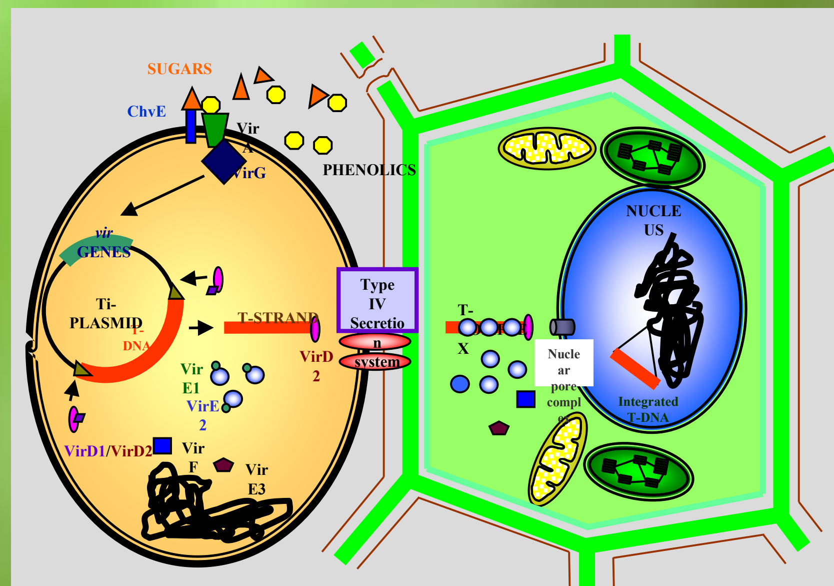
33rd Annual Mid-Atlantic Plant Molecular Biology Society

August 15, 16 2016

Soybean Monocracy

Germination 41%

PPFM Added
Germination 93%



National Wildlife Visitor Center
Patuxent Research Refuge
Laurel, MD

http://wp.towson.edu/mapmbs/

CONTENTS

You will find:	**Beginning on page**
Credits	**This page**
Welcome	**2**
Sponsors and Exhibitors	**3**
Meeting Schedule	**3-4 (Monday)** **5 (Tuesday)**
Speaker Abstracts	**7 (Monday)** **14 (Tuesday)**
Poster Titles and Abstracts	**21**
Participants	**37**

CREDITS

Lots of people provide the support and staffing for this meeting! Many thanks to all of them for the fine job they are doing.

PROGRAM
Ben Matthews
Bret Cooper
John Hammond
Siqun Wang
Richard Sicher
Hua Lu
Sue Mischke
Heven Sze
Chris Wozniak

PUBLICITY
Ben Matthews
Jim Saunders

VENDORS AND SPONSORS
Ben Matthews

PROGRAM BOOKLET & COVER DESIGN
David Puthoff

WEB PAGE
Nadim Alkharouf
Omar Darwish

REGISTRATION
Jim Saunders
Willow Saunders

LOCAL ARRANGEMENTS
Kim Campbell
Reham Youssef
Jim Saunders
Ben Matthews

AUDIO-VISUAL ASSISTANCE
Nadim Alkharouf
Chong Zhang

POSTER JUDGES
Jim Saunders
Kenneth Haymes
Bret Cooper
David Puthoff

TREASURER
Jim Saunders

WELCOME

Welcome to the 33rd annual Mid Atlantic Plant Molecular Biology meeting. Thank you for coming!!!

I am delighted so many new and returning faces are here today, and I hope this meeting is a good way to share the excitement of new scientific thinking and resources and help all of us in keeping up-to-date in the ever-broadening world of ideas, tools and advances in the plant (molecular) biological sciences. We have an outstanding group of speakers for this year's meeting, and we hope this meeting will be stimulating for all of you.

Our intention for this meeting is to provide an accessible, affordable high quality (and short) meeting in the mid-Atlantic region in a small and informal atmosphere so that scientists at all levels from undergraduate and graduate students to researchers and scientists in industry, universities and government can meet and mingle. We therefore provide lunch and breaks at the meeting so each participant has the opportunity to meet invited speakers and presenters.

Many people are involved in the planning and organizing of this meeting (see the previous page), and we thank them all for their efforts in making this another successful and productive meeting. We especially wish to thank our sponsors, and the exhibitors who furnish us with up-to-date products and services, and help to defray the cost of the meeting.

We always welcome your participation, comments and suggestions. **Also, if you are interested please join next year's organizing team and volunteer your services in planning next year's MAPMBS meeting.** This meeting was initiated 33 years ago, and several folks have participated all 33 years. Several of us are retired, and we especially hope to encourage more of you younger participants to attend the business meeting (Tuesday right before lunch) and step up and play a role in continuing this MAPMBS tradition. All are welcome at any stage of the planning and organizing process!

We thank you for your continued support and participation in the Mid Atlantic Plant Molecular Biology Society. You can keep up with MAPMBS on our website:

http://wp.towson.edu/mapmbs/

Benjamin F. Matthews
Chair

SPONSOR

Northeast Area USDA-ARS, Dr. Dariusz Swietlik, Area Director

EXHIBITORS

Cohen, Jesse	Bioline USA Inc.	(703) 217-2006 , Jesse.Cohen@bioline.com

2016 MAPMBS 33rd Annual Meeting Schedule

Monday, August 15, 2016

9:00 Registration and poster set-up

9:25 Welcome Ben Matthews, James Saunders, David Puthoff

Moderator: Stephen Mount
Dept. of Cell Biology and Molecular Genetics
University of Maryland, College Park

9:30 James Coker and Rana Kahn
The power and flexibility of on-line learning to STEM disciplines
University of Maryland University College

10:05 Wanyan Wang
Transcriptome Analysis of Resistance of Shrub Willow to *Empoasca fabae*
The Penn State University

10:30 Coffee break: Posters and Exhibitors
Presenters for posters 1-3 should make sure they get evaluated by the poster judges

11:00 Pal Maliga
Cell-to-Cell Movement of Organellar DNA in Plants
Waksman Institute of Microbiology; Department of Plant Biology, Rutgers University

11:35 Yinong Yang
Crop genome editing and precision breeding with CRISPR-Cas9
Department of Plant Pathology and Environmental Microbiology, and Huck Institutes of Life Sciences, Pennsylvania State University

12:10-1:30 MAPMBS business meeting
Lunch break: Posters and Exhibitors
Presenters for posters 4--8 should make sure they get evaluated by the poster judges

Moderator: Bret Cooper
USDA-ARS, Soybean Genomics and Improvement Lab, Beltsville, MD

1:30 N. Bhushan Mandava
BRASSINOSTEROIDS: Discovery, Isolation and Commercialization
Mandava Associates, LLC Repar Corporation

2:15 Angus Murphy
Multilevel regulation of root ABC auxin transporters in response to abiotic stress
Department of Plant Science and Landscape Architecture, University of Maryland, College Park

2:50-3:15 Coffee break: Posters and Exhibitors
Presenters for posters 9--11 should make sure they get evaluated by the poster judges

3:15 Introduction of Keynote speaker: David Puthoff
Frostburg State University, Frostburg, MD

3:20 Keynote address: Ralph Scorza
Intersections of Plant Biotechnology and Fruit Tree Breeding
USDA-ARS Appalachian Fruit Research Station

4:25 Close of day; depart the Visitor Center (building closes at 4:30)

{Speaker dinner in evening, for Invited speakers and MAPMBS program committee}

Tuesday, August 16, 2016

9:00 Registration, posters, exhibitors

9:25 Session moderator: John Hammond
USDA-ARS, United States National Arboretum, Floral and Nursery Plants Research Unit

9:30 Keynote speaker: Stanton B. Gelvin
Plant genes important for *Agrobacterium*-mediated plant genetic transformation
Department of Biological Sciences, Purdue University

10:30-11:00 Coffee break: Posters and Exhibitors
Presenters for posters 12--14 should make sure they get evaluated by the poster judges

11:00 Linda L. Kinkel
Diffuse symbioses: pathogen suppression in the soil microbiome
University of Minnesota

11:35 Mark A. Holland
Putting Symbiosis to Work: Developing Probiotics for Plants
Department of Biology, Salisbury University

12:10 – 1:45 Lunch break: Posters and Exhibitors
Presenters for posters 15 and greater should make sure they get evaluated by the poster judges

1:45 Poster competition award

Session moderator: David Puthoff
Frostburg State University, Frostburg, MD

1:50 John Hartung
Gene expression in sweet orange trees with tristeza and huanglongbing diseases
USDA-ARS, Molecular Plant Pathology Lab, Beltsville, MD

2:25 Break – coffee – posters

3:00 Jessica M Guseman
DEEPER ROOTING 1 Controls Root Orientation and Depth in Arabidopsis and Prunus Species
USDA-ARS Appalachian Fruit Research Station, Kearneysville

3:35 Burkhard Schulz
Herbicide resistance on the molecular level – what can we learn from gene expression data
University of Maryland, College Park

4:10 Close of day – posters down

The power and flexibility of on-line learning to STEM disciplines

James Coker and Rana Khan
University of Maryland University College

Online learning has its roots in distance education, which is a field that is over 170 years old. Its modern incarnation began in the 1960s and is currently being used by a vast number of institutions of higher learning, most of them public, to offer different options to millions of students and to draw a slightly different demographic to their institution. We will share some of the history and growth of online education and how UMUC, one of the pioneering public institutions in online education, is offering a broad spectrum of programs entirely online. Our focus will be on how we successfully offer several STEM programs, in particular our program in Biotechnology with four specializations. We will talk about how the curriculum is developed and delivered and the kind of activities that are part of a course. We will highlight the distinctive features of this program – professional science master's (PSM) designation, scholar-practitioner faculty, and a close working relationship with industry. We will discuss some of the recent trends in higher education/online learning and give a sneak peek of the new model of our programs at UMUC, which includes a shift in our approach to teaching and learning.

