35th Annual Mid-Atlantic Plant Molecular Biology Society

National Wildlife Visitor Center, Patuxent Research Refuge - Laurel, MD

August 14 & 15, 2018

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

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Organizing Committees: Lots of people provide the support and staffing for this meeting! Many thanks to all of them for the fine job they are doing. If you would like to join a committee and help, please let us know. We are always looking for dedicated volunteers!

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Jim Saunders
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Ben Matthews
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WELCOME

Welcome to the 35th annual Mid Atlantic Plant Molecular Biology meeting.

Thank you for coming!!! It will be great to see many old faces and meet many new faces. We have an outstanding group of speakers for this year's meeting, and we hope this meeting will be stimulating for all of you and help keep everyone up-to-date in the ever changing, exciting world of plant molecular biology. 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, who 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 35 years ago, and several folks have participated all 35 years. Several of us are retired, and we especially hope to encourage more of you younger participants to attend the business meeting (Monday 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/

Ben Matthews, chair MAPMBS 2018

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2018 MAPMBS 35th Annual Meeting Schedule

Tuesday, August 14, 2018

9:00 Registration and poster set-up

9:20 Welcome Ben Matthews, James Saunders, John Hammond, David Puthoff

Moderator: Ken Haymes USDA APHIS BRS

9:25 Anne Simon University of Maryland
RNA virus evasion of nonsense-mediated decay.

10:00 Pal Maliga Rutgers University
Engineered PPR10 RNA-Binding Protein for Regulated Gene Expression in Potato Amyloplasts.

10:20 Coffee break: Posters **Posters 1-4 to be judged (please be by your poster)**

11:00 John Jelesko Virginia Tech
First Emperical Evidence for Cardinal as the Penultimate Metabolite in Poison Ivy Urushiol Biosynthesis.

11:20 Chris Clarke USDA-ARS, Beltsville
Using the lens of molecular plant-microbe interactions to understand parasitization by the parasitic weeds of Orobanchaceae.

11:55-1:10 MAPMBS business meeting Lunch break: Posters
Posters 5-12 to be judged (please be by your poster)

Moderator: John Jelesko Virginia Tech

1:10 Jonathan Shao USDA-ARS, NEA, Beltsville
Automated detection of 'Ca. *Liberibacter asiaticus'* Infection in Citrus Using Immune Tissue Prints and Machine Learning using Convoluted Neural Networks.

1:45 Mike Axtell Pennsylvania State University
microRNAs from the parasitic plant *Cuscuta campestris* target host mRNAs involved in defense and phloem function.

2:20-2:25 **KeyGene** Walter Nelson

2:25 - 3:10 **Coffee break: Posters AND Rootbeer Floats** **Sponsored by KeyGene**
Posters 13-18 to be judged (please be by your poster)

3:10 **Introduction of Keynote speaker: Ben Matthews**

3:15 **The Leslie Wanner Keynote speaker:**
Somen Nandi UC Davis
Plant Made Pharmaceutical: Bench to Bedside.

4:15 Close of day; depart the Visitor Center (building closes at 4:30)

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

Wednesday, August 15, 2018

9:00 Registration, posters, exhibitors

9:20 Session moderator: Amanda Kenney USDA APHIS BRS

9:25 Bo Shen Dupont-Pioneer
High oil gene discovery, trait development, and challenges for commercialization.

10:00 Nye Lott
Development of Transgenic Poison Ivy (*Toxicodenron radicans*) Hairy Root Root Cultures: A Transformation-Regeneration Platform for Future Genome Editing Studies of an Irritating Native Weed.

10:20-11:00 Coffee break: Posters **Posters 19-22 to be judged (please be by your poster)**

11:00 Hyoun-Sub Lim Chungnam National University, Daejeon, South Korea
Characterization of new TuMV isolates from Korea as potential candidates for selection of markers to develop resistant crop lines.

11:35 Aimee Malzan University of Maryland
Insights and applications on temperature sensitivity of CRISPR-Cas12a systems in rice, Arabidopsis and maize

11:55 – 1:20 Lunch break: Posters **Posters 23-24 to be judged (please be by your poster)**

Session moderator: David Puthoff Frostburg State University

1:20 Yiping Qi University of Maryland
Plant genome editing with CRISPR-Cpf1 systems.

1:55 Wangshu Mou University of Maryland
The Ethylene Precursor, ACC, may be a Signal in Pollen Tube Guidance Toward Ovules in *Arabidopsis thaliana*

2:15 ****Poster competition awards ceremony****

2:25 Dimitre Mollov USDA-ARS, National Germplasm Resources Lab
High throughput plant virus diagnostics.

3:00 Joyce Van Eck Boyce Thompson Institute
Gene editing as a tool to advance improvement of underutilized crops.

3:35 Close of day – posters down; depart the Visitor Center (building closes at 4:30)

RNA VIRUS EVASION OF NONSENSE-MEDIATED DECAY

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Due to the obligate nature of viruses, host cellular RNA control pathways must be tolerated or circumvented for successful virus amplification. One such pathway is nonsense-mediated decay (NMD), which is ubiquitous in eukaryotes and normally removes aberrant mRNAs containing premature termination codons to prevent detrimental effects caused by expression of truncated proteins. NMD is also triggered by mRNAs with naturally long 3' UTRs, significantly reducing the half-life of these mRNAs in cells. For polyadenylated mRNAs with short 3' UTRs, translation termination includes an interaction between eukaryotic release factor 3 (eRF3) and cytoplasmic poly-A binding protein (PABPC1), which restricts binding of UPF1, the key factor that triggers NMD. Long 3' UTRs limit eRF3-PABPC1 interactions, allowing elevated levels of UPF1 to associate with the termination complex and increasing the likelihood of UPF1 initiating the decay pathway. Multiple genome-wide searches have identified a large number of mammalian mRNAs with exceptionally long 3' UTRs that are resistant to NMD, but the underlying mechanisms that allow these mRNAs to evade NMD are not understood. Many viral genomic (g)RNAs, especially those associated with 3'-co-terminal subgenomic (sg)RNAs, are templates for translation of only the 5' proximal ORF leaving a long 3' UTR (e.g., >3000 nt). Since this translation strategy makes these viruses optimal targets for NMD, most have likely evolved strategies to evade degradation. The few strategies that have been investigated (in animal viruses only) are mainly virus-specific, and include specific proteins that target NMD factors, or specific sequences that attract RNA-binding proteins thus restricting UPF1 binding. Elucidating additional NMD-evasion strategies employed by viruses, especially strategies that could be more widespread, is important for not only understanding virus fitness but also for providing insights into the poorly understood mechanisms used by NMD-resistant cellular mRNAs, which have no sequences or structures in common. Using an agroinfiltration-based NMD assay in Nicotiana benthamiana, we have identified multiple strategies used by the betacarmovirus Turnip crinkle virus (TCV) to evade NMD. The ribosome readthrough structure just downstream of the TCV p28 termination codon stabilized an NMD-sensitive reporter as did a frameshifting element from Pea enation mosaic virus 2. A short, unstructured region (USR) at the beginning of the TCV 3' UTR was also found to increase NMD resistance 3-fold when inserted into an unrelated NMD-sensitive 3' UTR just downstream from the stop codon. Several carmovirus 3' UTRs also conferred NMD resistance despite no sequence similarity in the analogous region. Instead, these regions all displayed a marked lack of RNA structure immediately following the NMD-targeted stop codon. NMD-resistance was unaffected by conversion of 19 pyrimidines in the USR to purines, but resistance was abolished when a 2-nt mutation was introduced downstream of the USR that substantially increased the secondary structure in the region through formation of a stable hairpin. The conserved lack of RNA structure among most carmoviruses at the 5' end of their 3' UTR would enhance sgRNA stability, thereby increasing expression of capsid proteins that also function as RNA silencing suppressors. Since unstructured RNA can serve as an internal ribosome entry site, we propose that the occasional movement of ribosomes past a stop codon via a readthrough/frameshifting element or after internal entry of ribosomes at an unstructured region displaces UPF1, protecting the RNA from NMD. These results demonstrate that the TCV genome has features that are inherently NMD-resistant and these strategies could be widespread among RNA viruses and NMD-resistant host mRNAs with long 3' UTRs. Furthermore, incorporation of a short unstructured region into transcripts expressed in plants with 3' UTRs longer than 200 nt should significantly increase the stability of these transcripts allowing for increased translation of encoded products.

Engineered PPR10 RNA-Binding Protein for Regulated Gene Expression in Potato Amyloplasts

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Constitutive, high-level expression of transgenes in the chloroplast compromises plant growth and interferes with development. To restrict transgene expression to potato tubers, we constructed a transgenic system, in which a Green Fluorescent Protein (GFP) is expressed from a mutant PPR10 Binding Site (BSgg) in plastids. Because the potato PPR10 protein does not recognize the mutant binding site, expression of GFP in the plastids has no impact on plant development and GFP accumulates only to low levels in tubers, 0.05% of total soluble protein. However, tuber-specific expression of PPR10gg enhanced GFP accumulation 40x, to about 2% of TSP.

