed yield of 2,370 kg ha^{-1}, 4.7% above mean yield of the standard cultivars (Table 1).

At State level, BRS Intrépido presented in Rio Grande do Sul State, 12.2 % higher yield than standard cultivars mean at the wet season and 11.1% higher in the dry season. For Santa Catarina State, 5.0 % at the wet season and -4.0% at the dry one, while in Paraná State, an equal mean yield at the wet season and 4.9% higher at the dry one. It has presented a good resistance level for antracnose and a type II plant architecture, being suitable to direct harvest.

BRS Intrépido has been registered for cultivation both at wet and dry seasons at the Ministry of Agriculture in 2015, receiving the Registration Number 33732, being suitable for cultivation in States of Rio Grande do Sul, Santa Catarina and Paraná.

Table 1. BRS Intrépido and standard cultivars mean seed yield (kg ha^{-1}) in State trials conducted in the Southern Region of Brazil, from 2005/2006 to 2007/2008.

Year	BRS Intrépido	Standard cultivars mean yield*	Relative yield (%)	Nº of trials
Rio Grande do Sul State (RS)				
Wet season – 2005/06 a 2007/08				
Mean	2544	2267	112,2	09
Dry season – 2007/08				
Mean	2239	2016	111,1	02
Santa Catarina State (SC)				
Wet season – 2006/07 e 2007/08				
Mean	3389	3238	105,0	10
Dry season - 2006/07 e 2007/08				
Mean	1446	1511	96,0	09
Paraná State (PR)				
Wet season – 2006/07 e 2007/08				
Mean	2189	2192	100,0	05
Dry season - 2006/07 e 2007/08				
Mean	2603	2481	104,9	06
Wet season – Southern Region				
General mean	2678	2523	106,1	24
Dry season – Southern Region				
General mean	2063	2003	103,0	17
All trials				
General mean	2370	2263	104,7	41 (TOTAL)

*Check cultivars: RS: 2005/06: BRS Expedito and BRS Campeiro; 2006/07 and 2007/08: Guapo Brilhante and BRS Valente; SC: 2006/07 and 2007/08: BRS Campeiro and BRS Valente; PR: 2006/07 and 2007/08: BRS Campeiro e Uirapuru

BRS PAISANO A NEW RELEASED COMMON BEAN CULTIVAR FOR SOUTHERN BRAZIL

Irajá Ferreira Antunes[1], Gilberto A. P. Bevilaqua[1]

Embrapa Clima Temperado, Caixa Postal 403, CEP 96001-970, Pelotas, RS, Brazil[1]; e-mail: iraja.antunes@embrapa.br

Common bean (*Phaseolus vulgaris* L.) breeding at Embrapa Clima Temperado (Temperate Climate Embrapa), a research center located in Pelotas, Rio Grande do Sul State, Brazil, has a long history, which began at the years sixties of the last Century, when the Ministry of Agriculture was in charge for the development of research activities with the species. At the creation of Embrapa, in 1973, research with common bean it was decided to concentrate at Goiás State, in Central Brazil Region, where was established the Centro Nacional de Pesquisa de Arroz e Feijão (National Research Center for Rice and Common Beans) – CNPAF, in 1974.

The research work that had been under way at Pelotas, was then discontinued from 1975 on. The lack of applied research with common beans from Embrapa in Rio Grande do Sul State, resulted in a demand from farmers for the return of the works with the species. The restoration of research on the theme occurred in 1986, and from then on, new cultivars have been released.

BRS Paisano is a new black bean cultivar for Southern Brazil. It results from the cross FT 206 X FT 90 10 654, conducted at Embrapa Clima Temperado, Pelotas, Rio Grande do Sul State, in 1994. Both parental lines are black seeded, and were originated at FT Sementes, a private company. At Embrapa Clima Temperado's experimental fields, were performed the F_2 to F_4 generation advances by single pod discent. In 1997, at the F_4 generation, was selected the plant that resulted, later on, in the line identified as TB 98-20. From 1999 to 2006 crop years, TB 98-20 has shown a favorable behavior both in preliminary as in advanced State trials. Based on these results, in 2006, the line was included in the Brazilian Southern Region Common Bean Research Network, covering the Southern States of Rio Grande do Sul, Santa Catarina and Parana, as well as São Paulo and Mato Grosso do Sul. From 2006/07 to 2009/10, from a total of 52 experiments scattered through the mentioned States, BRS Paisano showed a mean seed yield of 2,118 kg ha^{-1}, equal to the mean yield presented by the standard cultivars (Table 1).

At State level, presented in Rio Grande do Sul State, 2.7% higher yield than standard cultivars mean at the wet season and 13.8% higher in the dry season. For Santa Catarina State, 7.8 % at the wet season and -4.0% at the dry one, while in Paraná State, -3.1 in the wet season and -20.0% in the dry one. It has presented a good resistance level for antracnose and good performance under organic cultivation systems at farmers' fileds.

BRS Paisano has been registered for cultivation both in wet and dry seasons at the Ministry of Agriculture in 2015, receiving the Registration Number 33733, being suitable for cultivation in States of Rio Grande do Sul, Santa Catarina, Paraná, São Paulo and Mato Grosso do Sul.

Table 1. BRS Paisano and standard cultivars mean seed yield (kg ha^{-1}) in State trials conducted in the Southern Region of Brazil, from 2002/2003 to 2007/2008.

Year	BRS Paisano	Standard cultivars mean yield*	Relative yield (%)	Nº of trials
Rio Grande do Sul State (RS)				
Wet season – 2002/2003 to 2007/2008				
Mean	2.153	2.097	102,7	19
Dry season – 2007/2008				
Mean	2.243	1.971	113,8	02
Santa Catarina State (SC)				
Wet season – 2006/2007 and 2007/2008				
Mean	3.587	3.238	107,8	10
Dry season - 2006/2007 and 2007/2008				
Mean	1.477	1.539	96,0	09
Paraná State (PR)				
Wet season – 2006/2007 and 2007/2008				
Mean	2125	2192	96,9	05
Dry season - 2006/2007 and 2007/2008				
Mean	1.879	2.350	80,0	07
Southern Region				
Wet season				
General mean	2.622	2.509	104,5	34
Dry season				
General mean	1.866	1.953	95.5	18
All trials				
General mean	2.118	2.108	100,5	52 (Total)

*Standard cultivars: **RS**: 2002/2003: TPS Soberano and BRS Valente; 2003/2004: BRS Expedito and
BRS Valente; 2004/2005 and 2005/2006: BRS Expedito and BRSCampeiro;
2006/2007 and 2007/2008: Guapo Brilhante and BRS Valente; **SC**: 2006/2007 and 2007/2008: BRS
Campeiro and BRS Valente; **PR**: 2006/2007 and 2007/2008: BRS Campeiro and Uirapuru