Transcriptome Analysis of Resistance of Shrub Willow to *Empoasca fabae*

Wanyan Wang, John Carlson
The Penn State University, 321 FRB, University Park, PA, 16802

Shrub Willow (*Salix* spp.), a short rotation woody biomass, has superior properties as a perennial energy crop for the Northeast and Midwest US. The potato leafhopper *Empoasca fabae* (Harris) is an insect pest that poses a serious threat to shrub willow. Currently used cultivars displayed varying susceptibility towards potato leafhopper infestation. At present, use of resistant cultivars is the optimal strategy for pest control. However, the knowledge of resistance genes is not available for breeding selection. In our study, transcriptome analysis using RNA-Seq was conducted on two shrub willow parents ('94006' and 'Jorr') in a time series (leaf tissue was collected at 0, 6, 24 and 96h after pest challenge). On average, 3.1 million paired-end reads per library were generated on the Illumina Hiseq2500 platform and mapped to the Salix *purprea* reference genome transcript sequences (*Salix purpurea* v1.0, DOE-JGI). Analysis revealed that cultivar-specific defense genes, especially in potato leafhopper induced secondary cell wall modification, play an important role in the defense mechanism in a resistant cultivar. In a highly susceptible genotype, genes involved in programed cell death were highly expressed, which explained the pest-derived symptoms like necrosis, leaf curling and early leaf drop on susceptible genotype. Overall, identifying the resistance genes from the resistance genotypes can provide genomic resources for shrub willow breeding. This is part of the project The Northeast Woody/Warm-season Biomass Consortium (NEWBio), which is funded by the USDA Agriculture and Food Research Initiative program of the National Institute of Food and Agriculture grant # 2012-68005-19703.

CELL-TO-CELL MOVEMENT OF ORGANELLAR DNA IN PLANTS

Pal Maliga
Waksman Institute of Microbiology, Rutgers University, Piscataway, NJ
Department of Plant Biology, Rutgers University, New Brunswick, NJ

To detect organelle movement between cells, we grafted two different species of tobacco, *Nicotiana tabacum* and *Nicotiana sylvestris*. We initiated tissue culture from sliced graft junctions and selected for clonal lines in which gentamycin resistance encoded in the *N. tabacum* nucleus was combined with spectinomycin resistance encoded in *N. sylvestris* plastids. We obtained evidence for cell-to-cell movement of the entire 161-kb plastid genome in these plants, most likely in intact plastids [1]. In some of the clones, mitochondrial DNA movement was also detected in regenerated plants by restoration of pollen fertility in the cytoplasmic male sterile (CMS) graft partner [2]. Homologous recombination yielded fertile and sterile mitochondrial genomes due to recombination at alternative sites, linking CMS to a unique open reading frame in CMS mitochondria. Cell-to-cell movement of plastids and mitochondria supports the universality of intercellular organelle trafficking and enables modification of the mitochondrial genome by DNA transmitted from a sexually incompatible species. A mitochondrial trait of commercial interest could be CMS in species, such as tomato, where CMS currently does not exist.

References

1 Thyssen, G., Svab, Z. & Maliga, P. Cell-to-cell movement of plastids in plants. Proc. Natl. Acad. Sci. USA 109, 2439-2443 (2012).
2 Gurdon, C., Svab, Z., Feng, Y., Kumar, D. & Maliga, P. Cell-to-cell movement of mitochondria in plants. Proc. Natl. Acad. Sci. USA 113, 3395-3400 (2016).

Crop genome editing and precision breeding with CRISPR-Cas9

Yinong Yang
Department of Plant Pathology and Environmental Microbiology, and Huck Institutes of Life Sciences, Pennsylvania State University, University Park, PA 16802

The bacterial cluster regularly interspaced short palindromic repeats (CRISPR)/CRISPR-associated protein 9 nuclease (Cas9) system has emerged as an efficient and versatile tool for genome engineering. Recently, we have demonstrated precise genome editing and targeted mutagenesis in both monocot and dicot plants with the CRISPR/Cas9 system. To increase the editing specificity and minimize potential off-target effects of CRISPR/Cas9, we performed genome-wide prediction of highly specific gRNA spacer sequences in model plants and major crops and developed bioinformatic database and web tool for gRNA design. To facilitate multiplex genome editing, a novel polycistronic tRNA-gRNA (PTG) strategy has been developed for simultaneous mutation of multiple genes, deletion of chromosomal fragments, and other more sophisticated CRISPR/Cas9 applications. The PTG strategy is effective for multiplex editing in many organisms and allows functional discovery of both nonessential and essential genes. For its application in precision breeding, different editing strategies have been employed to produce transgene-free, genetically modified crops. With improved tools and strategies for gRNA/Cas9 design, delivery and editing, CRISPR/Cas9 has become a powerful technology for genome engineering, functional genomics and precision breeding of agricultural crops.

BRASSINOSTEROIDS: Discovery, Isolation and Commercialization

N. Bhushan Mandava
Mandava Associates, LLC
Repar Corporation

Brassinosteroids are a new group of plant growth substances and are ubiquitous to both higher and lower plants. Brassinolide is the first substance in this group and was discovered by USDA scientists in Beltsville, MD in 1970. Brassinolide was isolated and its chemical structure was elucidated unequivocally as having a unique steroidal structure. Following this discovery, as many as 70 brassinosteroids were found to be present in several plants. It was soon realized that brassinolide is not an ideal candidate for commercialization because of the cost of the synthetic material. Instead, it was decided to commercialize homobrassinolide (HBR) which is an active analogue of brassinolide and is also naturally occurring. The Beltsville group has made HBR available by chemical synthesis.

In order to commercialize, HBR was approved by U.S. Environmental Protection Agency (EPA) for use on all crops. HBR was evaluated for crop yield increases and crop quality improvement of several fruit and nut crops as well as vegetable and cereal crops. HBR will be marketed on these crops in 2017 after conducting large scale field trials this year.

Multilevel regulation of root ABC auxin transporters in response to abiotic stress

Angus Murphy, Wendy Peer, Doron Shkolnik, Mark Jenness , Wiebke Tapken
Department of Plant Science and Landscape Architecture
University of Maryland, College Park MD, 20742

ATP-Binding Cassette (ABC) transporters exhibit greater diversity in plants than in other kingdoms. In animals, the B subclass of this family is notoriously associated with xenobiotic detoxification. However, in plants and fungi, plasma membrane detoxification is associated primarily with the G subclass of Pleiotropic Drug Transporters, and vacuolar sequestration of toxic metabolites involves the C subclass. Plant ABCB transporters are, instead, associated with transport of simple and aromatic organic acids, and notably function in export of the hormone auxin at the plasma membrane. In Arabidopsis and maize, rootward polar transport of auxin from sources in the shoot and leaves requires the activities of ABCB1,11/12, and 19. Proper localization and function of these transporters requires the chaperone activity of TWISTED DWARF1 / FK506 BINDING PROTEIN 42 and packing into sterol and sphingolipid-enriched membrane domains. Shootward auxin movement through the root epidermis regulates root hair and lateral root elongation and is mediated by another family member, ABCB4, which functions in conjunction with AUXIN1 (AUX1) and PINFORMED2 (PIN2). This shootward auxin transport stream is sensitive to salt stress and treatment with abscisic qcid (ABA) in wild type, *aux1* and *pin2*, but not in *abcb4*. Reductions in ABCB4 abundance at the plasma membrane (PM) are visible 10-20 minutes after ABA or salt treatment, suggesting a non-transcriptional response. PM depletion of ABCB4 involves cleavage by the APA2 saposin B - aspartic protease. Genetic, biochemical, and co-localization analyses suggest that the cleavage occurs in a *trans*-Golgi network compartment marked by SYNTAXIN OF PLANTS 61 (SYP61), which is involved in ABA and salt stress responses. In *apa2* mutants, ABCB4-GFP signals are ubiquitous, abundant, and insensitive to short-term ABA treatment. *APA2* overexpression abolished ABCB4-GFP signals and phenocopies *abcb4*. Subsequent transcriptional repression of *ABCB4* is primarily mediated by the ABA INSENSITIVE 4 transcription factor. These results clarify how stress alters root gravitropic responses by initiating endocytosis and turnover of ABCB4 in a SYP61 compartment to reduce shootward auxin transport. Further, they demonstrate that saposin B – aspartic proteases have a broader function in regulating cellular trafficking mechanisms than previously thought and warranr further investigation.