FIRST EMPIRICAL EVIDENCE FOR CARDINOL AS THE PENULTIMATE METABOLITE IN POISON IVY URUSHIOL BIOSYNTHESIS.

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Urushiol biosynthesis is presumed to originate from fatty acid biosynthesis and proceed through several metabolites that were predicted based upon organic chemistry first principles (Giessman 1967, Dewick 1997). However, none of the predicted urushiol intermediary metabolites have been subsequently experimentally confirmed. Here we report novel urushiol congeners and a novel proposed urushiol biosynthetic intermediate. A GC-MS analysis of TMS-derived extracts from very young germinating poison ivy seedlings was performed to evaluate urushiol congeners for geographic accessional differences. Statistically significant differences in total C15 and C17 urushiols were observed between poison ivy accessions from either Texas or Michigan relative to accessions from Iowa. Virginia and New Jersey accessions showed non-significant intermediate C15 and C17 levels relative to TX, MI, and IA. Thus, like previously reported biometric traits (Benhase and Jelesko 2013), poison ivy urushiol levels showed accession-level evidence for local adaptation in the USA. Previous reports in the literature identified a single C15:0 and two C15:1 (presumed cis/trans isomers at alkenyl double bond) urushiols (Baer et al. 1980). In the poison ivy seedlings, we identified two C15:0 and four C15:1 urushiol isomers. These additional urushiol isomers indicate previously undetected urushiol chemical diversity in the relative placement of the ortho hydroxyl groups to the pentadec(en)yl chain.
Dewick (1997) predicts that anacardic acid is the penultimate metabolite in urushiol biosynthesis. However, there was no compelling evidence for ions with M/Z ratios corresponding to anacardic acid or known fragmentation ions thereof in the poison ivy seedling extracts. Instead, substantial steady state cardinol accumulation levels were observed. This is the first report of cardinols in poison ivy. Moreover, there were two C15:0 cardinol isomers, suggesting two different positions of the single hydroxyl group relative to the pentadecyl chain on the benzene ring. The relative proportions of steady state C15:0, C15:1, and C15:2 cardinol levels were similar to the relative proportions of steady state C15:0, C15:1, and C15:2 urushiol accumulation levels, respectively. Based on these data, we propose that cardinol is the penultimate metabolite in urushiol biosynthesis, and there is more urushiol chemical diversity in poison ivy than previously estimated.

Literature Cited:

Baer, H., M. Hooton, H. Fales, A. Wu, and F. Schaub. 1980. Catecholic and other constituents of the leaves of Toxicodendron radicans and variation of urushiol concentrations within one plant. Phytochemistry 19:799-802.
Benhase, E. B., and J. G. Jelesko. 2013. Germinating and culturing axenic poison ivy seedlings. Hortscience 48:1-5.
Dewick, P. M. 1997. Medicinal natural products: a biosynthetic approach. John Wiley & Sons, Chichester, UK.
Giessman, T. A. 1967. The biosynthesis of phenolic plant products. Page 1209 in P. Bernfeld, editor. The biogenesis of natural compounds. Pergamon Press Ltd.

Using the lens of molecular plant-microbe interactions to understand parasitization by Orobanchaceae parasitic

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Parasitic plants in the family Orobanchaceae, including witchweed Striga hermonthica and broomrape Phelipanche aegyptiaca, are the leading biotic constraints on agricultural production in many locations in Africa, Asia and eastern Europe. Parasitic weeds have historically been studied through the lens of weed management. However, parasitic weeds are more similar to microbial plant pathogens in many ways. Parasitic weeds, like microbial plant pathogens, must interact closely with host plants to extract nutrients and suppress defense responses. In contrast to other classes of pathogens (e.g., bacteria, fungi), little is known about the host plant immune pathways that control the outcome (i.e. resistance or susceptibility) of parasitic weed infection. We screened 47 Arabidopsis mutants defective in known aspects of immune responses against other classes of plant pathogens for altered resistance to virulent P. aegyptiaca. Functional Jasmonic acid and Salicylic acid signaling pathways were essential for full susceptibility of Arabidopsis to P. aegyptiaca. Several genes involved in these pathways are also differentially expressed in response to P. aegyptiaca infection. The core immunity hub gene, PFD6, is essential for maximal resistance to P. aegyptiaca parasitization. Additionally, we cloned 28 putative secreted effector proteins from the P. aegyptiaca transcriptome. Five candidate effector proteins actively suppressed plant immunity in transient assays. Several of these candidate effectors localize to the plasma membrane and are hypothesized to be involved in disrupting pattern triggered immunity through molecular mimicry of a host plant protein.

DETECTION OF 'CA. *LIBERIBACTER ASIATICUS'* INFECTION IN CITRUS USING CONVOLUTED NEURAL NETWORKS

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3 Southwestern University, Chongqing, People's Republic of China

Huanglongbing, associated with infection by 'Ca. *Liberibacter asiaticus'*, has caused catastrophic losses to the Florida citrus industry, and is widespread in urban areas of southern California. The pathogen is known to have a very erratic distribution in infected trees, and a years-long latent period before symptoms appear but during which time the pathogen can be spread by the psyllid vector. Early detection of the pathogen is crucial and qPCR is required to confirm visual symptoms before regulatory actions are taken. Detection of the pathogen using immune tissue printing with a rabbit polyclonal antibody that recognizes epitopes of the major outer membrane protein of 'Ca. Liberibacter asiaticus' has been developed. The assay can be scaled to process large numbers of samples. However, scoring the tissue prints as positive or negative for the pathogen requires an expert to view images of the tissue print to determine if individual phloem cells are stained indicating infection. We have begun the development of an automated system using the Tensorflow® software to create convoluted neural networks for image recognition and risk assessment based on the tissue prints. Preliminary results are promising. We have trained our system using curated training sets of known positive and negative tissue prints that present salient features obtained from graft-inoculated and qPCR-verified trees. Our results will enable rapid, accurate and unbiased scoring of images for the presence of the pathogen and may facilitate the early removal of infected trees.

MICRORNAS FROM THE PARASITIC PLANT CUSCUTA CAMPESTRIS TARGET HOST MRNAS INVOLVED IN DEFENSE AND PHLOEM FUNCTION

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Cuscuta campestris is an obligate parasitic plant that attaches to the stems of host plants and can cause significant losses in several crop systems. Very little is known about *C. campestris* virulence factors. We found that *C. campestris* induces several microRNAs at the site of parasite-host contact. Several of these miRNAs actively target mRNAs from the host, strongly implying that they cross the species barrier. Targeted host mRNAs include defense-related transcripts and a key transcript involved in phloem function. Arabidopsis thaliana mutants in certain target genes support increased growth of C. campestris, consistent with the hypothesis that these microRNAs act as virulence factors during parasitism. A second Cuscuta species, *C. gronovii*, also induces numerous small RNAs at the site of host contact, and some of these also target host genes involved in defense. However, none of the trans-species microRNAs from C. campestris are homologous to any induced, trans-species small RNAs from *C. gronovii*. This implies that the trans-species microRNAs in the Cuscuta genus may have diversified rapidly. Interfering with the activity of trans-species Cuscuta microRNAs might be a new approach to interfere with Cuscuta parasitism.

The Leslie Wanner Keynote Address:

PLANT MADE PHARMACEUTICALS: BENCH TO BEDSIDE

Somen Nandi and Karen A. McDonald

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The ability to genetically engineer plants and plant cells along with a synthetic biology approach and tools creates unprecedented opportunities for the biotechnology industry, particularly for plant-made pharmaceuticals (PMPs). Efficient transformation, agrobacteria infiltration, transfection methods, targeted expression of products, engineered plant hosts, and reliance on well-understood process instruments now make it practical to produce value-added products such as biopharmaceuticals, nutraceuticals, and industrial enzymes using plant-based systems. Commercial production of heterologous proteins using synthetic genes from humans, animals, viral antigens, antibodies, and disease-fighting metabolites has already been achieved. Studies have shown that foreign proteins expressed in plant systems retain their essential structural, biochemical, and biophysical characteristics. Studies on production of effective recombinant molecules produced by using transgenic, transient, and cell culture platforms will be discussed. In addition, the costs associated with scaling-up and process engineering for any commercial product has to be competitive and is dependent on the production platform as well as the final product. Thus, the production cost will be the driving force for commercialization of recombinant proteins.

It is very important to keep the overall integration of process operation in mind during selection and development of any product. Early analysis of developed processes is pivotal in transforming an R&D protocol into a manufacturing process. This has an immense cost impact if processes are frozen at an early stage of clinical trial lot production. Our research indicates the high-level expression of recombinant proteins, along with key parameters for the process development, are crucial to success for PMP industry. The procedure and data required for development of representative bioactive molecules through integration of available knowledge and technology will be discussed.

Selected recent & relevant publications from our lab (http://mcdonald-nandi.ech.ucdavis.edu)

Nandi, S. and McDonald, K. (2014). Expression of Recombinant Proteins in Plant Cell Culture. In: Plant-Derived Pharmaceuticals for Developing Countries. Chapter 3, 20-41. Ed. Kathleen Hefferon. CAB International, Oxford, UK.