NOTICE OF NAMING AND RELEASE OF PINTO MONARCA, A NEW HIGH GRAIN QUALITY CULTIVAR FOR THE HIGHLANDS OF MÉXICO

Rigoberto Rosales-Serna[1*] and Hilario Flores-Gallardo[1]

[1]INIFAP-CIRNOC-Campo Experimental Valle del Guadiana. Carretera Durango-El Mezquital km 4.5. Durango, México. C. P. 34170. Tel. +52 (55) 3871-8700, ext. 82714.
*rosales.rigoberto@inifap.gob.mx

The Valle del Guadiana Experiment Station of the Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP; National Research Institute in Forestry, Agriculture and Livestock) announce the naming and release of Pinto Monarca, a new pinto bean cultivar. This new bean cultivar shows an indeterminate semi-prostrate growth habit, and is considered as well adapted and high yielding cultivar for irrigated conditions in the highlands of México, where showed disease resistance, mainly to anthracnose [caused by *Colletotrichum lindemuthianum* (Sacc. & Magnus) Lambs. Scrib.] and rust [caused by *Uromyces appendiculatus* (Pers.) Unger. var. *appendiculatus*]. Pinto Monarca, tested as the improved line PT14069, was derived from the simple cross (PTSaltillo/PTColoso-2-1), made in 2010. The cross was designed to increase seed size in Pinto Saltillo cultivar used as the maternal parent in the cross.

Pinto Saltillo is a mid-season pinto bean cultivar with indeterminate growth habit (Type III) adapted to the highlands of México. Despite its popularity for commercial plantings in Durango, Chihuahua and Zacatecas, seed size in Pinto Saltillo (30-34 g/100 seeds; Sánchez *et al*., 2009) was characterized as intermediate (CIAT, 1987), limiting its acceptance for domestic and international markets where larger seeds (> 38 g/100 seeds) are preferred. Pinto Coloso is an upright indeterminate (Type III) cultivar also developed at INIFAP´s Valle del Guadiana Experiment Station and was considered as a source for higher seed size (34-48 g/100 seeds; Rosales *et al*., 2010), also showing resistance to anthracnose and rust, as well as intermediate resistance to common bacterial blight [caused by *Xanthomonas campestris* Syn. *axonopodis* pv. *phaseoli* (Smith) Dye].

In 2011, the F_1 plants were bulk advanced under field conditions in the State of Durango, located in the highlands of México. In 2012, the F_2 populations were sowed directly into the field in Durango and individual $F_{2:3}$ plants were selected on the basis of plant vigor, pod load, disease resistance and grain quality (seed size and color). During 2013, $F_{3:4}$ families were bulk advanced in the Valle del Guadiana Experiment Station and in generation F_4 individual plant selections were made based on disease reaction, earliness, plant vigor and grain commercial quality (seed size, color and shape).

In 2014, uniform population was coded as an improved line (PT14069) to be tested extensively under irrigation in the State of Durango (trials conducted at locations above an altitude of 1,800 m). Pinto Monarca was selected considering its yield response and agronomic traits at four locations from 2014 to 2017. In experimental plots under irrigated conditions Pinto Monarca registered high yields averaging 2,911 kg ha^{-1}. During 2017, under semi-commercial plots, Pinto Monarca averaged 3,159 kg ha^{-1} and outyielded, by 30 %, the local check Pinto Saltillo (2,185 kg ha^{-1}). Seed size in Pinto Monarca averages 38.8 g/100 seeds, and value registered in 2017 was 43.2 g/100 seeds compared to 40.7 g/100 seeds in Pinto Saltillo.

Pinto Monarca shows intermediate growth cycle (maturing 99 days after sowing), photoperiod sensitivity and its seed colors include brown strips over cream-colored background and slow darkening seed coat, which enhances grain shelf-life period. Producers require these types of cultivars, such as Pinto Monarca, in order to increase productivity and to improve grain quality in pinto beans produced in Northern México.

Breeder and foundation seed classes are produced and maintained by the INIFAP´s Valle del Guadiana Experiment Station. Additional information and seed samples for experimental purposes may be obtained with: Dr. Rigoberto Rosales-Serna. A. P. 186, Carretera Durango-El Mezquital km 4.5. Durango, Dgo., México. C. P. 34170. Tel. (52) 5538718700, 01 (800) 088-22-22. Extensión 82714.

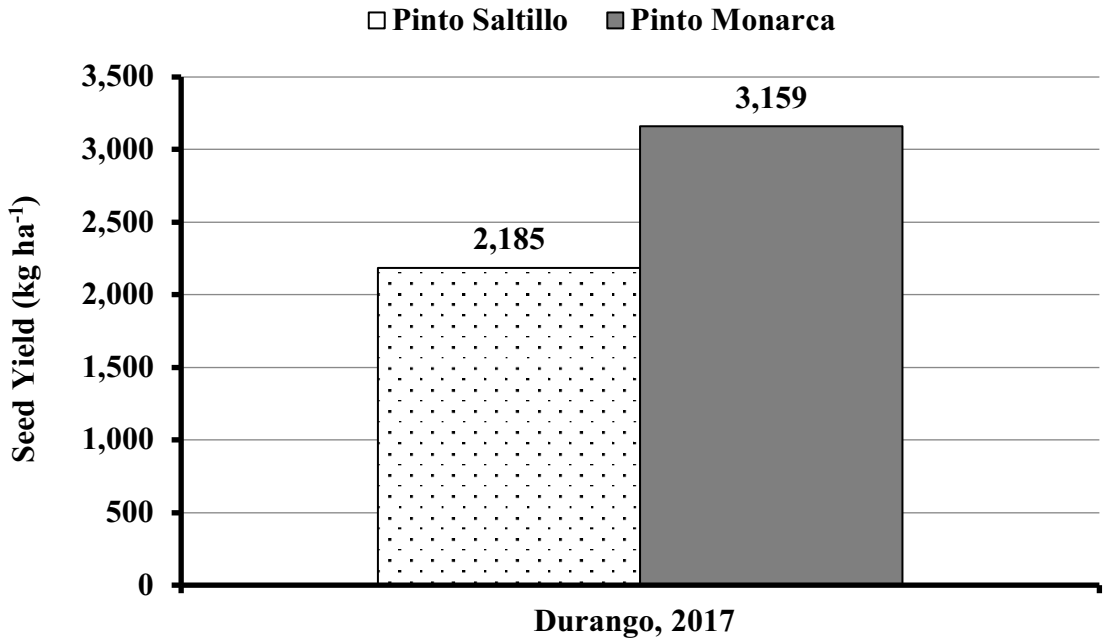


Figure 1. Seed yield observed with two pinto common bean cultivars sowed at INIFAP´s Valle del Guadiana Experiment Station. Durango, México. 2017.