Monday's Keynote Address

Intersections of Plant Biotechnology and Fruit Tree Breeding

Ralph Scorza, Chris Dardick, Ann Callahan, Richard Bell, Zongrang Liu, Chinnathambi Srinivasan, Amy Tabb, Courtney Hollender, Jessica Guseman, Kelsey Galimba
USDA-ARS Appalachian Fruit Research Station
2217 Wiltshire Road
Kearneysville, WV, 25430

Fruit tree breeding programs are beginning to utilize plant biotechnologies for the production of novel germplasm, for cultivar development, and in the evolution of new breeding technologies. The USDA-ARS fruit breeding programs at the Appalachian Fruit Research Station have, over time, integrated critical biotechnologies that have moved fruit breeding forward in significant ways. Gene identification combined with genetic engineering (GE) have been used to develop GE virus resistant plum germplasm and led to the release of a GE *Plum pox virus* resistant plum cultivar. Information on segregation of traits developed over decades of classical breeding has been utilized for the identification and functional analysis of genes that can now be used as precise molecular markers and for directly modifying tree growth through GE. Effective transformation systems and knowledge of gene function have made possible rapid-cycle fruit breeding, whereby generation times can be reduced from 4-6 years to one year for stone fruit species. Germplasm and cultivars with novel fruit traits are being developed using GE technologies combined with conventional breeding. Further opportunities for fruit tree improvement await exploitation. There is still much work to be done and there are bottlenecks to be expanded. Efficient, productive gene transfer technologies for most tree fruits, especially using somatic tissue explants, is a major research bottleneck that once overcome will pave the way for more robust application of GE to fruit tree improvement.

Tuesday's Keynote Address

Plant genes important for *Agrobacterium*-mediated plant genetic transformation

Stanton B. Gelvin
Department of Biological Sciences, Purdue University, West Lafayette, IN 47907 USA

Agrobacterium-mediated plant genetic transformation is a core technology for basic science research and for the Agricultural Biotechnology industry. Although transgenic plants have been generated since the mid-1980's, significant problems remain for transgenesis. Among these are host range: Many agronomically important species, or particular genotypes, remain highly recalcitrant to transformation. Additional problems stem from our lack of ability to direct T-DNA integration to particular genomic sites. Random integration of T-DNA frequently results in transgene silencing. In order to address these problems, it is important to understand the plant cellular mechanisms that underlie *Agrobacterium*-mediated transformation. For the past 20 years, our laboratory has helped define host genes and proteins important for transformation, and their roles in the transformation process. In particular, we have recently begun an analysis of plant genes and DNA repair/recombination pathways important for T-DNA integration. Manipulation of plant genes may improve both the quantity and quality of transformation events.

Diffuse symbioses: pathogen suppression in the soil microbiome

Linda L. Kinkel
Department of Plant Pathology, University of Minnesota, 495 Borlaug Hall, 1991 Upper Buford Circle, St. Paul, MN 55108

Microbes and plants exist within complex networks of interacting plant and microbial species, suggesting the inadequacy of traditional frameworks for understanding plant-microbe interactions. Our work explores the interacting roles of plant community diversity, plant host, and microbial species interactions in determining the pathogen-suppressive potential and composition of soil microbiomes. Our results show that both plant host and plant community diversity play significant roles in mediating microbial competitive interactions in soil, and, consequently, in facilitating specific microbial coevolutionary trajectories. Rhizosphere *Streptomyces* associated with the same plant host were significantly more pathogen-suppressive when the host grew in monoculture vs. within a high-diversity plant community. In contrast, populations of *Streptomyces* in the rhizosphere of plant hosts growing in high-diversity communities were more niche-differentiated than populations associated with the same host in monoculture. These data suggest that plant community diversity plays a critical role in determining the likelihood of antagonistic arms race coevolution vs. niche differentiation among sympatric soil populations, with significant implications for plant disease suppression. More broadly, our work illustrates how diffuse networks of species interactions over diverse spatial scales contribute to determining the pathogen-suppressive potential of indigenous soil microbes, and suggests that crop management approaches that target species interactions offer potential for sustainable disease control.

Putting Symbiosis to Work: Developing Probiotics for Plants

Mark A. Holland
Department of Biology, Salisbury University, Salisbury, MD 21801

Our work on bacteria with probiotic effects on plants began in 1989 when we were studying urease activity in soybean and its role in nitrogen metabolism. Soybean contains genes for several different urease enzymes and a number of genes related to expression of the enzymes. Mutants for these genes were identified and characterized, but for one of the enzyme activities, no mutants could be found. As it turns out, this enzyme was not a product of the soybean plant at all, but was being produced by PPFM bacteria (Pink-pigmented, facultatively methylotrophic bacteria: *Methylobacterium spp.*) living on the plants. It was something of a surprise to us that bacteria living on a plant could have a measureable effect on plant metabolism.

We soon found that these bacteria are distributed ubiquitously in nature and normally inhabit plant leaf surfaces in numbers that range from 10^5-10^7 cfu/gram fresh weight. They are vertically transmitted in seeds. We have surveyed dozens of species of plants and have never failed to find the bacteria. Even more interesting were the effects on plant growth and development of removing the bacteria. Seeds, for example, cured of their bacteria, failed to germinate. With a reduced bacterial load, their development was stunted and abnormal. These results suggested that sometimes seeds, particularly aged seeds or poorly stored seeds, fail to germinate because their bacteria have died. Bacterization of such seeds restored germination and normal growth

Early in our studies, we showed that the bacteria produce the plant growth regulator cytokinin. Significantly, we also demonstrated that the levels of tissue-extractable cytokinin are highly correlated with the size of the bacterial population on the plant. This suggests that the cytokinin produced by the bacteria is produced in metabolically meaningful amounts. Since that time, we have also demonstrated the production of gibberellin, and have confirmed observations by Russian colleagues that the bacteria also produce auxin. There is also some evidence that they can/do produce precursors to ethylene, abscisic acid and jasmonic acid.

Evidence for the probiotic effects of these bacteria on plants suggests ways in which they might be manipulated to the benefit of agriculture. We know now that adding PPFM bacteria to growing crops can increase growth and yield.

PPFM mutants, selected for overproduction of metabolites we would like to see increased in plants can be used to deliver these metabolites and thus alter the nutritional qualities of their plant host. Using this strategy, we have increased the methionine content of soy and have added vitamin B12 to salad greens. A low-tech biotech method for metabolic engineering!

Recent work in the lab has shown that PPFMs are also associated with micro and macro algae. Several strategies for applying this finding to algae culture are under development.

Many of the applications of PPFMs to plants have received patent protection. Others are working their way through the patent pipeline. All of this technology has now been licensed by NewLeaf Symbiotics (www.newleafsym.com).

CHARACTERIZATION AND PURIFICATION OF PROTEINS OF USED FOR THE PRODUCTION OF ANTIBODIES AGAINST '*CA*. LIBERIBACTER ASIATICUS'

Huawei Liu[1], Sagheer Atta [1,2], John S. Hartung[1]
[1] USDA ARS MPPL 10300 Baltimore Avenue, Beltsville, MD 20705
[2] Ghazi University, Dera Ghazi Khan, 32200, Pakistan
John.hartung@ars.usda.gov

'Candidatus Liberibacter asiaticus' (CaLas) is considered to be the most devastating pathogen of citrus worldwide. There is no effective control method for 'CaLas' infection or huanglongbing (HLB), the disease that it causes. We have previously described both polyclonal and single chain antibodies against several 'CaLas' proteins. Detection of 'CaLas' is efficient and accurate in practice in citrus tissues with these antibodies. Isolation of a sufficient amount of purified antigen is a key step in the production of antibodies. We describe the purification of six protein antigens used to create polyclonal antibodies. These proteins include a flagellar biosynthesis protein (FlhA), a dinucleoside polyphosphate hydrolase (InvA), a portion of the major outer membrane protein (OmpA), a component of type IV pilus (CapB), the polysiallic acid capsule expression protein (KpsA) and the outer membrane efflux protein (TolC). Different isolation protocols were used for different proteins based on their respective sequences and structures. Bioinformatic analysis showed great differences between of secondary and structure three-dimensional crystal structures of the antigens. The predicted structures of the antigenic epitopes of the six antigens were clustered in various types. Our data shows that different proteins require various purification protocols according to the secondary and tertiary structures of the proteins.

DEEPER ROOTING 1 CONTROLS ROOT ORIENTATION AND DEPTH IN ARABIDOPSIS AND PRUNUS SPECIES

Jessica M Guseman, Kevin Webb, Srinivasan Chinnathambi, Chris Dardick
USDA-ARS Appalachian Fruit Research Station, Kearneysville, WV

Root system architecture (RSA) influences essential root functions such as plant anchorage in the soil, uptake of water and nutrients, and biotic interactions with soil microbes. RSA is defined as the spatial distribution of roots within the soil, which is determined by multiple parameters including root length, growth rate, branching rate, and growth orientation, or angle. While few root angle genes have been cloned, recent studies have identified *DEEPER ROOTING1 (DRO1)* as a key player in determining root growth orientation. In rice, *DRO1* was found to influence root angle, with expression of the intact gene leading to deeper root systems and increased yield under water-limited conditions. We found that *DRO1* and *DRO1*-related genes can be found across plant phyla and fall within the IGT family, named for a conserved motif. The IGT family also includes *TILLER ANGLE CONTROL1 (TAC1)* and *LAZY1*, two genes shown to control lateral shoot organ angle in both monocots and dicots. Here we demonstrate a role for *DRO1* in controlling root architecture in Arabidopsis and Prunus species. Expression of *DRO1* and related *DRO2* are largely root specific. Loss of *DRO1* led to wide lateral root growth angles, and overexpression resulted in decreased root angle in Arabidopsis. Overexpression (OE) phenotypes required a conserved C-terminal EAR-like motif. Plums overexpressing peach *DRO1* (*PpeDRO1*) exhibited longer root systems than controls. Additionally, roots of *PpeDRO1* OE plums were able to form in shoot-proliferation media in tissue culture, a phenotype not seen in control plums. Together these data demonstrate a role for *DRO1* in regulating dicot root system architecture and highlight the potential for *DRO1* expression as a tool for improving roots in an agricultural setting.