Nandi, S. and Khush, GS. (2015). Strategies to increase heterologous protein expression in rice grains. In: Recent Advancements in Plant Expression in Crop Plants. Part II Chapter 7, 241-262, Ed: Kasi Azhakanandam; Aron Silverstone; Henry Daniell and Mike Davey. Springer, New York.

Nandi, S., Kwong, AT., Holtz, BR., Erwin, RL., Marcel, S., and McDonald, KA (2016) Techno-economic analysis of a transient plant-based platform for monoclonal antibody production. mAbs 8(8), 1456-1466.

Corbin, J.M., Kailemia, M.J., Cadieux, C.L., Alkanaimsh, S., Karuppanan, K., Rodriguez, R.L., Lebrilla, C.B., Cerasoli, D.M., McDonald, K.A. and Nandi, S. (2018). Purification, Characterization, and N-glycosylation of Recombinant Butyrylcholinesterase from Transgenic Rice Cell Suspension Cultures, Biotechnology and Bioengineering 115(5):1301-1310.

Xiong, Y., Li, Q., Kailemia, M.J., Lebrilla, C.B., Nandi, S. and McDonald, K.A., (2018). Glycoform Modification of Secreted Recombinant Glycoproteins through Kifunensine Addition during Transient Vacuum Agroinfiltration. International journal of molecular sciences, 19(3), 890.

HIGH OIL GENE DISCOVERY, TRIAT DEVELOPMENT, AND CHALLENGES FOR COMMERCIALIZATION

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Plant oil is an important renewable resource for biodiesel production and for dietary consumption by humans and livestock. Genetic mapping of the oil trait in plants has detected multiple quantitative trait loci (QTLs) with small effects. One major oil QTL encodes an acyl-CoA: diacylglycerol acyltransferase (DGAT1-2) that catalyzes the final step of oil synthesis. Overexpression of high oil DGAT1-2 in maize increases seed oil content by 25%. Stack of ZmDGAT1-2 and ZmWRI1 increases seed oil content as high as 60%. To further increase oil content in soybean, canola and sunflower, we used DNA shuffling to improve DGAT kinetic and stability. Overexpression of shuffled DGAT in soybean increases seed oil content by 20% without affecting seed protein content. The challenges for commercialization of high oil corn and soybean will be discussed. This work demonstrated that engineering of the native DGAT can further increase seed oil content in corn and soybean.

DEVELOPMENT OF TRANSGENIC POISON IVY (TOXICODENDRON RADICANS) HAIRY ROOT CULTURES: A TRANSFORMATION-REGENERATION PLATFORM FOR FUTURE GENOME EDITING STUDIES OF AN IRRITATING NATIVE WEED.

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Poison ivy is a native weed in North America best known for causing allergenic contact dermatitis on upwards of 50 million people per year (Epstein 1987). The poison ivy natural product urushiol is responsible for causing the delayed contact allergenic dermatitis symptoms. The incidence and severity of poison ivy allergenic dermatitis is expected to increase with predicted patterns of global change. Specifically, poison ivy grown in the presence of increased atmospheric CO2 levels results in faster growth, more biomass, and a shift in urushiol congener composition towards more potent allergenic urushiol congeners (Mohan et al. 2006, Ziska et al. 2007). Yet, most aspects of poison ivy physiology and ecology are very poorly characterized. Advances in understanding poison ivy urushiol chemical biology and ecology will require a molecular genetic approach. A de novo transcriptome assembly exists for poison ivy roots and leaves (Weisberg et al. 2017). We previously demonstrated transient transformation of poison ivy leaves (Dickinson et al. 2018).

Here we report the development of Agrobacterium rhizogenes-based poison ivy hairy root stable transformation and regeneration. A. rhizogenes strains R1000 and ATCC15834 were used to initiate poison ivy hairy roots showing auxin independent growth on synthetic media. Leaves were recalcitrant and hypocotyls were permissive to hairy root formation. Hypocotyls from etiolated plants were more permissive than light adapted plants. Between 1-5% of the total hairy roots retained auxin-independent growth after the second passage on selective media. Two independent poison ivy hairy root lines contained lower steady state levels of urushiol compared to wild type poison ivy roots. A. rhizogenes harboring a T-DNA binary vector with a cloned Fire Fly luciferase gene with an artificial intron (LUCINT) was used to generate independent hairy root cultures. Upwards of half of these poison ivy hairy root cultures displayed a bioluminescent phenotype, indicating stable integration and expression of the LUCINT transgene. These results demonstrate the feasibility of producing stable transgenic poison ivy hairy roots that accumulate (albeit lower levels of) urushiol. These findings set the stage for future genome editing of predicted urushiol biosynthetic genes using CRISPR-CAS9 technology.

Dickinson, C. C., A. J. Weisberg, and J. G. Jelesko. 2018. Transient Heterologous Gene Expression Methods for Poison Ivy Leaf and Cotyledon Tissues. Hortscience 53:242-246.
Epstein, W. L. 1987. Plant-induced dermatitis. Annals of Emergency Medicine 16:950-955.
Mohan, J. E., L. H. Ziska, W. H. Schlesinger, R. B. Thomas, R. C. Sicher, K. George, and J. S. Clark. 2006. Biomass and toxicity responses of poison ivy (Toxicodendron radicans) to elevated atmospheric CO2. Proceedings of the National Academy of Sciences of the United States of America 103:9086-9089.
Weisberg, A. J., G. Kim, J. H. Westwood, and J. G. Jelesko. 2017. Sequencing and de novo assembly of the Toxicodendron radicans (poison ivy) transcriptome. Genes (Basel).
Ziska, L. H., R. C. Sicher, K. George, and J. E. Mohan. 2007. Rising atmospheric carbon dioxide and potential impacts on the growth and toxicity of poison ivy (Toxicodendron radicans). Weed Science 55:288-292.

CHARACTERIZATION OF NEW TURNIP MOSAIC VIRUS ISOLATES FROM KOREA AS POTENTIAL CANDIDATES FOR SELECTION OF MARKERS TO DEVELOP RESISTANT CROP LINES

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Radish and Chinese cabbage are the most consumed vegetables in East Asia, and are used especially for Kimchi in Korea. Recently, due to increasing average temperatures, unexpected plant diseases have been reported resulting in loss of agricultural products. In particular, damage caused by viruses has been increasing every year. Because of the importance of monitoring viral diseases, we have investigated viral diseases including Turnip mosaic potyvirus (TuMV) affecting radish (Raphanus sativus) and Chinese cabbage (Brassica rapa). We have isolated twenty-five TuMV isolates from South Korea including Jeju island, and from Northwestern China. These 25 TuMV isolates were cloned in T7 and 35S derived dual-promoter based binary vectors, and differences in symptoms and pathogenesis were investigated by using these infectious clones. Sequence analysis data indicated most TuMV isolates belonged to BR group which infects both Raphanus sativus and Brassica rapa, and shared the highest identity with previously reported Japanese and Chinese isolates. Using hybrid constructs we revealed that the region including the P1, HC-Pro, P3, 6K1, and the N-terminal domain of CI includes a determinant of HR-like response in Nicotiana benthamiana. The interaction of this region with other component(s) of the potyvirus genome also affects the host reactions of commercial Brassica rapa cultivars carrying different resistance genes. We have generated chimeric constructs between mild and severe isolates, which were used to localize the determinant(s) of symptom severity. TuMV isolates carrying different identified determinants of symptoms and pathogenicity will be useful to screen breeding lines to identify additional sources of TuMV resistance.

Insights and applications on temperature sensitivity of CRISPR-Cas12a systems in rice, Arabidopsis and maize

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CRISPR-Cas12a (formerly Cpf1) is an endonuclease valued for its unique properties including, T-rich target sites and sizeable deletions. AsCas12a and LbCas12a systems have been employed in rice, soybean, and tobacco, although not always successfully. CRISPR proteins are part of bacterial adaptive immune systems, and as such, have evolved to function in environments unsuitable for plants. In order to improve Cas12a efficiency in a variety of species, we compared the effect of temperature on AsCas12a and LbCas12a in rice, and LbCas12a in Arabidopsis, maize, and tomato. Surprisingly, we found that temperature had a large effect on LbCas12a efficiency in Arabidopsis but not in rice. AsCas12a was more sensitive to temperature effects than Cas12a. Using these results, efficient production of germline mutants was demonstrated in maize and tomato at higher temperatures. These results provide temperature treatment guidelines for monocots and dicots to facilitate Cas12a genome editing. To further explore which Cas12a mechanism was temperature sensitive, we targeted PAP1 for repression at three different temperatures. PAP1 was best repressed under 22°C, not higher temperatures. This suggests that Cas12a binding is not affected by temperature.