REFERENCES

CIAT (Centro Internacional de Agricultura Tropical). 1987. Sistema estándar para la evaluación de germoplasma de frijol. *In*: A. van Shoonhoven y M. A. Pastor-Corrales (comps.). Cali, Colombia. 87 p.

Rosales S., R., J. A. Acosta G., F. J. Ibarra P., E. I. Cuéllar R. 2010. Pinto Coloso, nueva variedad de frijol para el estado de Durango. Revista Mexicana de Ciencias Agrícolas 1: 739-744.

Sánchez V., I., J. A. Acosta G., F. J. Ibarra P., R. Rosales S., E. I. Cuéllar R. 2009. Pinto Saltillo: variedad mejorada de frijol para el estado de Durango. Folleto Técnico Núm. 36. INIFAP-Campo Experimental Valle del Guadiana. Durango, México. 28 p.

NOTICE OF NAMING AND RELEASE OF NEGRO VENTURA, A NEW HIGH YIELDING CULTIVAR FOR THE HIGHLANDS OF MÉXICO

Rigoberto Rosales-Serna[1*] and Hilario Flores-Gallardo[1]

[1]INIFAP-CIRNOC-Campo Experimental Valle del Guadiana. Carretera Durango-El Mezquital km 4.5. Durango, México. C. P. 34170. Tel. +52 (55) 3871-8700, ext. 82714.
*rosales.rigoberto@inifap.gob.mx

The Valle del Guadiana Experiment Station of the Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP; National Research Institute in Forestry, Agriculture and Livestock) announce the naming and release of Negro Ventura, a new small opaque dry bean cultivar showing grains typical of the black bean seed class that meets the standards of domestic and international markets. This new bean cultivar shows a indeterminate semi-prostrate growth habit, and is considered as well adapted and high yielding cultivar for the irrigated conditions of the highlands of México, where showed disease resistance, mainly to anthracnose [caused by *Colletotrichum lindemuthianum* (Sacc. & Magnus) Lambs. Scrib.] and rust [caused by *Uromyces appendiculatus* (Pers.) Unger. var. *appendiculatus*]. Negro Ventura, tested as the improved line NGO14035, was derived from the simple cross (Jamapa/NGALtiplano-5), made in 2010. The cross was designed to obtain lines with rounded seeds with opaque black color, in order to increase acceptation in the Jamapa market class, cultivar used as the maternal parent in the cross.

Jamapa is a mid-season opaque black bean cultivar with indeterminate growth habit (Type II) adapted to humid tropics of México. Despite its popularity for commercial plantings in Chiapas, Veracruz and Nayarit, Jamapa is poor adapted in semi-arid highlands of México, showing susceptibility to drought and BCMV (Bean Common Mosaic Virus). Under drought conditions significant seed size reductions were observed in Jamapa seeds (>18 g/100 seeds) (CIAT, 1987), limiting its acceptance for domestic and international markets where 18 to 25 g/100 seeds are preferred. Negro Altiplano is an upright indeterminate (Type III) cultivar also developed at INIFAP´s Valle del Guadiana Experiment Station and was considered a source for higher seed size (22-27 g/100 seeds; Rosales *et al*., 2004), also showing resistance to rust, as well as intermediate resistance to anthracnose and common bacterial blight [caused by *Xanthomonas campestris* Syn. *axonopodis* pv. *phaseoli* (Smith) Dye].

In 2011, the F_1 plants were bulk advanced under field conditions at the State of Durango, located in the highlands of México. In 2012, the F_2 populations were sowed directly into the field in Durango and individual $F_{2:3}$ plants were selected on the basis of plant vigor, pod load, disease resistance and grain quality (seed size, color and shape). During 2013, $F_{3:4}$ families were bulk advanced in the Valle del Guadiana Experiment Station and in generation F_4 bulk selection was made based on disease reaction, earliness, plant vigor and grain commercial quality (seed size, color and shape).

In 2014, uniform population was coded as an improved line (NGO14035) to be tested extensively under irrigation in the State of Durango (trials conducted at locations above an altitude of 1,800 m). Negro Ventura was selected considering its yield response and agronomic traits at four locations from 2014 to 2017. In experimental plots under irrigated conditions Negro Ventura registered high yields averaging 2,960 kg ha^{-1}. During 2017, under semi-commercial plots, Negro Ventura registered 3,695 kg ha^{-1} and outyielded, by 53 %, the local check Negro San Luis (1,723

kg ha^{-1}) (Figure 1). Seed size in Negro Ventura averages 27.9 g/100 seeds, and the value registered in 2017 was 31.7 g/100 seeds, showing variations across environments between 25.7 to 31.7 g/100 seeds.

Negro Ventura shows intermediate to late growth cycle (maturing 105 days after sowing), photoperiod sensitivity, partially rounded seeds and its seed coat color is opaque black. Producers require these types of cultivars, such as Negro Ventura, in order to increase productivity and to improve grain quality of common beans produced in Northern México.

Breeder and foundation seed classes are produced and maintained by the INIFAP´s Valle del Guadiana Experiment Station. Additional information and seed samples for experimental purposes may be obtained with: Dr. Rigoberto Rosales-Serna. A. P. 186, Carretera Durango-El Mezquital km 4.5. Durango, Dgo., México. C. P. 34170. Tel. + 52 (55) 38718700, 01 (800) 088-22-22, Extensión 82714.