Herbicide resistance on the molecular level – what can we learn from gene expression data?

Kabelo Segobye[1], [3]Karthik Padmanabhan, [3]Michael Gribskov, [2]Stephen Weller, Burkhard Schulz[1].
[1]University of Maryland, College of Agriculture and Natural Resources, Department of Plant Science and Landscape Architecture, College Park, MD; [2]Horticulture Department, Purdue University, West Lafayette, IN; [3]Biology Department, Purdue University, West Lafayette, IN

The study of herbicide effects and herbicide resistance in plants has been focused to whole plant responses or physiological effects of herbicide treatments on plants upon treatment. Since the development of the first chemicals with selective herbicidal effects such as 2,4-D and other synthetic auxins more than 70 years ago, the effect of herbicides has been studied in the light of weed control through chemical treatments of plants. The use and often over-use of specific herbicides lead to selection of herbicide-resistant varieties of many weeds in agricultural and horticultural cropping systems. Many modes of herbicide resistance are not investigated and identified on the molecular level. The introduction of the non-selective systemic herbicide glyphosate and genetically engineered glyphosate tolerant crops increased the use of glyphosate to the most used herbicide worldwide. To date 35 mono- and dicot species have been found to have glyphosate-resistant biotypes. Different modes of glyphosate-resistance have been identified. Reduced uptake, transport, vacuolar sequestration, target gene amplification, and hypersensitive responses are ways that plants developed to resist death by glyphosate.
As an alternative to whole plant response analysis we started to analyze gene expression data as a hallmark of herbicide response in glyphosate resistant (GR) and sensitive (GS) biotypes of giant ragweed (Ambrosia trifida). Whole transcriptome data during a time course after herbicide treatment was analyzed from GR and GS plants. The data allows to address physiological pathways involved in response mechanisms in different biotypes.

Poster abstracts

#1
UNCOVERING TAC1 AND LAZY1 PROTEIN-PROTEIN INTERACTIONS USING A YEAST-TWO-HYBRID SCREEN

Emma Acly, Courtney A. Hollender, Jessica M. Guseman, Chris Dardick
USDA-ARS Appalachian Fruit Research Station, Kearneysville, WV

Shoot architecture can greatly influence both plant productivity and management. For example, peaches with a pillar phenotype allow growers to make more produce in a fixed amount of space, and trees with a weeping phenotype could make harvesting easier. Two closely related genes, *TAC1* and *LAZY1* control the angle of shoot branching. Mutations in the *TAC1* gene cause a pillar phenotype, while mutations in the closely related *LAZY1* gene lead to horizontal or weeping branches. We are interested in uncovering the molecular pathways that control branch angle. We used a yeast-two-hybrid screen to assay proteins that interact with TAC1 and LAZY1. Briefly, we used TAC1 or LAZY1 as a "bait" by fusing them to the binding domain (BD) of the yeast GAL4 transcription factor, and mated them with a library of peach proteins fused to the GAL4 activation domains (AD). When TAC1 or LAZY1 interact with a library protein, the GAL4 BD and AD will be reconstituted and can turn on a reporter gene. In this way, we can survey many peach proteins for interaction with TAC1 and LAZY1. Knowing these protein interactions will allow us to construct the molecular pathways controlling branch angle.

#2
IDENTIFICATION OF A WD-REPEAT PROTEIN THAT ACTIVATES THE DEUBIQUITINASE UBP3 AND INTERACTS WITH TWO E3 UBIQUITIN LIGASES.

Andrew Baskerville[*], Janet Donahue[‡], Glenda Gillaspy[‡], and Les Erickson[*]
[*]Department of Biological Sciences, Salisbury University, 1101 Camden Ave., Salisbury, MD
[‡]Department of Biochemistry, Virginia Polytechnic Institute and State University, 155 Otey Street NW, Blacksburg, VA
atbaskerville@salisbury.edu

Ubiquitination is an essential post-translational modification that regulates a variety of cellular processes. The ligation of the small peptide ubiquitin (Ub) onto substrate proteins by E3 Ub ligases can change protein localization, activity or stability. The most well known outcome for Ub-tagged proteins is degradation via the 26S proteasome. Deubiquitinases (DUBs) reverse the effects of ubiquitination by removing Ub from tagged substrates. In animals, Ubiquitin-specific protease (USP) 12 and USP46 are related DUBs that regulate key proteins in important growth and differentiation signaling pathways. USP12 and USP46 each form activated trimeric complexes with two WD-repeat (WDR) proteins, WDR20 and WDR48. Numerous animal and fungal DUBs are bound and activated by specific WDR proteins. In *Arabidopsis*, Ub-specific proteases 3 (AtUBP3) and 4 (AtUBP4) are related DUBs that have significant sequence similarities to animal USP12/46. Genetic studies have shown AtUBP3/4 are essential for proper pollen development and transmission. It is not yet known if WDR proteins bind and activate DUBs in plants. We have identified At2g37160 as a possible *Arabidopsis* WDR20 homolog based on amino acid sequence homology and the number and organization of WD motifs. We have shown At2g37160 (AtWDR20) interacts with both AtUBP3 and AtUBP4 in the yeast two-hybrid system. Biochemical characterization using recombinant proteins showed AtWDR20 increases the DUB activity of AtUBP3 over six-fold. A yeast two-hybrid screen of an *Arabidopsis* cDNA library using AtWDR20 as bait revealed interactions between AtWDR20 and two different E3 Ub ligases: Myb-30 Interacting E3 Ub ligase (MIEL1) and CHY Zinc-finger and Ring Protein 1 (CHYR1). Interactions between a DUB-associated WDR protein and E3 Ub ligases is intriguing, and implies AtWDR20 may function on both sides of the Ub cycle or is itself a target of ubiquitination. *In planta* and *in vitro* protein interaction analyses will further characterize the interactions between AtWDR20, AtUBP3, and these E3 ligases.

#3
IMMUNO TISSUE PRINT AND IMMUNE CAPTURE-PCR FOR DIAGNOSIS AND DETECTION OF *CANDIDATUS* LIBERIBACTER ASIATICUS

Fang Ding[1,2], Cristina Paul[1] and John S. Hartung[1]

[1] USDA ARS MPPL 10300 Baltimore Avenue, Beltsville, MD
[2] Hubei Key Laboratory of Plant Pathology, Huazhong Agricultural University, China

‘*Ca.* Liberibacter asiaticus’ (CaLas), associated with citrus Huanglongbing (HLB), is a non culturable member of the α-proteobacteria. Methods for *in situ* immuno tissue print and immuncapture followed by PCR were developed for CaLas. The anti OmpA polyclonal antibody was highly effective for the detection of CaLas from citrus petioles, stems, seeds and roots in a simple tissue printing format. The antibody was also used to capture bacteria from both citrus and periwinkle extracts for qPCR. When field samples of known CaLas-infected citrus were tested, about 80% of all samples analyzed tested positive with both immuno tissue printing and qPCR; whereas 95% were positive with at least one of these two methods. When asymptomatic citrus tissues were tested, the tissue printing method gave a higher rate of detection (83%) than the qPCR method (64%). This result is consistent with a lower concentration of CaLas DNA, but a higher proportion of viable cells, in the asymptomatic tissues. The immuno tissue printing method also preserves the detail of the spatial distribution of ‘*Ca.* Liberibacter asiaticus’ in diseased citrus tissues. Both the immuncapture PCR and immuno tissue printing methods offer the advantages of low cost, high throughput, ease of scaling for multiple samples and simplicity over current PCR-based methods for the detection of ‘*Ca.* Liberibacter asiaticus’.

#4
IDENTIFYING ROOT ANGLE GENES USING A SUPPRESSOR SCREEN IN ARABIDOPSIS

Courtney Knill, Jessica M. Guseman, Chris Dardick
USDA-ARS Appalachian Fruit Research Station, Kearneysville, WV

Root architecture is an important aspect of plant productivity since the shape of a plant's root system determines accessibility to water and nutrients. The gene *DRO1*, which was first discovered in rice, has been shown to control the orientation or angle of root growth. *DRO1* is a root-specific gene in the IGT family, which contains other genes known to control shoot architecture. In the model plant Arabidopsis thaliana, *DRO1* mutant roots have much wider growth angles than wild-type roots. Little is known about the molecular pathways surrounding *DRO1*, so our goal is to learn more about these pathways by performing a suppressor screen in Arabidopsis. We treated *dro1* mutants with the chemical mutagen EMS and screened for plants with normal root angles. We expect to find other genes that are involved in regulating root angle, and then be able to determine how they affect *DRO1*. The implications of being able to control root growth could have tremendous ramifications for orchards. One possible result would be to create modified fruit trees that can thrive in water limited conditions.