PLANT GENOME EDITING WITH CRISPR- CPF1 SYSTEMS

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Clustered regularly interspaced short palindromic repeats (CRISPR)-Cpf1(Cas12a) has emerged as an effective genome editing tool eukaryotic cells. We compared the activity of Cpf1 from Acidaminococcus sp. BV3L6 (As) and Lachnospiraceae bacterium ND2006 (Lb) in plants, using a dual RNA Polymerase II promoter expression system. LbCpf1 generated biallelic mutations at nearly 100% efficiency at four independent sites in rice T0 transgenic plants, while AsCpf1 showed suboptimal activity. LbCpf1 was further applied for successfully genome editing in maize and Arabidopsis. Whole-genome sequencing of LbCpf1 edited rice plants revealed no off-target mutations, suggesting Cpf1 is very specific for precise genome editing. Despite high activity and specificity of LbCpf1, its applications in plants are limited by its restrictive TTTV (V=A, C, G) PAM (protospacer adjacent motif) requirement. We demonstrated that a Cpf1 ortholog, FnCpf1, can edit many TTV PAM sites in plants. Further, we engineered protein variants of LbCpf1 and FnCpf1 that recognize CCCC, TYCV (Y=C, T) and TATG PAMs, which greatly expanded the target range of Cpf1 proteins for plant genome editing. Moreover, we repurposed AsCpf1 and LbCpf1 for efficient transcriptional repression in Arabidopsis and demonstrated reduction of target gene transcription >10-fold. Our data suggest promising applications of CRISPR-Cpf1 for editing plant genomes and modulating the plant transcriptome.

THE ETHYLENE PRECURSOR, ACC, MAY BE A SIGNAL IN POLLEN TUBE GUIDANCE TOWARD OVULES IN ARABIDOPSIS THALIANA

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It is well established that the phytohormone ethylene is synthesized by a two-step reaction starting from conversion of S-adenosylmethionine to 1-aminocyclopropane-1-carboxylic acid (ACC) by the enzyme ACC synthase (ACS), followed by conversion of ACC to ethylene by ACC oxidase. Numerous ethylene response studies in flowering plants are based on applying ACC (which can be directly added to growth media) in place of ethylene gas. However, our findings here provide an example in which the ACC-ethylene relationship is uncoupled. The acs octuple mutant (with all 8 functional ACS genes knocked out or knocked down) constructed by Tsuchisaka et al. (2009) has shorter siliques as well as fewer seeds than the wild type (WT), but this phenotype is not rescued by ethylene treatment nor is it detected in ethylene-insensitive mutants. This lack of correlation with ethylene raised the possibility that ACC itself could be an independent signal in regulating fertility in Arabidopsis. Here, we analyzed the defect more closely, starting with a series of crosses that revealed that the reproduction defect in the acs octuple mutant derives from the female sporophyte and is dominant over WT. Based on hand crosses and aniline blue staining of pollen tubes, we discovered that WT and mutant pollen tubes do not turn toward ovules of the acs octuple mutant, as well as they do toward WT ovules. In addition, pollen tubes of ethylene-insensitive mutants can turn normally in WT pistils but exhibit less turning in mutant pistils, suggesting that turning of pollen tubes toward the ovules is affected by ACC, not by ethylene. Moreover, a semi-in vivo pollen tube guidance assay with WT and acs octuple ovules in competition with each other indicated that the sporophytic tissue of acs octuple mutant ovules is responsible for the defect in pollen tube attraction. When we pre-treated the ovules with ACC, we could rescue this defect in the semi-in vivo assay. Our findings identify ACC in the female sporophyte is a signal for pollen tube guidance, indicating a novel role for ACC independent of its role as the ethylene precursor in Arabidopsis.

USING HIGH THROUGHPUT SEQUENCING IN PLANT VIRUS DIAGNOSTICS

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Plant viromes are defined by the assembly of viral nucleic acids, both DNA and RNA, associated with any plant sample or community of plants. Plant viral metagenomics is the genomic analysis of the plant virome. High throughput sequencing (HTS) is a metagenomics-based technique used to detect viruses of individual organisms or entire populations. This review aims to demonstrate both benefits and pitfalls to HTS methods.

HTS is now beginning to supersede more traditional plant virus diagnostic methods such as biological indicators, PCR, RT-PCR, ELISA, and electron microscopy. The advantages of HTS are: extreme sensitivity, reliability, and the relative ease of obtaining a whole viral genome sequence. HTS has other applications for plant virology, including studying pathogen diversity, the discovery of new and uncharacterized viruses, and as a tool for virus surveys and large-scale epidemiological studies.

An essential part of the HTS process is to consider the nucleic acid (NA) (DNA or RNA, single or double stranded, size selection, with or without enrichment, etc.) used in the downstream workflow. These considerations provide several sequencing options: total NA and shotgun approach, double stranded RNA, virion purified NA, and small interfering RNAs.

Using HTS for virus detection and characterization requires comprehensive computational efforts. Among many of the challenges with HTS data is the de novo assembly and accurate sequence mapping to known references or newly assembled contigs.

HTS raw data reads consist of NA from viruses, prokaryotic and eukaryotic organisms. To identify and associate these reads with their true biological meaning is a major challenge for the HTS methods. In many occasions reads and contigs are identified with very low identities (~20-40%) to viral taxa and could suggest they represent an uncharacterized virus. However, it can also be a false alarm and such data needs to be carefully evaluated. In most cases a PCR based approach and Sanger sequencing is necessary to validate HTS results. A single NA extract sent for HTS often yields a complete viral genome but one test may have many potential bioinformatic outcomes: 1) a single sequence of the target; 2) multiple, positive target sequences; 3) fragmentary sequence of target; 4) no good assembly evident; 5) heavy contamination sample-to-sample: cross talk between samples and false positives. Another important challenge is the sensitivity of HTS. Processing and analyzing samples with low viral NA titer can lead to overlooking potential infection and result in a false negative detection. Specific examples are presented to illustrate these points.

HTS has been gaining popularity and has aided in the discovery of numerous new viruses. However, there is a gap between HTS data and its biological meaning, which poses yet another challenge for satisfying Koch's postulates. The application of HTS may even cause a methodological shift from disease etiology to metagenomics approaches and creates ambiguity for plant quarantine regulations. Despite its current challenges HTS is very promising approach in plant virus diagnostics and virus characterization.

GENE EDITING AS A TOOL TO ADVANCE IMPROVEMENT OF UNDERUTILIZED CROPS

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The availability of gene editing technologies, especially CRISPR/Cas, has greatly advanced gene function studies and provides the long-term benefit of approaches to precisely manipulate phenotypes to advance crop improvement. These improvements have the potential to secure agricultural productivity by enhancement of characteristics such as yield and resilience to stresses imposed by climate extremes. Traits of interest to us are those that when modified can transform a plant species that is underutilized because of undesirable agronomic characteristics into one with potential to diversify options for agricultural production. Our early work with the Solanaceae family member tomato (Solanum lycopersicum) as a model centered on investigation of gene function as it relates to plant architecture, meristem development, and fruit-related characteristics. Results from this work led us to believe that gene editing could be exploited to fast-track improvement, in a sense fast-track domestication of underutilized plant species. Our subsequent work has transitioned to other solanaceous species, including the closest tomato wild relative, Solanum pimpinellifolium, and members of the distantly related Physalis genus to determine if what we learned from our earlier work with tomato is translatable to improvement or domestication of these species. Within the Physalis we are working with two different species, Physalis pruinosa (groundcherry), which is a diploid and Physalis peruviana (goldenberry) a tetraploid. Through application of CRISPR/Cas-mediated gene editing, we have observed timely improvements of undesirable phenotypes that cements our belief that this technology can indeed be exploited to turn an underutilized species into one with desirable agronomic characteristics within a realistic timeframe. To date, we have targeted a number of genes to affect characteristics such as plant growth habit and fruit size. We observed a more compact growth habit in both tomato and groundcherry by targeting the Self Pruning gene (SP, homolog of Arabidopsis TFL1) and its homolog SP5G. Related to fruit characteristics, we have recovered groundcherry fruit with a 20% increase in weight by editing the CLAVATA1 (CLV1) gene as compared to the wild type, non-edited control. As our research has progressed, we have identified additional traits to improve in Physalis that would be considered undesirable from an agricultural productivity perspective. Through this work we intend to establish editing strategies for key genes that most affect traits such as growth habit, productivity, harvestability and others that if improved would increase the likelihood of underutilized plant species being part of a solution to strengthen food and agricultural security.

Poster #1
EVALUATION OF ANTIMICROBIAL AND ANTITUMOR ACTIVITIES OF GOLDEN OYSTER MUSHROOM (PLEUROTUS CITRINOPILEATUS) FRUITING BODIES EXTRACTS

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The golden oyster mushroom *Pleurotus citrinopileatus* is a popular edible mushroom in the world especially in the eastern countries and have many valuable nutritional and medicinal properties beneficial to human health, a limited number of previous studies on its antimicrobial and antitumor activities. The Infectious disease and cancer are the major leading cause of human death worldwide. Many of these deaths occur because several patients do not have access to effective and affordable antimicrobial and antitumor compounds. The use of medicinal properties of edible mushrooms extracts had advantages over the use of chemical compounds, as edible mushrooms extracts are natural, have less unwanted side effects and some can overcome the bacterial resistance. In this study extraction by different solvents from golden oyster mushroom of P. citrinopileatus fruiting bodies were tested for their ability to inhibit the growth of some pathogenic fungal and bacterial species using agar diffusion method. Also, the extracts were tested for their ability to inhibit the growth of different human cancer cell lines including human liver carcinoma (Hep G2), the human colonic epithelial carcinoma (HCT 116), the human cervical cancer cells (HeLa) and the human breast adenocarcinoma (MCF-7) using 3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. Furthermore, the cytotoxicity effect of the different extracts was tested against isolated mouse hepatocytes. We observed that the hot water and methanol extracts of fruiting bodies were most effective in inhibiting the growth of most fungi and bacteria. The highest effect reported was by hot water extract with 28±2 mean diameters of inhibition zone (MDIZ) against *Staphylococcus aureus* and with 25±2 MDIZ against *Candida albicans* and *Aspergillus fumigatus*. while, The highest antitumor activity was recorded by cold water extract with half maximal inhibitory concentration (IC50) values of 7.1±0.2, 6.1±0.1, 5.8±0.2 and 6.8±0.3 µg/ml against Hep G2, HCT 116, HeLa and MCF-7 cells, respectively with non-significant effect on the normal mouse hepatocytes. In conclusion, extracts of edible mushroom P. citrinopileatus are good sources for antimicrobial and antitumor compounds and I recommend further chemical studies to isolate and identify the active antimicrobial and antitumor compounds from these beneficial extracts.