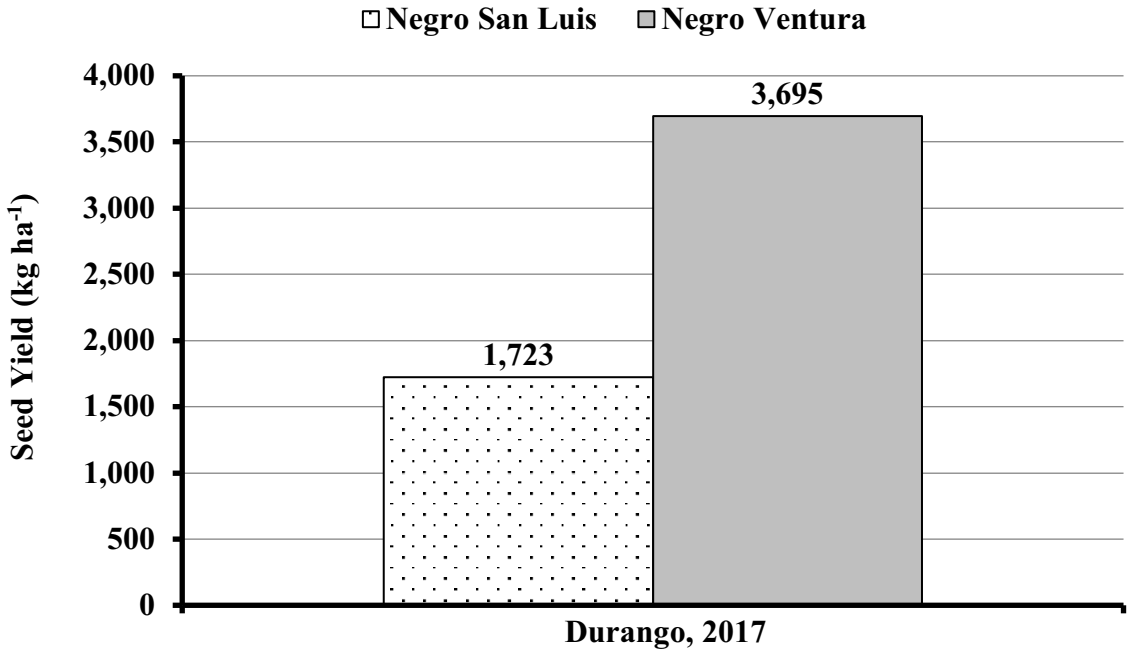


Figure 1. Seed yield observed in two pinto common bean cultivars sowed at INIFAP´s Valle del Guadiana Experiment Station. Durango, México. 2017.

REFERENCES

CIAT (Centro Internacional de Agricultura Tropical). 1987. Sistema estándar para la evaluación de germoplasma de frijol. *In*: A. van Shoonhoven y M. A. Pastor-Corrales (comps.). Cali, Colombia. 87 p.

Rosales S., R., J. A. Acosta G., J. S. Muruaga M., J. M. Hernández C., G. Esquivel E. y P. Pérez H. 2004. Variedades mejoradas de frijol del Instituto Nacional de Investigaciones Forestales Agrícolas y Pecuarias. Libro Técnico 6. Campo Experimental Valle de México. INIFAP. Chapingo, Edo. de México. 148 p.

SUBJECT MATTER INDEX

Mirka Maily Acevedo Romero
Andador Playa Roqueta # 316 Fracc.
Canelas Durango, Dgo. MEX
618-122-3389
mirkamar.itvg@hotmail.com

Anatercia Ferreira Alves
Fitotecnia- Biotecnologia e Melhoramento de Plantas (UFV)
Universidade Estadual do Maranhão-CESI
77405-090
BRAZIL (99) 982696751
anaterciaa@yahoo.com.br

Rodrigo Anschau
Univerisidade Estadual de Londrina
Londrina - Parana 86020-000
BRAZIL 55 43 99619-6411
rodrigo_anschau@hotmail.com

Iraja Ferreira Antunes
Embrapa Clima Temperado - C.
Postal 403
Pelotas, Rio Grande do Sul 96001-970
BRAZIL 53-275-8434
iraja.antunes@embrapa.br

Demerson Arruda Sanglard
ICA - UFMG
Avenida Universitaria 1.000 Bairro Universitar
BRAZIL 5.5389223121e+011
demerson.ufmg@gmail.com

Ignacio Aspiazú
Av, Reinaldo Viana 2630 - Bico da Pedra
Janaúba - MG - 39440-000
BRAZIL 55 38 38211378
ignacio.aspiazu@unimontes.br

Fabio Aurelio dias Martins
Santa Rita 153
Jardin Gloria
Lavras - MG 37200-000
fabioaureliod@gmail.com
BRAZIL

Parthiba M. Balasubramanian
Agriculture & Agri- Food Canada
Lethbridge Research Centre
Lethbridge, Alberta T1J 4B1
403-317-2275
Parthiba.Balasubramanian@AGR.GC.CA

Jim Ballerstein
NYSAES, Hedrick Hall
Geneva, NY 14456-0462
315-787-2223
jwb2@cornell.edu

Amber Bassett
1066 Bogue St., RM A364
East Lansing, MI 48824
(865)384-9657
basset31@msu.edu

Messias Jose Bastos de Andrade
Departmento de Agricultra
Universidade Federal de Lavras
Lavras-MG 35-3829-1327 BRAZIL
mandrade@ufla.br

Vanet Batista de Souza
Osvaldo Cruz St, 603
Maringa BRAZIL
55 44 997588482
vanetbatista@yahoo.com.br

James S. Beaver
Dept. of Crop and Agro- Environmental Sci
Univ. of Puerto Rico, Mayaquez
Mayaguez, PR 00681-9000
320-200-8787
j_beaver@hotmail.com;
james.beaver@upr.edu

Steve Beebe
CIAT
7343 NW 79th Terrace
Medley, FL 33166-2211
650-833-6625
s.beebe@cgiar.org

Casper Beneke
P.O. Box 14466
Kempton Park, Gauteng 14366
casper@starkeayres.co.za
SOUTH AFRICA

Erika Berghauer
Seminis Vegetable Seed
7202 PORTAGE ROAD
DEFOREST, WI 53532
(608) 842-1435
erika.mary.berghauer@monsanto.com

Kirstin Bett
Dept. of Plant Sciences
University of Saskatchewan
Saskatoon, SK S7N 5A8
306-966-4947
k.bett@usask.ca

Gilberto Bevilaqua
Embrapa CPACT
Pelotas - RS 96010-280
gilberto.bevilaqua@embrapa.br
BRAZIL

Harbans Bhardwaj
AGRIL. Research Station
BOX 9061
Petersburg, VA 23896
HBHARDWJ@VSU.EDU

Fred A. Bliss
214 Inca Pl.
Davis, CA 95616
530-756-5154
Fbliss@dcn.org

Jeffrey Boersma
U112, 3912-77th Avenue
Leduc, Alberta T9E 0B6
jeff97boersma@yahoo.com.au

Nolan Boronowski
Plant, Soil, & Microbial Sciences
Michigan State University
East Lansing, MI, 48824
nmbornowski@gmail.com

Joao Bosco dos Santos
Departmento de Biologia
UFLA, C.P. 3037
Lavras-MG BRAZIL
jbsantos@dbi.ufla.br

Mark A. Brick
Dept. of Soil & Crop Sciences
CSU, Fort Collins, CO 80524
970-491-6551
mbrick@colostate.edu

Ana Campa Negrillo
SERIDA
Apdo.13, 33300 Villaviciosa
Asturias SPAIN
acampa@serida.org