#5
IDENTIFICATION OF A HALOTOLERANT MUTANT VIA IN VITRO MUTAGENESIS IN THE CYANOBACTERIUM *FREMYELLA DIPLOSIPHON*

Tabatabai, B.[1], Arumanayagam, A.[2], and Sitther, V.[1]
[1]Department of Biology, Morgan State University, Baltimore, MD; [2]Department of Pathology, Methodist Hospital Research Institute, Houston, TX
Corresponding author's e-mail address: viji.sitther@morgan.edu

Fremyella diplosiphon is a freshwater cyanobacterium that has great potential as a biofuel agent due to its fast generation time and ability to grow in different wavelengths of light. A recent mutagenesis-based effort in *F. diplosiphon* resulted in a halotolerant mutant that tolerates up to 20 g/L NaCl. In the present study, protein upregulation and gene expression in the halotolerant mutant was evaluated. Proteomic approach using SDS-PAGE and two-dimensional polyacrylamide gel electrophoresis revealed up-regulation of five substrate-binding and associated proteins identified by matrix-assisted laser desorption/ionization time of flight mass spectrometry in the mutant. A spot corresponded to a secondary tripartite ATP-independent periplasmic (TRAP) transporter solute receptor gave a significant hit with a score of 669 with 21% sequence coverage. A three-fold increase in the expression of a TRAP transporter solute receptor encoding gene was detected using reverse transcription-quantitative PCR. Results of the study indicate that salt tolerance in *F. diplosiphon* could be enhanced by overexpression of the TRAP transporter solute receptor, thereby potentially increasing its survival in brackish waters. Further studies will be aimed towards identifying and incorporating additional halotolerance genes using biotechnological approaches to enable its growth in 35 g L^{-1} NaCl, the average salinity of seawater.

#6
The Circadian Clock Component LUX ARRHYTHMO Regulates Arabidopsis Defense Through Salicylic Acid

Chong Zhang, Nicholas C. Seitz, William Angel, Daniela Lin, Amelia Hallworth, and Hua Lu
Department of Biological Sciences, University of Maryland Baltimore County, Baltimore, Maryland 21250 USA

Recent studies showed that two morning clock genes regulate Arabidopsis defense independently of the key defense signaling mediated by salicylic acid (SA). To further understand the defense role of the circadian clock, we tested a mutant impaired in the evening clock gene *LUX ARRHYTHMO* in defense responses. We found that the *lux-1* mutant was compromised to both basal and R-gene mediated defense against *Pseudomonas syringae* and expression of the *LUX* gene was suppressed by *P. syringae*. We also found that *lux-1* had transiently reduced SA accumulation after infection with a virulent *P. syringae* strain. Consistent with this result, the double mutant *acd6-1lux-1* displayed suppression on dwarfism, cell death, and constitutive defense phenotypes, compared with *acd6-1*, which has been used as a convenient genetic tool in gauging the change of defense levels. We further found that two downstream targets of LUX also could modulate resistance to *P. syringae* via the SA pathway. Together our results showed that LUX regulates Arabidopsis defense, possibly through affecting SA signaling. These data further support crosstalk between the circadian clock and plant innate immunity and also reveal different molecular mechanisms underlying clock-defense crosstalk.

#7

VIRUS-DIRECTED PHLOEM ALTERATIONS: IMPACT ON VIRUS MOVEMENT AND DISEASE DEVELOPMENT IN STONE FRUIT CROPS

James N. Culver,[1,2] Christopher Dardick[3], Chinnathambi Srinivasan[3] and Tamara D. Collum[1,2]

[1]Dept of Plant Science and Landscape Architecture, UMD, College Park, MD
[2]Institute for Bioscience and Biotechnology Research, College Park, MD
[3]USDA-ARS, Appalachian Fruit Research Laboratory, Kearneysville, WV
jculver@umd.edu

Within the United States fruit and nut crops in the genus Prunus including peaches, plums, cherries and almonds have an annual value of over $5.4 billion. However, these crops are subject to both endemic and epidemic viruses that negatively impact both productivity and market value. The goals of this study are to utilize a translatome approach to elucidate the molecular mechanisms used by viruses to usurp the vascular tissue of stone fruit crops to facilitate movement and induce disease. This approach permits the characterization of vascular associated mRNAs in response to virus infection as well as in response to seasonal changes of growth and dormancy. Transgenic plum lines expressing a tagged ribosomal protein from either phloem specific promoters PSUC2 or PSULTR2;2, or control CaMV 35S promoter have been created and characterized. Immunopurifcation of tagged ribosomes from translatome plum lines allowed for the recovery of high quality translatome RNA. Future experiments will focus on three economically important tree fruit viruses: *Plum pox virus* (PPV), *Prunus necrotic ringspot virus* (PNRSV) and *Tomato ring-spot virus* (ToRSV). Similar *N. benthamiana* translatome lines were also created and will be infected with PPV, PNRSV and ToRSV for comparison to the studies being initiated in plum. We anticipate that findings from these studies will enhance our understanding of the transcriptional effects viruses have on the vascular tissues of tree fruit crops, the role these effects play in the development of disease, and the translational status of viral RNAs through periods of seasonal growth and dormancy.

#8
POISON IVY DRUPE MICROBIOME AND AVIAN-INDUCED FUNGAL PHYTOSANITATION OF POISON IVY DRUPES.

Deepak Poudel[1], Jessica Cahill[1], Laila Kirkpatrick[2], Regina Hanlon[1], Dana Hawley[2], David Schmale[1] and John G. Jelesko[1]
1) Dept. of Plant Pathology, Physiology, & Weed Science, 2) Biology Dept; Virginia Tech, Blacksburg, VA 24060.
poudel12@vt.edu

Microbial contamination, particularly by the phytopathogenic fungus *Colletotrichum fioriniae*, is a major obstacle during poison ivy seedling germination under lab conditions (Benhase and Jelesko, 201, HortSci, 48, 1; Kasson et. al., 2014, Plant Disease, 98, 995). Given the acid scarification requirement for seedling germination, we hypothesized that avian consumption of poison ivy drupes promotes seedling germination by digestion of endophytic fungi and bacteria residing either on the exocarp or within the mesocarp, thereby reducing fungal populations on the defecated poison ivy drupes. Poison ivy drupes were collected from the Virginia Tech Golf Course in 2014 and 2015. Microbial colony forming units were extracted from drupes and enumerated by on acidified Potato Dextrose Agar and Nutrient Agar. Drupes collected in 2014 had more bacterial CFUs than fungal CFUs. In contrast, drupes from 2015 had significantly more fungal CFUs and fewer bacterial CFUs, relative to 2014. Single spore isolated fungi were isolated and the ITS regions were sequenced. As expected, we isolated the poison ivy phytopathogen *C. fioriniae*. The increased fungal populations in 2015 drupes showed more dark colored fungal colonies, compared to 2014. The abundant dark colored fungi from 2015 drupes mostly belonged to the genera *Phoma* or *Cladosporium*. The majority of both fungi and bacteria were associated with poison ivy mesocarp tissues, indicated that they were endophytes. Thus, both fungal species composition and total numbers varied between years.
Poison ivy drupes from 2015 were fed to captive house finches to evaluate the impact of avian digestion on fungal loads in drupes. Poison ivy seeds passing through the avian gut resulted in complete removal of the exocarp, and varying degrees of mesocarp removal. Drupes that had passed through the avian gut showed significantly reduced total fungal CFUs, regardless of the degree of mesocarp removal. Thus, avian digestion of poison ivy drupes results in significant removal of total fungi, and thus potentially promotes the survival of germinating poison ivy seedlings by phytosanitizing the drupes of phytopathogenic fungi such as *C. fioriniae*.

#9
OPTIMIZING TRANSGENIC METHODS IN POISON IVY

Cody Dickinson and John Jelesko
Virginia Tech, 540 Latham Hall, Blacksburg, Virginia
chrisd1@vt.edu

Urushiol is the natural product produced by poison ivy (*Toxicodendron radicans*) that is responsible for the contact dermatitis symptoms for which this plant is widely known. One of the goals of the Jelesko Lab is to elucidate the enzymes and genes responsible for urushiol production in poison ivy. This will require the use of reverse genetic procedures in poison ivy. Currently, no reverse genetic techniques are documented in poison ivy. However mango, a related plant species also in the Anacardiaceae, has been shown to respond to transgenic gene expression using both Agrobacterium infiltration and particle bombardment procedures. Therefore, a firefly luciferase (*LUC*) construct was used to evaluate poison ivy transformation efficiency using Agrobacterium infiltration and biolistic methods.
For agrobacterium infiltrations, the target tissues were compound true leaves from various ages and growth regimes. Optimal LUC expression was observed in young leaves from plants grown in magenta boxes, whereas negligible LUC expression was observed in older leaves from potting soil grown plants. Agrobacterium infiltrated leaves also showed accumulation of colored compounds at the sites of infiltration, suggesting that the plants were mounting a defense response to the infiltrated bacteria. For this reason, biolistic transformation methods were pursued.
LUC biolistic transformation was directed at poison ivy cotyledons and first true leaves. Both tissues showed effective LUC expression, without any of the discoloration symptoms observed in the Agrobacterium transformations. However, the LUC expression varied considerably from one shot of the gene gun to another. These experiments lay the foundation for poison ivy transformation with other transgenic constructs suitable for evaluating cloned poison ivy genes predicted to be involved in urushiol biosynthesis (e.g. viral induced gene silencing constructs). The long term goal of these studies is to develop stable “poison-less ivy” plants.