Poster #2
DEVELOPMENT OF A PCR HIGH-RESOLUTION MELT ASSAY FOR ARTEMISIA ABSINTHIUM

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Artemisia absinthium (wormwood) contains the compounds alpha-thujone and beta-thujone, formerly associated as a primary component of absinthe. When thujone is consumed in large amounts, the consumer can experience psychoactive effects and in severe cases, failure of the renal and nervous system. Although thujone is a regulated by the FDA, wormwood and its compounds are not controlled under the DEA Controlled Substances Act, allowing for the purposeful consumption of thujone in a tea, oil, or pellet to experience a "legal high". Wormwood seeds are legal and can easily be purchased in the United States. We demonstrate a real-time polymerase chain reaction high resolution melt (PCR-HRM) assay with a melt temperature of 84.75 ± 0.068°C using Radiant Green to detect trace biological material from *A. absinthium*. DNA primers were developed based on the NCBI genomic data available on wormwood. The assay was sensitive to 0.01 ng of A. absinthium, with reproducible results. The size of the amplicon was estimated using an agarose gel. The specificity of the assay developed, as well as the results of duplexing this assay against another plant used to obtain a legal high, *Datura stramonium* (jimson weed), will be presented. Continuing to develop HRM assays allows for an inexpensive and efficient way of identifying legal high substances using trace DNA.

Poster #3
Deciphering the Speciation of Crocanthemum genus and assessing the genetic diversity of its species using AFLP Markers

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The genus Crocanthemum belongs to the family Cistaceae a major group of Angiosperms. The species of this genus are native to North, South and Central America, and West Indies. A prominent feature ofCrocanthemum is its production of dimorphic flowers in all but the Californian species. The objectives of this study were to determine if C. dumosum is genetically distinct from C. canadense and to assess the genetic diversity of the four Crocanthemum species populations using AFLP markers. A total of 140 samples were used in the study (17 bicknelli, 23 canadense, 94 dumosum, and 6 propinquum). The six AFLP primers used generated a total of 843 alleles (mean=140.5) of which all were polymorphic. The PIC of primers ranged from 0.96-0.98 (mean = 0.97). PCoA clustered C. dumosum together with C. canadense implying close genetic similarity. C. bicknelli and C. propinquum clustered separately meaning they are genetically distinct from each other and from dumosum and canadense. AMOVA indicated that 56 % variation was within species and 44% variation among species. Based on this study, it can be concluded that dumosum and canadense are a single species or one is a sub-specific variant of the other. Further studies using DNA barcoding are needed to elucidate more into the speciation of the two. These findings need to be taken into consideration in the conservation management of C. dumosum.

Poster #4
ROLE OF INOSITOL PYROPHOSPHATES IN LIPID REMODELING IN ARABIDOPSIS THALIANA

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Phosphate (Pi) is an essential nutrient for plants, required for plant growth and seed viability. Under Pi stress, plants undergo dynamic morphological and metabolism changes to leverage available Pi, including the breakdown of membrane phospholipids. Plants have been shown to "remodel" their lipid membrane profiles under phosphate starvation, degrading phospholipids in the cell membranes and utilizing the generated phosphorus for essential biological processes. By concomitantly inducing a phospholipid hydrolysis pathway and galactolipid biosynthetic pathway, membrane phospholipids are replaced by non-phosphorus containing galactolipids and sulfolipids. The inositol phosphate signaling pathway is a crucial element of the plant's ability to respond to changing energy conditions. Inositol phosphates (InsPs) are synthesized from the cyclic 6-carbon polyol scaffold, myo-inositol. Inositol hexakisphosphate (InsP6) is the most abundant InsP signalling molecule and can be phosphorylated further by VIP kinases, resulting in inositol pyrophosphates (PPx-InsPs). PPx-InsPs have high energy bonds, and have been linked to maintaining phosphate (Pi) and energy homeostasis in yeast. Using tandem mass spectrometry, we are examining the lipid profile of an Arabidopsis vip double mutant, in response to phosphate depletion, to address the role of PPx-InsPs in Pi sensing.

Poster #5
Arabidopsis RNA degradome: Insights about the contribution of exoribonuclease XRN4 in mRNA turnover and developmental processes

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In plants, XRN4 is the 5' to 3' exoribonuclease that functions in cytoplasmic mRNA decay and is homologous to XRN1 of yeast and animals. XRN4 preferentially catalyzes 5' monophosphorylated RNA, such as those found on miRNA-targets following miRNA-guided cleavage and decapped mRNA. XRN4, also known as Ethylene Insensitive 5 (EIN5), impacts ethylene and ABA signaling, seed dormancy, flowering time, and plant responses to heat stress in addition to playing a major role in post-transcriptional gene silencing (PTGS). There is mounting evidence to indicate that XRN4 impacts gene expression at the post-transcriptional level by fine-tuning mRNA stability. Our study investigated XRN4 substrates on a global scale to gain insight about the biological impacts of the enzyme in Arabidopsis. RNA-seq and degradome (Parallel Analysis of RNA Ends, PARE) analysis demonstrated that xrn4 mutants overaccumulate many more decapped, deadenylated intermediates compared to those that are polyadenylated. XRN4 also contributes to nonsense-mediated decay (NMD), an mRNA surveillance pathway, and xrn4 overaccumulates decay intermediates of select NMD targets. Further transcripts carrying DST-element in their 3'UTR were also among XRN4 substrates indicating that the enzyme has a role in sequence-specific mRNA decay. Among XRN4 substrates, some of the over-represented gene ontology (GO) categories include gene products from primary carbon and nitrogen metabolism, hormone signaling (auxin and ethylene), and abiotic stress responses. Of these XRN4 substrates, auxin-responsive Aux/IAA genes were of greatest interest since some code for unstable proteins and are repressors of auxin-responsive gene expression impacting root growth. In xrn4, Aux/IAA transcripts IAA2 and IAA3 showed elevated mRNA levels than the WT. Following transcription inhibition, RNA decay kinetics in seedlings showed that these Aux/IAAs are more stable in xrn4 ($t_{1/2}$>36 min) than the WT ($t_{1/2}$ <20 min), indicating that XRN4 is required for their decay. Intriguingly, validation of PARE using 5'RACE analysis indicated that these Aux/IAA transcripts not only overaccumulated decapped mRNAs, but also 5'capped mRNAs in xrn4. This suggests that in xrn4 either the decapping is inefficient or transcription is elevated for select transcripts. Based on our analyses, we propose that the former is a more likely scenario due to the strong impact of xrn4 on Aux/IAA mRNA stability. Collectively, our results point towards post-transcriptional regulation via XRN4-mediated RNA turnover as one of the mechanisms maintaining cellular levels of Aux/IAA repressors. We further show that xrn4 mutants are insensitive to the stimulatory effects of optimal nitrogen supply and produce significantly fewer (>50%) lateral roots than WT. This root branching defect could be attributed to stable levels of Aux/IAA mRNAs, potentially elevating their protein levels. Further analysis is underway to examine the global RNA stability levels and protein profiles in the roots of xrn4 seedlings subjected to different nitrogen regimes.

Poster #7
ELUCIDATING THE ROLES OF ATVIP1 AND ATVIP2 IN PLANT PHOSPHATE SIGNALING AND INOSITOL PYROPHOSPHATE SYNTHESIS

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Inositol phosphates (InsPs), including inositol hexakisphosphate (InsP6), are unique messengers consisting of a myo-inositol ring and six single phosphate moieties. They have been suggested to serve as proxies for inorganic phosphate (Pi) in Pi sensing and signaling pathways. InsP6 can be converted to InsP7 and InsP8 through phosphorylation events to contain diphospho- and triphospho- groups. These high energy inositol pyrophosphates (PPx-InsPs) are suggested to play a role in Pi sensing though the genes involved in this conversion are still unknown in plants. Multiple yeast and mammalian kinases, including VIP1, phosphorylate InsP6 and InsP7. Our group identified two orthologous plant kinases, AtVIP1 and AtVIP2, that catalyze the conversion of InsP7 to InsP8. We are interested in how levels of InsP6, InsP7, and InsP8 impact Pi sensing. Our approach has been to develop a set of mutants with altered InsP and PPx-InsP levels and test their responses to varying levels of Pi. We describe work in progress using loss- and gain-of function mutants in the Inositol Phosphate Kinase 1 (IPK1) and the AtVIPs. In this mutant set, AtVIP double mutants and IPK1 overexpressor mutants grew more favorably under higher Pi conditions while IPK1 loss-of function mutants were more negatively impacted. These results provide evidence that AtVIP1 and AtVIP2 are key players in how plants sense and respond to varying Pi levels.