Jacqueline Campbell
2604 Stange Road, Apt 3
Ames, IA 50010
515-441-2857
jdjax@iastate.edu

Steve Cannon
1017 Crop Genome Informatics Lab
Ames, IA 50010
515-294-6971
steven.cannon@ars.usda.gov

Ivon Cerda-Hurtado
Centro de Biotecnologia Genomica-IPN
Blvd. Del Maestro esq. Elias Pina
Tamaulipa MEX

SimonChang
Syngenta Seeds, Inc.
6338 HWY 20 - 26
Nampa, ID 83687
208-465-8538
simon_jc.chang@syngenta.com

Syama Chatterton
Lethbridge Research Center AAFC
5403 1 Ave South
Lethbridge, AB T1J 4B1
403-317-2226
Syama.Chatterton@agr.gc.ca

Rowland Chirwa
Coordinator, SABRN
Chitedze Res. Stat.
Lilongwe, MALAWI
781-182-76722
r.chirwa@cgiar.org

Karen Cichy
USDA-ARS
434 Plant & Soil Sciences Bldg.
East Lansing, MI 48824-1325
517-355-0271x210
karen.cichy@ars.usda.gov

Régisde Araujo Pinheiro
Rua Uruguay 1888 apartamento 3
Pelotas - RS 96010-630
BRAZIL (053) 999592587
regispinheiroagro@gmail.com

Carlos Alessandrode Freitas
431, Dr. José Adriano Arrobas Martins Ave,
Jardim Nova Aparecida
Jaboticabal-SP, 14883-300
BRAZIL 55 16997796215
carloscaf77@gmail.com

Marcelo Muellerde Freitas
40 Vitorio st. apt 204
Jaboticabal-SP, 14883-360
BRAZIL 5.5169819559e+012
freitasmm@hotmail.com

Trazilbo Josede Paula, Jr.
EPAMIG
Vila Gianetti 47
Vicosa, MG 36570-000
BRAZIL 55-313891-2646
trazilbo@gmail.com

Antonio M. de Ron Pedreira
Mision Biologica de Galicia
El Palacio-Salcedo
36243 Pontevedra, SPAIN
34-986-854800
amderon@mbg.csic.es

Daniel G. Debouck
CIAT
7343 NW 79th Terrace
Medley, FL 33166-2211
650-833-6625
d.debouck@cgiar.org

Maria Josedel Peloso
Doutora em Genetica e Melh. de Feijao Comum
EMBRAPA Arroz e Feijao
75 375-000 Santo Antonio De Goias
BRAZIL 55-62-3533-2158
mariajose.peloso@embrapa.br

Jessica Delfini
Agronomic Institute of Paraná
Londrina, Paraná
BRAZIL 55 43 999084702
jessica_delfini@hotmail.com

Brett Despain
ADM –Edible Bean Specialties, Inc.
Caldwell, ID 83607
208-455-7728
brett.despain@adm.com

Fábio Aurélio Dias Martins
Rua Santa Rita, 153
Barrio Jardim Glória
Lavras-MG 37200-000
BRAZIL 55353829-1327
fabioaureliod@gmail.com

Josédos Santos Neto
Agronomic Institute of Paraná - IAPAR
Area of breeding and plant genetics
Londrina, Paraná BRAZIL
(51) 43 998075063/ 43 33762495
js.neto@iapar.br

Robert Duncan
Dept. of Plant Science
222 Agriculture Bldg, 66 Dafoe Rd
Winnipeg, MB R3T 2N2
202-474-6076
duncanrw@cc.umanitoba.ca

Emmalea Ernest
University of Delaware
Carvel Research & Education Center
Georgetown, DE 19947
302-856-7303
emmalea@udel.edu

J. Alberto Escalante Estrada
Colegio de Postgraduados
Campus Montecillo
Montecillo, MEX 56230
595-2-0247
jasee@colpos.mx

Sydney Everhart
Department of Plant Pathology
University of Nebraska
Lincoln, NE 68583-0722
(402) 472-2879 everhart@unl.edu

Kathryne Everts
27664 Nanticoke Rd.
Salisbury, MD 21801
410-742-8788
keverts@umd.edu

Felipe Favoretto Furlan
Universidade Estadual de Londrina
Parana CEP 86200-000
BRAZIL 55 43 999730764
fforettofurlan@gmail.com

Juan Jose Ferreira
SERIDA
Apdo.13, 33300 Villaviciosa
Asturias, SPAIN
34 985 890066
jjferreira@serida.org

Deidre Fourie
ARC-Grain Crops Institute
Private Bag X1251
Potchefstroom 2520
S. AFRICA 27-18-299-6312
FourieD@arc.agric.za

Thais Freitas Santos
Rua Anicuns
Quadra 02 Lote 08,
Iporá, Goiás
BRAZIL 64 999035179
thaisfreitassantos26@gmail.com

Valérie Geffroy
Institut de Biologie des Plantes
Université Paris Sud
91405 ORSAY cedex
FRANCE 33 1 69 15 33 65
valerie.geffroy@u-psud.fr

Robert J. Gehin
Harris Moran Seed Co.
1677 Muller Rd.
Sun Prairie, WI 53590
608-837-6574
r.gehin@hmclause.com

Dimitar Genchev
Dobroudja Agricultural Institute
9520
General Tochevo
BULGARIA 359-58-653-234
genchev@dai-gt.org;
dd_genchev@abv.bg

Paul Gepts
Dept. of Plant Sciences/MSI
1 Shields Avenue
Davis, CA 95616
530-752-7743
plgepts@ucdavis.edu

Chris Gillard
Ridgetown College
120 Main St., E.
Ridgetown, ON N0P 2C0
519-694-1632
cgillard@ridgetownc.uoguelph.ca

Humberto Godoy Androcioli
Celso Garcia Cid Road, Km 375
Londrina, PR 86001-970
BRAZIL 55 43 3376-2298
handrocioli@iapar.br

Deidrah Goldoff
Harris Moran Seed Co.
1677 Muller Road
Sun Prairie, WI 53590
608-772-9799
d.goldoff@hmclause.com

Maria Celeste Goncalves Vidigal
Av. Colombo 5790-cep:87020-900
Univ. Estadual de Maringa
Maringa, Parana, 87020-900
BRAZIL 442635036
mcgvidigal@uem.br

Pedro Vidigal Filho
Av. Colombo 5790-cep:87020-900
Univ. Estadual de Maringa
Maringa, Parana, 87020-900
BRAZIL 442635036
vidigalfilhop@gmail.com

Kenneth F. Grafton
NDSU Dept. 7500
314 Morrill Hall, 7500
Fargo, ND 58105-6050
701-231-6693
k.grafton@ndsu.edu