#10
CELL TYPE SPECIFIC URUSHIOL CONGENER ACCUMULATION IN POISON IVY (*TOXICODENDRON RADICANS*) STEM TISSUES.

Jordan Winston[1], Drew Sturtevant[2], Minas Aziz[2], Kent Chapman[2], and John Jelesko[1]
1) Dept. of Plant Pathology, Physiology, and Weed Science, Virginia Tech, Blacksburg, VA; 2) Dept. of Biological Sciences, Univ. of North Texas, Denton, TX.
jelesko@vt.edu

Urushiol is the natural product responsible for the characteristic allergenic skin rashes caused by contact with any poison ivy (*Toxicodendron radicans*) tissue. Urushiol is not a single chemical but rather composed of congeners of pentadec(en)yl-catechol or heptadecy(en)yl-catechol with varying degrees of unsaturation on the alkyl chain. Poison ivy produces mostly pentadec(en)yl-catechol (C15-urushiol), and substantially less heptadec(en)yl-catechol (C17-urushiol) congeners. Moreover, the degree of unsaturation of the C15- and C17-urushiols is often different, suggesting that they may be produced in different cell types.
To investigate this hypothesis, we subjected poison ivy stem cross sections to 2-Dimentional MALDI-MS (2D-MALDI-MS) analysis using a high resolution Obitrap-XL mass spectrometer. Validation of urushiols by MALDI-MS detection was performed on a chloroform extract of the sap from the Japanese lacquer tree (*Toxicodendron vernicifluum*), which is previously characterized as having a urushiol congener composition very similar to poison ivy. When imaging poison ivy young seedling hypocotyls and internode cross sections, some urushiol congener adducts ionized better than others. Nevertheless, 2D-MALDI-MS imaging demonstrated that C15-urushiols were mostly confined to apparent resin duct tissues, whereas the C17-urushiols accumulated in cortex and vascular bundle tissues. Cross sections from the same plants were also subjected to GC-MS. The latter confirmed the presence of urushiol congeners identified in 2D-MALDI-MS, as well as greater C15-urushiol levels relative to C17 urushiols. Based upon these localization patterns, C15-urushiols are likely synthesized at high levels in resin ducts, whereas C17-urushiols are likely synthesized at much lower levels in the cortex and vascular bundles in poison ivy stems.

#11
Molecular analysis of glyphosate resistance in giant ragweed (Ambrosia trifida)

Kabelo Segobye[1] and Burkhard Schulz[1]
[1]University of Maryland, College of Agriculture and Natural Resources, Department of Plant Science and Landscape Architecture, College Park, MD

Giant ragweed *(Ambrosia trifida)* is a competitive, annual weed. The introduction of glyphosate resistant agronomic crops ("Roundup®-ready") in 1996 provided an effective tool to manage giant ragweed. The physiological mechanism of glyphosate's herbicide effect is inhibition of EPSPS, a key enzyme in the shikimate pathway. The use of glyphosate drastically increased after 1996 in glyphosate resistant crops. This resulted in tremendous selection pressure for glyphosate resistant giant ragweed (GRGR). We are investigating the mechanism(s) of glyphosate resistance in GRGR biotypes. The goal of our project is to discover glyphosate resistance genes. We hypothesize that the basis of resistance in GRGR biotypes is related to reduced translocation of glyphosate and a rapid response of glyphosate treated leaves in GRGR, which show a hypersensitive-like (HR) reaction to herbicide treatments. This HR results in leaf abscission within a day of treatment. GRGR plants do not die from glyphosate treatments but re-grow from shoot and axillary meristems and reproduce. Glyphosate sensitive plants transport the herbicide throughout the entire plant, which leads to leaf chlorosis and eventual death of the treated plants after 2-3 weeks.

The progression of the response and symptoms in GRGR when treated with glyphosate resemble a typical hypersensitive response similar to that observed on some plants after pathogen attack. To assess the reaction of sensitive and resistant biotypes to glyphosate on the molecular level we analyzed the total transcriptome of treated and untreated plants after glyphosate application. We have identified a list of genes that were differentially expressed between the two biotypes as the first step in identifying genes responsible for the glyphosate resistance observed. We study the pathogen response pathway in plants and assess the role of defense hormone salicylic acid (SA) as a basic immune signal and how it is involved in controlling plant sensitivity to glyphosate treatment.

#12
DEVELOPMENTAL MECHANISMS UNDERLYING FLESHY FRUIT DIVERSITY IN ROSACEAE

Authors: Kelsey Galimba[1,2], Ann Callahan[1], Chris Dardick[1], Zhongchi Liu[2]
[1]USDA-ARS Appalachian Fruit Research Station, 2217 Wiltshire Road, Kearneysville, WV
[2] University of Maryland, College Park, MD
kelsey.galimba@ars.usda.gov

The family Rosaceae includes a number of economically-important fruit and ornamental crop species and exhibits an impressive diversity of fleshy fruit morphologies, which evolved independently from a dry fruit ancestor. Here, we compare the development of two types of these fleshy fruits: stone fruits and pomes. In stone fruits such as peach, the flesh arises from the mesocarp of the ovary wall, while in pome fruits such as apple, the ovary wall forms the core and the flesh is derived from the hypanthium. We are interested in determining what specific genes and regulatory gene networks have evolved to initiate and specify fleshy tissue development in the different floral parts of each fruit type. Although the majority of studies in fruit development focus on later events like ripening, there is a growing list of genes determined to be involved in fruit flesh initiation. We focus on describing the changing morphology of apple and peach fruits, from pre-pollination to embryo maturity. We also analyze the expression patterns of a number of MADS-box fruit development transcription factors, comparing the hypanthium (fleshy in apple and non-fleshy in peach), to the ovary (non-fleshy in apple and fleshy in peach). Determining the genetic control of fruit flesh initiation will not only give insight into how this important developmental process takes place, but may also potentially be applied to secure fruit production as environmental factors threaten agriculture across the globe.

#13

THE ARABIDOPSIS SPLICING FACTOR GENE *SR45* AND CRYPTIC GENETIC VARIATION FOR SIZE

Sonal Patel, Heba Ibrahim and Stephen M. Mount
Center for Bioinformatics and Computational Biology
Dept. of Cell Biology and Molecular Genetics
University of Maryland College Park, MD 20742
smount@umd.edu

Loss-of-function mutations in the *Arabidopsis thaliana* gene *SR45*, which encodes a conserved SR protein splicing factor, have surprisingly mild phenotypes. However, much more severe phenotypes are sometimes observed in crosses between *sr45-1* in the Columbia background and other strains of Arabidopsis. In particular, extremely small plants have been observed among the progeny of crosses between *sr45-1* (a TDNA insertion in the Col background) and recombinant inbred lines (RILs) derived from Col and Ler. Because *sr45-1* alone does not have the small phenotype on either parental background, we hypothesize that the trait is a result of a three-way genetic incompatibility between variants in the two accessions and *sr45-1*. By backcrossing and selfing, we have obtained a line in which small size segregates as a recessive Mendelian trait.

In this line, we compared small and large (wild-type) plants using RNA-seq. Replicates were very similar. We observed 306 up-regulated and 37 down-regulated genes. The set of up-regulated genes was significantly enriched for genes with metabolic functions, including transporters.

#14
TRACING THE BOTANICAL ORIGIN OF A PDO (PROTECTED DESIGNATION OF ORIGIN) WHITE TEA PRODUCT USING SINGLE NUCLEOTIDE POLYMORPHISM (SNP) MARKERS

Lin Zhou[1,2], Huawei Tan[1,2], Lyndel W. Meinhardt[1], Sue Mischke[1], and Dapeng Zhang[1]
[1] USDA-ARS-NEA, Sustainable Perennial Crops Laboratory, BARC
[2] Nanjing Agricultural University, College of Horticulture, Nanjing, Jiangsu, China

Abstract: The market for specialty tea has been rapidly expanding due to the increased revenues and profits brought to growers and industry. Accurate verification of genetic identity is of critical importance to ensure authentication of specialty tea products. Fuding white tea from Fujian province, China, is one of the PDO tea products made from local traditional varieties, and it commands a premium market price. Adulteration in this product has been pervasive, both in domestic and international markets. It is in the interests of commerce to guarantee the botanical origin of white tea and other specialty products. In return, this assurance provides economic incentive for on-farm conservation of traditional tea germplasm. The objective of this work is to develop a method for authentication of the botanical origin of Fuding white tea. A total of 30 white tea products from domestic and international markets were sampled and analyzed, using a panel of 192 Single Nucleotide Polymorphism (SNP) markers. The panel of samples also included 50 reference varieties, representing a majority of the traditional tea varieties in China. The genotyping was performed using nanofluidic 96x96 array plates. Based on the SNP profiles, the true-to-type commercial Fuding white tea can be unambiguously differentiated from the fraudulent products. Moreover, the positive identification can be achieved using a single tea bud or leaf, which enabled the establishment of a link between a particular individual tea leaf with a specific tea variety. This method can be readily applied to other PDO tea products, including green and oolong teas, thus significantly enhancing the capacity to protect the unique character and reputation of these premium tea products. The same method can also be used as an effective tool for genotype verification in germplasm management, breeding and seed propagation.