Poster #8
Development of Brace Root Primordia.

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A major difference between plants and animals is that plants can form post-embryonic organs de novo and adapt to a changing environment. These new organs can be of the same type as the originating tissue, or organs with completely different identities from their tissue of origin. The study of de novo organogenesis in plants has primarily focused on the development of organs of the same type as the originating tissue (cis-organogenesis). For example, the mechanisms of lateral root development from a parent root is an extremely well-studied process. What has received less attention is the development of organs that are different from their tissue of origin (trans-organogenesis).
Trans-organogenesis in plants is exemplified by the development of roots from stems. These stem-borne roots can be found in a variety of plant species, from tropical mangroves to banyan trees to cultivated maize. Our lab focuses on the development of stem-borne roots in maize (brace roots) due to the vast molecular and biological resources available. Maize brace roots are ubiquitous across genotypes, but the mechanisms regulating their initiation are unknown. In this project, our goal is to define the morphological and molecular mechanisms associated with the initiation of trans-organogenesis in maize.
In this project, we aim to provide the first systematic study of trans-organogenesis and define the developmental processes through which a new organ can be initiated from an organ with a different identity. The results from this proposal will provide insights into de novo organogenesis and allow us to address future questions related to the differences between cis-organogenesis and trans-organogenesis. In addition, the molecular data obtained from this proposal will enable us to apply targeted strategies to disrupt or modify brace root initiation and investigate the impact of developmental changes on function.

Poster #9
Development of Maize and Sorghum

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Our labs goal is to uncover the role and development of brace roots within monocots While maize and sorghum have been widely studied on a whole, little effort has been put into studying brace roots in these systems or in any system. In this study we aim to compare the internal anatomy of maize and sorghum throughout their growth. By doing so we hope to find differences and similarities that may elucidate the function and development of these brace roots.

Poster #10
Genetic Variation Within the Translatome of Plum Pox Virus in Response to Leaf Development and Vernalization

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Plum pox virus (PPV), a member of the Potyviridae is a worldwide threat to stone fruit production, causing serious economic losses. Previous studies have identified a range of genetic variants within infected trees with the potential to impact disease. To further investigate the dynamics of PPV sequence variants, we analyzed high-throughput sequence data generated from ribosomal-associated PPV genomes (termed the translatome) isolated from the leaves of infected plum trees at two, four and six week developmental as well as pre-vernalization buds. Translatomes were taken from both whole leaf and the leaf vascular phloem. Sequences from translatome genomes likely represent those being actively translated and, thus, contributing to the infection process. Additionally, transcripts derived from a cDNA clone of Tobacco mosaic virus (TMV) were used to control for inherent sequencing errors. The translatomes of PPV infected tissues, derived from ribosome pull-downs represent a unique means to identify active virus infection levels. Results from this study show that PPV variants occur in proportionally higher levels in newly developing plum tissues that have the lowest levels of infection while more mature tissue that show high levels of infection display proportionately lower numbers of variants. Combined, these results suggest that tissue developmental stage can impact the level of sequence variation present in a virus population.

Poster #11
THE ETHYLENE PRECURSOR, ACC, ITSELF MAY FUNCTION AS A PLANT HORMONE IN THE LIVERWORT MARCHANTIA POLYMORPHA

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The plant hormone ethylene functions in numerous aspects of development and environmental responses. In higher plants, it is well established that ethylene is synthesized from the precursor 1-aminocyclopropane-1-carboxylic acid (ACC) by the activity of ACC oxidase (ACO). Interestingly, ACO homologs capable of efficiently converting ACC to ethylene are found only in higher vascular plants, whereas the synthesis of ACC appears to be well conserved, even in basal land plants, raising the question of what role ACC plays in these plants. We addressed this question using the model system Marchantia polymorpha (liverwort), a basal land plant. We discovered that treating Marchantia with ACC induces a phenotype that is quite distinct from that of ethylene treatment. In Marchantia gemmalings, ethylene treatment increases overall plant size. Mp-ctr1 knockout mutants created by CRISPR/Cas9 are larger than the wild type (WT), consistent with a constitutive ethylene response, while Mp-ein3 knockout mutants are smaller than WT, consistent with ethylene insensitivity. In contrast, ACC treatment during the early stages of gemmaling development results in severe inhibition of cell differentiation and growth not seen with ethylene treatment.These ACC effects are non-toxic and reversible. A knockout of one of the two ACC synthase (ACS) gene homologs in Marchantia, Mp-acs1, is interestingly larger in size compared to the WT. We have thus unmasked a reversible ACC response that is distinct from ethylene response, leading us to propose that ACC itself serves as plant hormone in Marchantia. We speculate that ACC may be an important signaling molecule that evolutionarily predated the ability of higher land plants to efficiently convert ACC to ethylene.

Poster #12
Conservation of Tunicamycin Biosynthetic Gene Clusters Across Rathayibacter Species

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The APHIS-listed select agent *Rathayibacter toxicus*, causal agent of annual rye grass toxicity, is a Gram-positive bacterium that infects a variety of forage grasses through its close association with seed gall nematodes belonging to the genus Anguina. *Rathayibacter toxicus* produces a tunicamycin-related toxin under undefined field conditions, resulting in sporadic disease outbreaks that cause morbidity and mortality among grazing livestock. At present, little is known about this toxigenic bacterium and the regulatory mechanisms involved in toxin production. Sequencing of several R. toxicus strains has demonstrated the conservation of a putative tunicamycin-like biosynthetic cluster that spans 14 genes, in which 12 of these genes (tunA-tunL) are homologous to the tunicamycin gene cluster (TGC) of Streptomyces chartreusis. Our objective was to characterize the diversity of the TGC within R. toxicus, and determine if the TGC was unique to R. toxicus or present in other Rathayibacter species. Genomic analyses of available toxigenic and atoxigenic grass-associated Rathayibacter species identified two novel tunicamycin-related gene clusters in R. iranicus and an undescribed South African Rathayibacter sp. termed ‘EV’. The potential introduction of any plant-associated toxigenic Rathayibacter species, along with our native Anguina nematode population, could have widespread and severe implications for U.S.agriculture.

Poster #13
EVALUATING GENETIC VARIATION OF ARONIA GERMPLASM ACCESSIONS USING NOVEL SSR MARKERS

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Aronia is a deciduous plant native to eastern North America belonging to the Rosaceae family. Its two most commonly known species are; *A. arbutifolia* and *A. melanocarpa*. A third species, *A. prunifolia*, is thought to be a hybrid of the two but is hard to distinguish from A. melanocarpa. A fourth species, *A. mitschurinii*, is also known but is an intergeneric hybrid between sorbus and Aronia. The fruit of A. melanocarpa have been reported to have significant health benefits and more antioxidants than any other temperate fruit and are targeted for breeding. The objective of the study was to use novel Simple Sequence Repeat (SSR) markers to assess the genetic diversity of the Aronia accessions currently maintained at the University of Connecticut to support Aronia breeding efforts in the United States. The seven Aronia SSR loci yielded 86 alleles ranging in size from 132 to 351 bp, all of which were polymorphic. The Polymorphic Information Content (PIC) of loci ranged from 0.735 to 0.954 (mean = 0.80). Analysis of molecular variance (AMOVA) partitioned 21% and 79% of the total variation to among and within species, respectively. This implies that greater genetic variation exists among accessions within the same species than among species. UPGMA generated dendrogram based on Jaccard's coefficient of similarity grouped accessions into species specific clusters. In conclusion, this study found that the Aronia germplasm collection contains significant genetic diversity among and within its species to support Aronia breeding program.

Poster #14
Evaluating a starch biosynthesis pathway gene for its potential to develop resistant starch wheat

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Over 30 million people in the United States suffer from diabetes and more than 36% of the population are obese. A significant contributor to this statistic is the abundance of nutritionally insufficient food sources for the general public. Consumption of foods containing high levels of resistant starch (RS) can lead to lower postprandial glucose levels and insulin responses, which, in turn, would help those at risk in developing diabetes. Wheat covers more acreage than any other food crop, and being an optimal source of carbohydrates, amino acids, and vitamins and minerals, it is a nutritionally significant food source. Amylose and amylopectin are the two polymers of glucose that make up starch in wheat, and high RS wheat can be developed by altering the amylose: amylopectin ratios in its grains. Starch branching enzymes (SBE) and starch synthase enzymes (SSIIa) are the key enzymatic factors in the regulation of the amylose: amylopectin ratios within wheat. Reports show that alterations in SBE contributes to high RS in wheat, however, little research has been done on SSIIa. In this study, a hard red wheat (HRW) variety, Jagger, was chosen to develop a mutagenized population due to HRW's high nutritional value. The SSIIa mutations were screened in each of the A, B, and D genomes. Within the mutant population, at least one knock-out of the SSIIa gene was identified in each of the genomes, and a full null SSIIa mutant was developed by genetically combining the three mutations. With the A, B, D, and full null mutants, we will now be able to compare amylose:amylopectin ratios and RS content between wild type and mutant HRW by perfoming digestion assays (Megazyme International Ireland Ltd.).