Tom Grebb
Central Bean Co, Inc.
P.O. Box 215
Quincy, WA 98848
509-787-1544

Michael Grusak
USDA-ARS RRVARC
1605 Albrecht Blvd N
Fargo, ND 58102
701-239-1371
mike.grusak@ars.usda.gov

John Hart
2704 Derbyshire Rd
Maitland, FL 32751
607-342-7772
jph248@cornell.edu;
johnh@earthworkseeds.com

Jerry Haynes
Jack's Bean Company LLC
402 N. Interocean Ave
Holyoke, CO 80734-1000
970-854-3702
office@jacksbean.com;
jerry@jacksbean.com

Jim Heitholt
Dept Plant Sciences - 3354
1000 E University Ave
Laramie, WY 82071
307-766-3104
jim.heitholt@uwyo.edu

Luiz Henrique Campos de Almeida
Rua Alexander Graham Bell 560
3701 Parue Jamaica
Londrina - PR 86063-250
caluizhenrique@msn.com
BRAZIL

Sanjuana Hernandez-Delgado
Instituto Politecnico Nacional
Reynosa, MEX
shernandezd@ipn.mx

Becky Higgens
Dept. of Plant Pathology
1875 No. 38th, 406 PSH
Lincoln, NE 68583-0722

Azize Homer
1115 Reynolds St.
Laramie, WY 82072
307-742-5161
ademirbas@hotmail.com

George L. Hosfield
208 Artists Alley
Blowing Rock, NC 28605-9615
828-295-6727
georgehosfield@bellsouth.net

Khwaja G. Hossain
SB 108
330 3rd Street, NE
Mayville, ND 58257
701-788-4728
k.hossain@mayvillestate.edu

Anfu Hou
Unit 100-101
Route 1Y5
Morden, Manitoba R6M 1Y5
204-822-7228
houa@agr.gc.ca

Benjamin Hughey
Pure Line Seeds, INC
P.O. Box 746
Warden, WA 98857
608-438-2554
ben.hughey@purelineseed.com

Oscar P. Hurtado
316 Lyric Lane
Silver Springs, MD 20901
ophurtado@gmail.com

Francisco Ibarra-Perez
CE Cotaxtla, INIFAP
Carretera Veracruz- Cordoba km 34.5
Verecruz MEX 94270
011 52 229 262 2233
fcojip@hotmail.com

Carmen Jacinto-Hernandez
Tepetlaoxtoc Mna-5,
Texcoco, Estado de México. CP 56253
595-4-2877
carmenjh9@yahoo.com

Venugopal Kalavacharla
205 Baker Annex
Delaware State University
Dover, DE 19901-2277
302-857-6492
vkalavacharla@desu.edu

Kelvin Kamfwa
Department of Plant Science
University of Zambia
Lusaka, ZAMBIA
2.6097360256e+011
kelvinkamfwa@gmail.com

Kris Kappenman
ADM-Seedwest
Decatur, IL 62525-1820
217-451-4707
kappenman@adm.com

Alexander Karasev
University of Idaho
Dept of PSES, AgSci Rm. 242
Moscow, ID 83844-2339
208-885-2350
akarasev@uidaho.edu

Chris Kelley
Kelley Bean Company
1520 Ave "B"
Scottsbluff, NE 69361
308-633-7333
ckelley@kelleybean.com

James D. Kelly
1066 Bogue St
Michigan State University
East Lansing, MI 48824
517-353-0169
kellyj@msu.edu

Paul Kimani
Dept of Crop Science-Kabete
University of Nairobi
Nairobi, KENYA
pmkimani@uonbi.ac.ke

Ted Kisha
Curator, Phaseolus Collection
WRPIS
Pullman, WA 99164-6402
509-335-6898
Theodore.kisha@ars.usda.gov

Ken Kmiecik
714 Seneca Pl.
Madison, WI 53711
608-698-5198
kakmiecik@sbcglobal.net

Josue Kohashi-Shibata
Centro de Botanica. Col. De Postgrad
Montecillo, Edo. De Mexico
C.P. 56230
595-95-20200
jkohashi@colpos.mx

David Kramer
DOE-Plant Research Laboratory
S220 Plant Biology Building
East Lansing, MI 48824
kramerd8@msu.edu

Paul Kusolwa
Sokoine Univeristy of Agriculture
Department of Crop Science
P.O. Box 3005, Morogoro
TANZANIA
kusolwap@gmail.com

Regina Lucia Ferreira Gomes
Rua Manoel Felicio de Carvalho
1864,Ininga
Teresina - PL 64-49-690
BRAZIL 86-3215-5754
r.lfgomes@hotmail.com

Alice MacQueen
University of Texas
Austin, TX 78759
alice.macqueen@gmail.com

Domenico Magnifico
Tera Seeds SRL Cons.
47035 Gambettola (FC)
ITALY 139-547653884
dmagnifico@teraseeds.com

Frédéric Marsolais
Southern Crop Protection & Food Res Centre
AAFC
London, ON N5V 4T3
519-953-6718
Frederic.Marsolais@agr.gc.ca

Mark Massoudi
AG BIOTECH INC.
9701 Blue Larkspur Lane
Monterey, CA 93940
831-324-0585
info@agbiotech.net

Netzahualcoyotl Mayek-Perez
Centro de Biotecnologia Genomica-IPN
Blvd. Del Maestro esq. Elias Pina
Tamaulipa, MEX
52 899-9243627
nmayek@ipn.mx

Phil McClean
Department of Plant Sciences, NDSU
Dept # 7670
PO Box 6050, 270B Loftsgard
Fargo, ND 58108-6050
701-231-8443
phil.mcclean@gmail.com

Cirano Cruz Melville
Jaboticabal, SP CEP: 14883-900
BRAZIL 55 16 98129 6298
ciranomelville@outlook.com

Phil Miklas
USDA-ARS-IAREC
24106 No. Bunn Road
Prosser, WA 99350-9687
509-786-9258
phil.miklas@ars.usda.gov

Wezi Mkwaila
Dept of Horticulture
LUANR
Lilongwe, MALAWI
265 0 998331376
wezimkwaila@gmail.com

Vania Moda-Cirino
IAPAR
Rod. Celso Garcia Cid (PR-445), Km 375
Londrina - Paraná
BRAZIL +55 (43) 3376-2123
vamoci@iapar.br

Bertrand Monsimier
Vilmorin
Route Du Manoir
bertrand.monsimier@vilmorin.com
FRANCE

James R. Myers
Dept. of Horticulture, ALS 4017
Oregon State University
Corvallis, OR 97331
541-737-3083
myersja@hort.oregonstate.edu