#15
INTRASPECIES VARIATION IN GREEN ASH RESPONSE TO AN INVASIVE INSECT

Di Wu, John E. Carlson
The Penn State University, 321 FRB, University Park, PA, 16801
dxw5099@psu.edu

Green ash (*Fraxinus pennsylvanica*) is a medium-sized, ecologically and economically valuable tree species native to the eastern and central United States. However, the widely distributed green ash species in North America is under severe threat from the rapid invasion of emerald ash borer (*Agrilus planipennis*; EAB), an Asian wood-boring beetle. To understand the mechanism of the defense response, transcriptomes were prepared for six green ash genotypes exposed to EAB infestation, using an RNA-seq approach. Mapping these reads to the *de novo* assembled reference of 107,611 transcript contigs, prepared from 98 Gb of RNAseq data from multiple tissues and treatments (www.hardwoodgenomics.org/node/68249), enabled differentially expressed genes to be identified between resistant and susceptible genotypes and between control and treated bark samples. The enrichment analysis showed that most of the overrepresented GO terms were related to stress response in resistant genotypes. In addition, our results indicate that the response process was associated with induced, rather than suppressed, gene expression.

To understand more about this serious forest health issue and to assist in green ash protection and restoration, a genetic diversity study was also conducted with 429 green ash accessions collected from 60 provenances across the species' natural range, using SSR markers. Our results revealed three distinct sub-groups of provenances. Northern provenances fell into one group, southern provenances into a second group, and the third sub-group of provenances consisted of admixtures of northern and southern genotypes. In addition to genetic variation, phenotypic variation in growth and in EAB resistance and susceptibility was assessed and compared between the three groups of provenances. Our results showed that the admixture group has significantly larger diameters than the distinct Northern and Southern groups. Although no significant difference has been detected in canopy health ratings among the three groups, we observed that both the Northern group and admixed group had significantly higher numbers of EAB exit holes per m^2 the provenances in the Southern group.

We hope that this study will support further research on the basis of apparent low frequency natural EAB resistance in green ash and lead to strategies for eventual restoration of the species. This project was supported by a grant from NSF's Plant Genome Research Program (IOS-1025974).

Participant	Mailing address	Phone and Email
Abdelkreem, Reham	USDA-ARS Soybean Genomics and Improvement Laboratory, 10300 Baltimore Ave, Bldg 006, Beltsville, MD 20705	301-504-0259, Reham.youssef@ARS.USDA.GOV
Acly, Emma	USDA ARS Appalachian Fruit Research, 2217 Wiltshire Rd, Kearneysville, WV 25430	3047253451 220, Elizabeth.Lutton@ARS.USDA.GOV
Adusei, Afua	Morgan State University, 1700 E. Coldspring Lane, Baltimore, MD 21251	4438853540, afadu1@morgan.edu
Alkharouf, Nadim	Towson University, 8000 York Road, Dept of Computer and Information Sciences, Towson, MD 21252	410-704-3149, nalkharouf@towson.edu
Ashia, Joseph	Salisbury University, Dept. of Biology, 1101 Camden Ave., Salisbury, MD 21801	4105485590, ajoseph2@gulls.salisbury.edu
Assadian, Mariam	Macherey-Nagel Inc., 2850 Emrick Blvd., Bethlehem, PA 18020	915-920-1231 , massadian@mn-net.com
Baskerville, Andrew	Salisbury University, Dept. of Biological Sciences, 1101 Camden ave, Salisbury, MD 21801	atbaskerville@salisbury.edu
Bodnar, Anastasia	USDA Aphis BRS, 4700 River Road, unit 150, Riverdale, MD 20737	301202-4644, anastasia.l.bodnar@aphis.usda.gov
Brewer, Eric	USDA, ARS, BARC, NGRL, 10300 Baltimore Ave., Bldg 004, Rm 025 BARC West, Beltsville, MD 20705	301-504-5458, eric.brewer@ars.usda.gov
Campbell, Kimberly	USDA-ARS, 10300 Baltimore Ave, Bldg 006, Rm 213, Beltsville, MD 20705	301-504-0330, kimberly.campbell@ars.usda.gov
Cohen, Jesse	Bioline USA Inc.	(703) 217-2006 , Jesse.Cohen@bioline.com
Coker, Jim	University of Maryland, University College, ,	james.coker@umuc.edu
Collins, Ron	USDA, ARS, SPCL, 10300 Baltimore Ave, Bldg 001, Rm 232, BARC-West, Beltsville, MD 20705	301-504-6135, Ron.Collins@ars.usda.gov
Collum, Tamara	University of Maryland, Institute for Bioscience & Biotechnology, 4291 Fieldhouse Dr., Plant Sciences Bldg, Rm 5115, College Park, MD 20742	3014052852, tdfisch@umd.edu
Cooper, Bret	USDA-ARS, 10300 Baltimore Ave, Bldg 006, Rm 213, Beltsville, MD 20705	301-504-9892, bret.cooper@ars.usda.gov
Dardick, Chris	USDA ARS Appalachian Fruit Research, 2217 Wiltshire Rd, Kearneysville, WV 25430	3047253451 220, chris.dardick@ars.usda.gov
Darling, Tylen	Howard Commnity College, 10901 Little Patuxent Pkwy, Columbia, MD 21044	4435183406, tdarling@howardcc.edu
Dickinson, Cody	Virginia Tech, 220 Ag Quad Lane, Blacksburg, VA 24061	8044454328, Chrisd1@vt.edu
Eicholtz, Matthew	ARS USDA BARC NGRL, 10300 Baltimore Ave, Bldg 004, RM 025, Beltsville, MD 20705	301 504 5458, Matthew.eicholtz@ars.usda.gov

Enitan, Oluwatomisin	Morgan State University, 1700 E. Coldspring Lane, Baltimore, MD 21251	4438853540, oleni2@morgan.edu
Erickson, Les	Salisbury University, Dept. of Biological Sciences, 1101 Camden ave, Salisbury, MD 21801	4106670788, flerickson@salisbury.edu
Fathabad, Somayeh	Morgan State University, 1700 E. Coldspring Lane, Baltimore, MD 21251	4438853540, sogha1@morgan.edu
Flurer, Brittany	Salisbury University, Dept. of Biology, 1101 Camden Ave., Salisbury, MD 21801	4105485590, bflurer1@gulls.salisbury.edu
Frederick, Reid	USDA ARS FbWSRU, 1301 Ditto Ave, Fort Detrick, MD 21702	301-619-7344, Reid.Frederick@ars.usda.gov
Fu, Shimin	USDA ARS MPPL, 10300 Baltimore Ave., Beltsville, MD 20705	3015046374, xiaomin.fu@ars.usda.gov
Fuentes-Bueno, Irazema	ARS USDA BARC NGRL, 10300 Baltimore Ave, Bldg 004, RM 025, Beltsville, MD 20705	301 504 5458, irazema.fuentes@ars.usda.gov
Galimba, Kelsey	USDA, ARS, Appalachiam Fruit Research Station, 2217 Wiltshire Rd, Kearneysville, WV 25430	304-725-3451, Kelsey.galimba@ars.usda.gov
GELVIN, Stanton B	Purdue University , Dept. of Biological Science, 201 S. University St., West Lafayette, IN 47907	765-494-4939, gelvin@purdue.edu
Grinstead, Sam	ARS USDA BARC NGRL, 10300 Baltimore Ave, Bldg 004, RM 22, Beltsville, MD 20705	3015045458, sam.grinstead@ars.usda.gov
Gupta, Subhash	USDA BRS International Collaboration, 4700 River Road # 147, Riverdale, MD 20737	301-851-3956, subhash.c.Gupta@aphis.usda.gov
Guseman, Jessica	USDA ARS Appalachian Fruit Research, 2217 Wiltshire Rd, Kearneysville, WV 25430	3047253451 220, Jessica.guseman@ars.usda.gov
Hammond, John	USDA ARS USNA FNPRU, 10300 Baltimore Ave. Bldg. 010A , Beltsville, MD 20705	3015045313, john.hammond@ars.usda.gov
Hartung, John	USDA ARS MPPL, 10300 Baltimore Ave, , Beltsville, MD 20705	301-504-6374, john.hartung@ars.usda.gov
Haymes, Kenneth	USDA Aphis BRS, 4700 River Road, Riverdale, MD 20737	3018513879, kenneth.m.haymes@aphis.usda.gov
Holland, Mark	Salisbury University, Dept. of Biology, 1101 Camden Ave., Salisbury, MD 21801	4105485590, maholland@salisbury.edu
Hurtado Gnzales, Oscar	USDA/ARS/BARC/SGIL, Bldg. 006. 10300 Baltimore Ave., Beltsville, MD 20705	301-504-5484, oscar.gonzales@ars.usda.gov
Ibrahim, Heba	University of Maryland College Park, Dept Cell Biology & Molecular Genetic , 4065 Campus Dr., College Park, MD 20742	3014059904, heba.m.m.ibrahim@gmail.com
Islam, Nazrul	USDA, ARS, SGIL, 10300 Baltimore Ave., Bldg 006, BARC West, Beltsville, MD 20705	301-504-5316, Ronghui.yang@ars.usda.gov
Jelesko, John	Virginia Tech, 220 Ag Quad Lane, Blacksburg, VA 24061	5402313728, jelesko@vt.edu
Jones, Richard	USDA ARS/ GIFVL, 10300 Baltimore Ave, Bldg 010A, Rm 311 Barc West, Beltsville, MD 20705	301-504-8395, richard.jones@ars.usda.gov