Poster #15
HOST- PATHOGEN STUDIES PROVIDE NEW OPPORTUNITIES ON ANTHRACNOSE RESISTANCE IN PEPPER AND TOMATO

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Anthracnose caused by a diverse number of Colletotrichum species poses a great threat to Solanaceous species such as pepper and tomato. As a hemibiotrophic pathogen, anthracnose caused by Colletotrichum is primarily observed on mature pepper and tomato. However, in recent years, anthracnose has become a more serious problem on immature pepper. Thus, genetic resistance to anthracnose in both immature and mature peppers and tomatoes is highly desirable since other means of protection such as fungicide application are often incomplete and cost-inefficient.
USDA-ARS maintains a large collection of Colletotrichum isolates that originated from infected pepper and tomato fruits. Those isolates display differential host pathogenicity and extensive phylogenetic diversity. Multi-locus phylogenetic analysis grouped these isolates within the four aggregate groups. Lineages were identified that distinguished isolates that rotted both immature and mature pepper fruit from conventional ripe fruit rotting isolates. To determine the gene(s) that are responsible for causing anthracnose lesion development in pepper, we chose 10 isolates for whole genome sequencing. Three of these isolates can cause anthracnose in immature and mature pepper (green fruit rotter), and the other seven rotted ripe but not immature pepper (non-green fruit rotter). Two of these isolates cause lesions only in ripe tomato fruit and do not infect pepper. De-novo assembly and gene prediction were performed to obtain protein sequences. Pan-genome analysis showed that 103 orthogroups were found exclusively shared among green fruit rotters, whereas the number of non-green-rotters-only orthogroups was 37. Predicted proteins were characterized with multiple database. Functional studies are currently underway to identify the gene(s) responsible for anthracnose development in pepper.
Synteny among Solanaceous species and conservation of gene function provide opportunities to apply knowledge about anthracnose resistance from tomato to pepper. A small-fruited tomato breeding line (95L368) provides a high level of ripe fruit rot resistance to a wide range of Colletotrichum species, including isolates that rot immature pepper fruit. SNP-based mapping studies using 95L368 revealed three chromosomal regions associated with anthracnose lesion development. To characterize the underlying molecular basis of this resistance and apply this knowledge to pepper, we obtained tissue extracts from 95L368 and a susceptible control (Rio Grande). Results demonstrate that only the tissue extracts from 95L368 inhibited the growth of C. scoviellei isolate 13NJRP5 on Tomato Broth Agar media. Changes to extract pH and heat and protease treatments did not alter inhibitory effects of the 95L368 extracts, suggesting that the inhibition could be caused by a heat-stable, pH-insensitive compound, not a protein product. Further fractionation indicated that the compound is highly polar. HPLC-MS and NMR will be used to determine and report the chemical structure of this compound. Pathogen functional studies, together with identification of the inhibitory compound in tomato and further analysis of candidate genes mapped to chromosomal regions associated with lesion development, provides unique opportunities to exploit relatedness among Solanaceous species for developing durable anthracnose resistance in pepper and tomatoes.

Poster # 16
ELUCIDATING THE ROLE OF THE FLOWERING ACTIVATOR FLK IN PATHOGEN DEFENSE IN ARABIDOPSIS THALIANA

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Recent studies using the model plant Arabidopsis thaliana have elucidated the crosstalk between the genetic pathways governing flowering time control and pathogen defense. Metabolically, flowering and defense control are costly processes that likely compete for the same resources during plant growth and development. Our laboratory has an Arabidopsis mutant, acd6-1, characterized by constitutive defense and diminutive size. The small size of acd6-1 is inversely proportional to the defense level, which makes acd6-1 an ideal readout to quickly assess defense levels in genetic analyses of defense related mutants. In a mutant screen for acd6-1 suppressors, we identified an allele (flk-5) ofFLK, a canonical flowering activator encoding a putative RNA binding protein that localizes to the nucleus. flk loss-of-function mutants were previously shown to exhibit delayed flowering. We confirmed suppression of acd6-1 with anotherflk allele (flk-1). Additionally, we complemented the late flowering phenotype of flk-1 with a wildtype FLK gene translationally fused with the GFP reporter. To further assess the defense role of FLK, we infected plants with the virulentPseudomonas syringae strain DG3 and found that both flk-1 and flk-5 mutants exhibited increased bacterial growth and diminished accumulation of salicylic acid, a key defense signaling molecule. Interestingly, flk-1 and flk-5 exhibited enhanced resistance to infection with the fungal pathogen Botrytis cinerea. flk-1 and flk-5 showed reduced response to treatment with flg22, a defense elicitor derived from the conserved region of P. syringae flagellin proteins, for reactive oxygen species (ROS) production, root growth inhibition, and callose deposition at the cell wall. Treatment with methyl viologen, an inducer of superoxide production, also yielded a compromised response in flk-1 and flk-5. These results support the involvement of FLK in multiple defense signaling pathways, illustrating the crosstalk between pathogen defense and flower development. Further studies are necessary to elucidate the molecular mechanism underlying the defense role of FLK.

Poster #17
Lipoxygenase Gene Family: Identification and Expression Analysis During Plant Growth & Development, Fruit Ripening and Upon Induced Abiotic Stresses

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Lipoxygenases (LOXs) (EC 1.13.11.12) catalyze the oxygenation of fatty acids and produce oxylipins including the stress hormone jasmonate (jasmonic acid/methyl jasmonate). Little is known about the LOX gene family members in tomato, and less so about which members impact growth and development or those that differentially respond to various abiotic stresses. We have carried out genome-wide identification of 19 LOX gene members in tomato which map to 12 different chromosomes, cluster into four distinct clades and contain specific lipoxygenase domains. Expression patterns highlight gene members that are specifically expressed during either growth, development or fruit ripening. Exposure-time dependent kinetics of expression in response to imposed cold, drought, heat, salinity, or wounding stress defined LOX gene members that were induced or suppressed early during each stress, and the non-responders. A medley of different responses of LOX gene family members to specific plant stress highlights the complexity involved as well as providing gene members that can be utilized for developing plant resistance to different abiotic stresses. In summary, the novel information presented here on LOX gene family members in tomato should help to strategize their genetic manipulation for developing robust, stress tolerant and flavor-rich plants.

Poster #18
ENHANCED LIPID PRODUCTION BY OVEREXPRESSION OF STEROL DESATURASE GENE IN THE CYANOBACTERIUM, FREMYELLA DIPLOSIPHON.

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The freshwater cyanobacterium, Fremyella diplosiphon, has great potential as a commercial biofuel agent due to its fast generation time, available sequenced genome, and capacity to grow under a wide spectrum of light. Previous efforts in our laboratory to overexpress the lipid producing gene, sterol desaturase (SD), in the wild type (WT) F. diplosiphon strain resulted in a transformant B481-SD, with a 63-fold increase in transcript abundance. In the present study, we evaluated the total lipid content and fatty acid methyl ester (FAME) composition in WT and B481-SD strains to determine the effect of overexpression in lipid production. Total lipid yield determined by gravimetric analysis revealed a significant increase in the transformant relative to the WT. FAME analysis of tranesterified lipids detected methyl palmitate, the methyl ester of hexadecanoic acid (C16:0) as the most abundant species, which accounted for 76.35 and 65.93% produced in WT and B481-SD respectively. No significant differences in hexadecanoic acid (C16:0), methyl hexadecenoate (C16:1), methyl tetradeconate (C14:1) and methyl octadecanoate (C18:0) were observed. On the contrary, significant increases in methyl octadecenoate (C18:1), and methyl octadecadienoate (C18:2) were detected in B481-SD, indicating that overexpression enhanced unsaturated fatty acids such as oleic and linoleic acids. Results of our study revealed that overexpression of the sterol desaturase gene increased total lipid content and essential fatty acids, which are primary fatty acid components in biofuels. Future studies will be aimed towards determining FAME abundance in B481-SD using two-dimensional gas chromatography-time of flight mass spectrometry analysis to enable its use a commercial scale biofuel agent. (Supported by Strengthening Historically Black Graduate Institutions Award P031B1410005 and the National Institutes of Health Award UL1GM118973).

Poster #19
IS DIHYDROFLAVONOL 4-REDUCTASE (DFR) GENE RESPONSIBLE FOR ANTHOCYANINLESS PHENOTYPE?