Susan Nchimbi-Msolla
Dept. of Crop Science and Production
Sokoine University of Agriculture
Chuo Kikuu Moragoro
TANZANIA 2.5575484997e+011
nchimbi@suanet.ac.tz; smsolla@yahoo.com

Berlin Nelson
Dept. of Plant Pathology #7660
Walster Hall 306
Fargo, ND 58105-6050
701-231-7057
berlin.nelson@ndsu.edu

Luciano Nogueira
Rodovio GO 330, km 241
Ipameri GO 75780-000
BRAZIL 55 64993211556
lucianonogueiraagro@gmail.com

Barry Ogg
Dept. of Soil & Crop Sciences
Colorado State University
Fort Collins, CO 80523-1170
970-491-6354
Barry.Ogg@colostate.edu

Atena Oladzadabbasabadi
Department of Plant Sciences, NDSU
Dept # 7670
PO Box 6050, Loftsgard
Fargo, ND 58108-6050
701-318-8192
atena.oladzad@ndsu.edu

Dâmiany Pádua Oliveira
Rua Lasmar 116
Vista Alegre
Perdoes - Minas Gerais,37260-000
damy_agro84@hotmail.com;
damiany.padua.oliveira@gmail.com
BRAZIL

Eli Carlos Oliveira
Rua Luiz Lerco, 399
Ap # 705 Torre # 01
Londrina – Paraná 86047 – 610
BRAZIL 55 43 9631 6040
elioliveira.agro@gmail.com

Marina Borges Oliveira Silva
Rua Alfonso Pena
São Gonçalo
BRAZIL Janaúba MG 39440-000
55 38 38211457
mariunim@yahoo.com.br

Arie Oppelaar
Monsanto Holland BV
Wageningse Afweg 31
6702 PD Wageningen
NETHERLANDS 31317468364
arie.oppelaar@monsanto.com

Juan M. Osorno
Dept. of Plant Science
NDSU Dept. 7670, P.O. Box 6050
Fargo, ND 58108-6050
701-231-8145
juan.osorno@ndsu.edu

Esteban S. Osuna Ceja
km 32.5
Carretera. Ags.-Zac.
Pabellon de Arteaga, Ags.
MEX 01495-65-8-01-67
osuna.salvador@inifap.gob.mx

James Palmer
Michigan Crop Improvement Assoc.
P.O. Box 21008
Lansing, MI 48909
517-332-3546
palmerj@michcrop.com

Julie S. Pasche
NDSU
Walster Hall 323
Fargo, ND 58108-6050
701-231-7077
Julie.Pasche@ndsu.edu

Talo Pastor-Corrales
Soybean Genomics and Improvement Laboratory
Bldg. 006, Room 118, BARC-West
Beltsville, MD 20705
301-504-6600
talo.pastor-corrales@ars.usda.gov

Peter Pauls
44 James St W
Guelph Ontario N1G 1E4
ppauls@uoguelph.ca

Alexis Plouy
Filer, ID 83328
alexis.plouy@monsanto.com

Tim Porch
USDA ARS SAA TARS
2200 P.A. Campos Ave., Suite 201
Mayaguez, PR 00680
787-831-3435 x254
timothy.porch@ars.usda.gov

Bodo Raatz
CIAT
7343 NW 79th Terrace
Medley, FL 33166-2211
5724450079
b.raatz@cgiar.org

Magno Antonio Patto Ramalho
Dept. de Biologia - UFLA
Cx. Pos. 3037
37200-000 Lavras, M.G
BRAZIL 035-829-1352
magnoapr@ufla.br

John Rayapati
ADM
4666 Faries PKWY 050
Decatur, IL 62526
john.rayapati@adm.com

Guiherme Renato Gomes
Rua Alexander Graham Bell 560
Londrina CEP 86069-250
BRAZIL 55 43 984257925
guilhermerenatogomes@hotmail.com

Hendrik Rietman
Storm Seeds
Heidebloemstraat 2-1
3660 Opglabbeek
BELGIUM 31-6-1996-406
hendrik@rietman.nu

Charlene Robast
Vilmorin
Route Du Manoir
FRANCE 02 4179 4179
charlene.robast@vilmorin.com

Leticia Rodrigues Sousa
Rua dos Imigrantes Q. 2 L. 10
Iporá- GO
BRAZIL (+55) 064 9.9653-2770
letyrodrigues21@gmail.com

Maria Teresa Rodriguez Gonzalez
Colegio de Postgraduados
Campus Montecillo
Montecillo MPIO. De Texcoco 56230
MEX 01-595-95-20200
mate@colpos.mx

Gonzalo Rojas-Cifuentes
Dept. of Plant Science
NDSU Dept. 7076
Fargo, ND 58108-6050
701-231-8168
Gonzalo.Rojas@ndsu.edu

Rigoberto Rosales Serna
Encinos 158
Residential Los Pinos
Durango, Dgo. Mex. 34162
MEX rigoberto_serna@yahoo.com

Juan Carlos Rosas
EAP/ZAMORANO
Tegucigalpa,
504-2287-2000 ext 2314
jcrosas@zamorano.edu

Janice M.W. Rueda
ADM
4666 Faries Parkway
Decatur, IL 62526
217-451-7722
Janice.Rueda@adm.com

Ivan A. Russkikh
Belarus State University
Department of Genetics
220030 Minsk, BELARUS
375 447193920
russkikh@bsu.by

Jeff Safe
Crites Seed Inc.
16500 Rd. 5 NW
Quincy, WA 98848
509-787-1446
jeff@critesseed.com

Gopesh C. Saha
Brotherton Seed Company, INC.
451 S. Milwaukee Ave
Moses Lake, WA 98837
509-750-2756
gopesh@brothertonseed.com

Nubia Andrea Villota Salazar
Instituto Politécnico Nacional, CBG
Reynosa Tamps, MEX
andreavillota17@yahoo.com.mx,
nvillotas1000@alumno.ipn.mx

Jim Schild
Scotts Bluff County Extension
4502 Avenue I
Scottsbluff, NE 69361
308-632-1480
Jschild1@unl.edu

Howard F. Schwartz
C205 Plant Sciences
Dept. of Bioagr. Sci. & Pest Mgmt.
Fort Collins, CO 80523-1177
970-491-6987

Luke Shellenberger
Pro Vita
PO Box 628
Kuna, ID 83634
208-463-7624
ron@provita-inc.com

Matt Shellenberger
Pro Vita
PO Box 628
Kuna, ID 83634
208-463-7624
Matt@Provita-Inc.com

Thomas H. Smith
Crop Science Building
University of Guelph
Guelph, ON, N1G 2W1
519-824-4120 ext 58339
thsmith@uoguelph.ca