Jordan, Ramon	US National Arboretum, USDA ARS, 10300 Baltimore Ave, Bldg 010A, Rm 129, Beltsville, MD 20705	3015045646, ramon.jordan@ars.usda.gov
Jurick, Wayne	USDA ARS, 10300 Baltimore Ave., Beltsville, MD 20705	3015046980, wayne.jurick@ars.usda.gov
Khan, Rana	University of Maryland, University College, 3501 University Blvd. East, Adelphi, MD 20783	2406842430, rana.khan@umuc.edu
Kim, Joonyup	USDA/ARS/BARC/SGIL, Bldg. 006. 10300 Baltimore Ave., Beltsville, MD 20705	301-504-6091, joonyup.Kim@ars.usda.gov
Kinkel, Linda	Department of Plant Pathology, University of Minnesota, 495 Borlaug Hall, 1991 Upper Buford Circle, St. Paul, MN 55108	612 625 0277, kinkel@umn.edu
Knill, Courtney	USDA ARS Appalachian Fruit Research, 2217 Wiltshire Rd, Kearneysville, WV 25430	3047253451 220, Elizabeth.Lutton@ARS.USDA.GOV
Lawrence, Susan	USDA, ARS, BARC, IIBBL, 10300 Baltimore Ave, Bldg 007, Rm 301 BARC-west, Beltsville, MD 20705	301-504-6144, susan.lawrence@ars.usda.gov
Li , Haiyan	USDA-ARS, BARC, MPPL, 10300 Baltimore Ave., Bldg. 004/Rm121, Beltsville, MD 20705	301-504-7118, haiyan.li@ars.usda.gov
Liu, Huawei	USDA ARS MPPL, 10300 Baltimore Ave., Beltsville, MD 20705	3015046374, Huawei.Liu@ars.usda.gov
Maliga, Pal	Waksman Institute of Microbiology, Rutgers University, 190 Frelinghuysen Rd, Piscataway, NJ 08854	(848) 445-5329, maliga@waksman.rutgers.edu
Mandava, N. Bhushan	Mandava Associates, LLC, 1050 Connecticut Avenue, N.W., Suite 500, Washington, D.C. 20036	(202) 223 - 1424, bhushan@mandava.com
Matthews, Ben	USDA-ARS Soybean Genomics and Improvement Laboratory, ,	(443) 280-2492, benfmatthews@gmail.com
McGonigle, Brian	DuPont Crop Protection, 1090 Elkton Rd., Newark, DE 19711	3023665322, brian.megonigle@dupont.com
Mclaughlin, Margaret	ARS USDA BARC NGRL, 10300 Baltimore Ave, Bldg 004, RM 22, Belstville, MD 20705	3015045458, margaret.mclaughlin@ars.usda.gov
McMahon, Michael	USDA ARS FbWSRU, 1301 Ditto Ave, Fort Detrick, MD 21702	3016192232, michael.mcmahon@ars.usda.gov
Mollov, Dimitre	ARS USDA BARC NGRL, 10300 Baltimore Ave, Bldg 004, RM 22, Belstville, MD 20705	3015045458, dimitre.mollov@ars.usda.gov
Mount, Stephen	University of Maryland College Park, Dept Cell Biology & Molecular Genetic , 4065 Campus Dr., College Park, MD 20742	3014059904, smount@umd.edu
Murphy, Angus	Dept. of Plant Science and Landscape Architecture, University of Maryland, College Park, MD 20742	3014056244, asmurphy@umd.edu
Natarajan, Savithiry	USDA-ARS Soybean Genomics and Improvement Laboratory, 10300 Baltimore, Ave. Bldg. 004, Rm 211, Beltsville, MD 20705	301-504-5258, savithiry.natarajan@ars.usda.gov

Pedley, Kerry	USDA ARS FbWSRU, 1301 Ditto Ave, Fort Detrick, MD 21702	301-619-1668, Kerry.pedley@ars.usda.gov
Pinney, Stephen	USDA, ARS, SPCL, 10300 Baltimore Ave, Bldg 001, Rm 232, BARC-West, Beltsville, MD 20705	301-504-7317, Stephen.pinney@ars.usda.gov
Poudel, Deepak	Virginia Tech, 220 Ag Quad Lane, Blacksburg, VA 24061	5712086116, poudel12@vt.edu
Puthoff, David	Frostburg State University, 101 Braddock Road, Frostburg, MD 21532	301-687-4172, dpputhoff@frostburg.edu
Reinsel, Michael	USDA Floral and Nursery Plant Research Unit, Bldg 010A. BARC West, 10300 Baltimore Blvd., Beltsville, MD 20705	3015049424, michael.reinsel@ars.usda.gov
Rowland, Jeannie	USDA-ARS, GIFVL, BARC-West, 10300 Baltimore Ave, Bldg 010A, Rm 247, BARC-West, Beltsville, MD 20705	301-504-6654, Jeannine.Rowland@ars.usda.gov
Ruck, Amy	USDA ARS FbWSRU, 1301 Ditto Ave, Fort Detrick, MD 21702	301-619-0517, Amy.ruck@ars.usda.gov
Saunders, James	Towson University, 14590 Triadelphia Mill Road, Dayton, MD 21036	443-386-4695, jsaunders@towson.edu
Saunders, Willow	Salisbury University, 14590 Triadelphia Mill Road, Dayton, MD 21036	2405652595, willowbrook3kid@aol.com
Schultz, Burkhard	University of Maryland, Plant Science Dept., College Park, MD 20742	3014051317, bschulz1@umd.edu
Scorza, Ralph	USDA-ARS-AFRS, 2217 Wiltshire Road, Kearneysville, WV 25430	3047253451, Ralph.Scorza@ars.usda.gov
Sechler, Aaron	USDA ARS FDW SRU, 1301 Ditto Ave, Fort Detrick, MD 21702	301-619-2232, aaron.sechler@ars.usda.gov
Segobye, Kabelo	University of Maryland, Plant Science Dept., College Park, MD 20742	3014051317, ksegobye@umd.edu
Serrels, Joanne	USDA, 4700 River Road, Unit 147, Riverdale, MD 20737	3018513867, joanne.m.serrels@aphis.usda.gov
Shen, Zhengxing	USDA, 4700 River Road, Unit 147, Riverdale, MD 20737	3018513922, zhengxing.shen@aphis.usda.gov
Smigocki, Ann	USDA-ARS, BARC, MPPL, 10300 Baltimore Ave., Bldg. 004/Rm122, Beltsville, MD 20705	301-504-7118, ann.smigocki@ars.usda.gov
Stone, Christine	Foreign Disease Weed Science Research Unit, 1301 Ditto Ave, Fort Detrick, MD 21702	301-619-2862, Christine.stone@ars.usda.gov
Tabatabai, Behnam	Morgan State University, 1700 E. Coldspring Lane, Baltimore, MD 21251	4438853540, betab1@morgan.edu
Tucker, Mark	USDA, ARS, SGIL, 10300 Baltimore Ave., Bldg 006, BARC West, Beltsville, MD 20705	301-504-6091, mark.tucker@ars.usda.gov
Tumakong, Yvette	USDA, ARS, BARC, NGRL, 10300 Baltimore Ave., Bldg 004, Rm 025 BARC West, Beltsville, MD 20705	301-504-5458, yvette.tumakong@ars.usda.gov
Ulrey, Thomas	Mount St. Mary's University, 3 W. 5th St., Frederick, MD 21701	2404463782, thulrey@email.msmary.edu

Urrutia, Cesar	ARS USDA BARC NGRL, 10300 Baltimore Ave, Bldg 004, RM 025, Beltsville, MD 20705	301 504 5458, Cesar.urrutia@ars.usda.gov
Wang, Wanyan	Pennsylvania State University, 321 Forest Resources Bldg., University Park, PA 16803	814-7777998, wuw123@psu.edu
Wu, Di	Pennsylvania State University, 321 FRB, University Park, PA 16802	814-9542336, dxw5099@psu.edu
Wu, Liping	USDA, ARS, BARC, NGRL, 10300 Baltimore Ave., Bldg 004, Rm 025 BARC West, Beltsville, MD 20705	301-504-5458, liping.wu@ars.usda.gov
Yang, Yinong	Pennsylvania State University, 405C Life Science Building, University Park, PA 16802	814-867-0324, yuy3@psu.edu
Zhang , Chong	University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21052	443-641-3959, zh1@umbc.edu
Zhang, Dapeng	USDA, ARS, SPCL, 10300 Baltimore Ave, Bldg 001, Rm 223, BARC-West, Beltsville, MD 20705	301-504-7477, Dapeng.Zhang@ars.usda.gov
Zhang, Deshui	USDA, 4700 River Road, Unit 147, Riverdale, MD 20737	3018513922, deshui.zhang@aphis.usda.gov
Zhou, Lin	USDA, ARS, SPCL, 10300 Baltimore Ave, Bldg 001, BARC-West, Beltsville, MD 20705	301-504-7477, lin.zhou@ars.usda.gov