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While biosynthetic pathways of anthocyanins, along with the isolation of corresponding genes, have been well characterized in species such Arabidopsis thaliana (L.), we have investigated the gene responsible for the green (anthocyaninless), phenotype within Wisconsin fast plants. Through the use of PCR of five different genes, designed from BAC end sequences compared to the Arabidopsis thaliana genome, one gene was found to be different. The dihydroflavonol 4-reductase (DFR) gene showed a point mutation within the coding region. In order to confirm that the nonfunctional DFR gene was the cause for the green phenotype, PCR product from an amplification of the wild type DFR gene was cloned into pBI121 vector. Non-purple fast plants were transformed using agrobacterium containing the DFR gene by floral dip method. The few transformed plants that were obtained did not have purple stems. DNA extracted from the transformed plants showed the presence of the CAMV promoter, kanamycin and DFR gene. Expression of the DFR gene could not be confirmed at the RNA level.

Poster # 20
IDENTIFYING CANDIDATE GENES INVOLVED IN STONE CELL FORMATION IN PLUM

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In the early 1990's, Luther Burbank started a breeding program that resulted in the release of two stoneless plum cultivars 'Miracle' and 'Conquest'. These cultivars had only a 'grain of sand' for the stone.. A potential remnant of this breeding program, 'Stoneless' was chosen to study in order to understand the genes that control the determination and differentiation of fruit endocarp (stone cells). Fruit tissue was collected from two trees of 'Stoneless' as well as from two different plum cultivars with normal stones. RNA was extracted and sequenced (RNAseq) to look for genes that were expressed differently in normal vs abnormal stone cultivars. RNAseq data showed that a number of genes were found to have different expression levels that associated with the 'Stoneless' cultivar. To confirm these results reverse transcriptase qPCR was used. In order to do that, genes that were expressed equally at all stages were needed to compare to the differentially expressed genes. Based on the RNAseq data, nine genes were identified that varied less than 10% between samples ranging from flower buds to endocarp tissue in the different cultivars. Our results suggested that four of the nine genes could be used as a standard for qPCR measured gene expression within these tissues. Using gnorm q base + software to determine which the best 'housekeeping' gene was, no optimal targets were found due to the variability between the normalization factors. Thus all 4 genes were used for normalization and we analyzed the expression of 7 candidate genes using genorm software. Confirming the results obtained with the RNA sequencing data, it was found that a receptor-like protein kinase and a potential cytochrome P450 monooxygenase are expressed at significantly higher levels in Stoneless Fruit. These genes could be involved in inhibiting the formation of the stone. A serine-type endopeptidase and a homeodomain protein are expressed at higher levels in the normal stone'Reine claude' and these genes could be involved in the formation of the stone. Further work will need to be done to follow up on the function of these genes.

Poster #23
EXPLORING THE VIRULENCE GENETICS OF ERWINIA TRACHEIPHILA ON 'AMBROSIA' MELON

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Erwinia tracheiphila, one of the most destructive pathogens of cucurbits, causes bacterial wilt of cucurbits. The two vectors *of E. tracheiphila*, striped (*Acalymma vittatum* F.) and spotted (Diabrotica undecimpunctata howardi B.) cucumber beetles, transmit the disease through their mouthpieces and their residual frass. Transmission occurs when the bacteria enter a wound present on the cucurbit plant. The objective of this project was to explore the virulence genetics of E. tracheiphila. An *E. tracheiphila* Tn5 transposon mutant library was created and screened for mutants that could not cause disease on 'Ambrosia' melon seedlings. About 2,000 mutants were inoculated on 'Ambrosia' melons at the cotyledon stage, as first true leaves began to emerge. Symptoms were observed over the course of ten days. Several mutants were found to cause no wilt or very limited wilt. The DNA flanking the Tn5 transposon insertion sites in mutants that were defective in virulence was isolated and sequenced to determine the genes that were interrupted. Potential functions for some of these mutated genes as related to the plant disease process will be explored. The results indicate that purine and exopolysaccharide biosynthesis are necessary for E. tracheiphila to cause disease on melons. This study has applied molecular genetic tools to the understanding of E. tracheiphila virulence, which is an area that is mostly unexplored. The study paves the way for additional studies of *E. tracheiphila* virulence genetics, including avirulence functions, which could be useful for developing plants resistant to bacterial wilt caused by *E. tracheiphila*.

Poster #22
ON MICROCYSTIN PRODUCTION AND PROTEIN EXPRESSION IN SELECTED CYANOBACTERIA.

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Cyanotoxins such as microcystins produced by cyanobacteria are known to cause major ecological and human health problems worldwide. Previous efforts in our laboratory have shown that exposure of Anabaena cylindrica B629 to 4 g/L NaCl, and A. cylindrica 29414 and Fremyella diplosiphon SF33 to 2 and 4 g/L NaCl resulted in a 13-84% increase in microcystin production suggesting that elevated levels of NaCl induces greater microcystin release. The objective of the present study was to determine the effect of salinity on microcystin production over time and protein expression in these strains. Cultures were grown in standard BG-11/HEPES medium and microcystin levels were compared to those exposed to 1, 2, and 4 g/L NaCl. Three replicates were maintained and cultures grown for 0, 5, and 10 days under fluorescent white light adjusted to 30 μmol m-2s-1 with initial optical density of 0.1 at 750 nm. Microcystin concentrations were quantified using enzyme-linked immunosorbent assay (ELISA) according to the manufacturer's protocol and data analyzed using ANOVA and Tukey's honest significant difference test. In the second phase, total protein of NaCl-treated A. cylindrica B629 & 29414 and F. diplosiphon SF33 cells was extracted and compared to the control. Results of the ELISA study revealed significant differences in day 0 and 5 microcystin levels of A. cylindrica 29414 and F. diplosiphon SF33 exposed to 2 g/L NaCl and A. cylindrica B629 exposed to 1 & 4 g/L NaCl. In addition, significant differences in microcystin concentrations were observed at day 5 in A. cylindrica 29414 cultures amended with 2 g/L NaCl compared to control, 1, and 4 g/L NaCl. Sodium dodecyl sulphate-polyacrylamide gel electrophoresis (PAGE) revealed the presence of an additional band in A. cylindrica B629 cultures exposed to 2 and 4 g/L NaCl at 120-130 kDa suggesting that higher levels of NaCl caused alterations in the proteome. In future studies, protein expression in A. cylindrica B629 & 29414 and F. diplosiphon SF33 will be determined by two-dimensional PAGE and matrix-assisted laser desorption/ionization time-of-flight/time-of-flight mass spectrometry to identify specific microcystin-associated proteins. These findings will assist with the development of preventative measures to control toxic blooms which are significantly detrimental to ecosystem and human health. (Supported by the National Institute of General Medical Sciences of the National Institutes of Health awards #'s UL1GM118973 and TL4GM1189742).

Poster #23
In Vitro Antibacterial Activity of Garlic and Tea Tree Oil

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Background:
To evaluate the antibacterial activity of tea tree oil, and fresh pure garlic against six strains of bacteria.
Methods:
The selected essential oils were screened against one gram-negative bacteria (Escherichia coli) and five gram-positive bacteria (Bacillus cereus, Staphylococcus epidermidis, Bacillus subtilis, and Micrococcus luteus). The 3 different concentrations (1:1, 1:25, 1:50 and mix) using the disc diffusion method.
Results:
The tea tree essential oil and fresh crushed garlic showed antibacterial activity against one or more bacterial strains. The different concentrations were used to test for antibacterial activity (1:1, 1:50, 1:25, and a mix) using the disc diffusion method.
Conclusion:
The 100% tea tree essential oil and fresh crushed garlic exhibited significant inhibitory effects against the tested bacterial strains. tea tree oil and the crushed fresh garlic showed promising inhibitory activity even at low concentrations. In general, E. Coli and M. Luteus were the most susceptible. In Conclusion, the tea tree oil and crushed fresh garlic showed antibacterial activity against the tested strains. They both can be a good source of antibacterial agents.

Poster #24
Effect of Hop stunt viroid on hop cultivars and its distribution in central Washington

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Hop stunt viroid (HSVd) is a major threat to hop cultivation globally. In 2004, HSVd was first detected in hop yards of Washington State, the major hop growing region of the United States. To understand the effect of HSVd on symptomatic and asymptomatic cultivars, a study was conducted using six cultivars of hop to determine the impact on yield. Average yield in case of infected symptomatic cultivars Glacier, Cascade and Willamette was reduced by 62 %, 14% and 34 % respectively compared to non-inoculated healthy plants. No significant yield reduction was observed in case of Nugget, Columbus and Galena cultivars. To identify the current distribution of HSVd in central Washington, a survey was conducted by testing 1,635 hop plants in which 285 plants (17.4 %) were infected with HSVd. Symptom expression is highly variable among different hop cultivars infected with HSVd. Thorough testing and removing infected material is essential to prevent the further spread of HSVd. However, the development of improved diagnostic tools to accommodate large sample numbers was needed. Reverse transcription-polymerase chain reaction (RT-PCR) has been the major technique previously used for HSVd diagnosis; however, it is costly and sample handling is technically challenging. We developed a more robust reverse transcription-recombinase polymerase amplification (RT-RPA) assay to facilitate the processing of multiple samples. The assay was optimized with all major variants of HSVd from other host species in addition to the hop variant. Green house and farm samples were tested by RT-RPA and RT-PCR; a 100% correlation was obtained between results of the two techniques.

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