Svetla Sofkova-Bobcheva
Massey University
Palmerston North
svetla.sofkova@gmail.com
NEW ZEALAND

Fabrícia Sousa Silva
Avenida Rio XII, no. 742
Iporá, Goiás
BRAZIL 64 99964-1558
fabriciasosilva@outlook.com

Elaine Souza
Departmento de Biologia
UFLA, C.P. 3037
Lavras-MG
BRAZIL 35 3829 1354
easouza@dbi.ufla.br

Maria da Conceição Martinianode Souza
Rua Capitão Rui Lucena 71
Apto 903
Recife PE 50070-080 BRAZIL
mariamartiniano@hotmail.com

Thiago Souza
Embrapa Rice and Beans
GO-462, km 12, Zona Rural
Santo Antonio de Goiás, GO
BRAZIL 55 (62) 3533-2129
thiago.souza@embrapa.br

James R. Steadman
Dept. of Plant Pathology
1875 No 38th - 406 PSH
Lincoln, NE 68583-0722
402-472-3163
jsteadman1@unl.edu

Kathy Stewart-Williams
Idaho Crop Improvement Association
2283 Wright Ave, Suite C
Twin Falls, ID 83303
208-733-2468
kathysw@idahocrop.com

John Theuws
Kempenlaan 7
B-3600 Genk, BELGIUM
32-89-85-2931
johntheuws@telenet.be

Henry J. Thompson
Colorado State University
Cancer Prevention Lab
Fort Collins, CO 80523-1173
970-491-7748
henry.thompson@colostate.edu

Alyson Thornton
Harris Moran
1677 Muller Rd.
Sun Prairie, WI 53590
608-837-6574
A.Thornton@hmclause.com

Joseph Michel Tohme
C I A T
7343 NW 79th Terrace
Medley, FL 33166-2211
415-833-6625
j.tohme@cgiar.org

Oscar H. Tosquy-Valle
CE Cotaxtla, INIFAP
Carretera Veracruz-Cordoba
Verecruz, MEX 94270
011 52 229 262 2233
tosquy.oscar@inifap.gob.mx

Jennifer Trapp
Seneca Foods Corp.
1201 N 4th St
LeSuer, MN 56058
509-521-5507
jtrapp@senecafoods.com

Shing-Jy Tsao
1184 Fynes Ct.
San Jose, CA 95131
408-332-1955
jocelyn@ntu.edu.tw

Mark A. Uebersax
2846 West Braden Road
Perry, MI 48872
517-204-2723
uebersax@tds.net

Carlos Urrea
Panhandle Research & Extension Center
4502 Avenue I
Scottsbluff, NE 69361
308-632-0556
Currea2@unl.edu

Sonia Valdez Ortega
Calle Esmeralda no. 303
C.P. 34237
Durango, Dgo., MEX
52 (618)2998168
sonia_valdez@hotmail.com

Arlene Valmadrid
East-West Seed Co., Inc.
Km 54 Cagayan Valley Road
San Rafael, Bulacan 3008
PHILIPPINES (044) 766-4952
arlene.dionglay@eastwestseed.com

Gerthonvan de Bunt
Pop Vriend Seeds B.V.
P. O. Box 5
1619 ZG Andijk
NETHERLANDS 31-22859-1462
gvandebunt@popvriendseeds.nl

Ana Vargas
Dept. of Plant Sciences
51 Campus Drive, Univ of Saskatchewan
Saskatoon, SK S7N 5A8
anavargaspal.2@gmail.com;
agv937@mail.usask.ca

Greg Varner
MI Dry Bean Res. Board
8439 N. Blair Road
Breckenridge, MI 48615-9726
989-751-8415
varnerbean@hotmail.com

Elise Vendeuvre
Vilmorin
Route Du Manoir
elise.vendeuvre@vilmorin.com
FRANCE

Rogerio Faria Vieira
Grain Legume Researcher
EPAMIG - Vila Gianetti 47
Vicosa, MG 36571-000
BRAZIL 55-31-3891-2646
rfvieira@epamig.br

Diego Viteri
Condominios Laderas del Mar 202
Aguadilla, PR 00603
dviterid@hotmail.com

Oswaldo Voysest
1225 Bushnell St
Beloit, WI 53511-6430
608-313-8606
ovoysestv@aol.com

Dan Wahlquist
Syngenta Seeds, Inc.
6338 HWY 20 - 26
Nampa, ID 83687
208-465-8510
dan.wahlquist@syngenta.com

J. G. Waines
Botany and Plant Sciences
University of California
Riverside, CA 92521-0124
951-682-3838
giles.waines@ucr.edu

Lyle Wallace
3405 NW Orchard Ave
Apt 252
Corvallis, OR 97330
847-942-2849
LW2671@gmail.com

Ivo Eduardo Wellington
Thereza Cristina de Jesus Julião, nº740
Jardim Nova Aparecida
Jaboticabal, SP CEP: 14883-296
wellington_ie@hotmail.com
BRAZIL

Molly Welsh
P.O. Box 6
Colton, WA 99113
509-330-0546
wandh@pullman.com

Jeffrey White
ALARC, USDA-ARS
21881 North Cardon Lane
Maricopa, AZ 85138
520-316-6368
jeffrey.white@ars.usda.gov

Jason Wiesinger
USDA Robert Holley Center for Agriculture and Health
Cornell University
Ithaca, NY 14853
607-255-8002
jaw456@cornell.edu

Andi Woolf
Idaho Bean Commission
821 W. State St.
Boise, ID 83702
208-334-3520
andi.woolf@bean.idaho.gov

Evan Wright
Michigan St. Univ.
4450 Beaumont Rd.
Lansing, MI 48910
517-355-2287
wrigh294@msu.edu

2017 FINANCIAL STATEMENT
BEAN IMPROVEMENT COOPERATIVE

BALANCE AS OF January 1, 2017 **$ 15,892.45**

INCOME

		2017
2017 Dues	$	2,950.00
Extra Reports	$	0.00
2017 Dues prepaid	$	0.00
Back Issues	$	240.00
BIC 2017 Meeting excess	$	pending
Bank Interest	$	119.28
TOTAL INCOME	$	3309.28

EXPENSE

Labor charges	$	515.00
Postage, Copy Charges and Office Supplies	$	235.00
Book editing fees	$	1,137.00
PayPal Fees	$	152.34
Graduate Student Presentation and Technical Merit Awards – 2017 BIC meeting	$	700.00
Wire transfer fee	$	76.00
Student travel awards – 2017 BIC meeting	$	4,500.00
TOTAL EXPENSE	$	7,315.34

BALANCE AS OF December 31, 2017 **$ 11,886.39**