**2019 Poster Session Abstracts**

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The effect of physiological resistance on behavioral response of *Tetranychus urticae*.

Adekunle Adesanya, Laura Lavine, Fang (Rose) Zhu, and Doug Walsh

Washington State University, IAREC, Prosser, Washington

**Abstract:** Due to its rapid development, high fecundity, overwintering strategy and also adaption to multiple host plants, the two spotted spidermite *Tetranychus urticae* has developed resistance toward a plethora of synthetic and natural pesticides. Hence, the need to adequately study resistance for sustainable integrated pest management. However, most studies on acaricide resistance in the two spotted spidermite have focused on the acute toxicity of the acaricides with no or little information on the contact irritancy and spatial repellency of acaricides on *T. uriticae.* Furthermore, the impact of physiological resistance on the behavioral effect (irritancy, repellency and oviposition) remains unknown in most pest species including *T. urticae*. In this study, we tested the effect of acaricide resistance on the contact toxicity, irritancy, and repellency of widely used acaricide on *T. urticae* strains. Using fenazaqin (Magister®), an inhibitor of mitochondrial electron transport complex I (MET-I) as our model acaricide, we observed that the development of acaricide resistance can modify the behavioral response of *T. urticae* to the acaricide. Similar physiological resistance within MET-I acaricides (fenpyroximate, fenazaquin and pyrabiden) did not lead to corresponding behavioral responses. The repellency and irritancy of an acaricide to *T. urticae* does not appear to be directly linked to their resistance status to the acaricide. Hence, the behavioral effect of an acaricide should be considered in designing integrated mite management programs.

The impact on hop-derived volatile compounds in beer by dry-hopping at different points of fermentation using different strains of hops and yeast.

Justin Alexander, Stacey Williams

New Belgium Brewing, Fort Collins, Colorado

**Abstract:** Multiple factors can influence the flavor profile of dry hopped beers, including when hops are added in the process (pre/post fermentation), the selected hop variety, the age of the hops, the type of hop product, and the time, temperature and mixing parameters used during dry hopping.  Many breweries are now focusing on using dry hopping to produce more “fruit” forward beers.  By taking advantage of the complex biochemical reactions that take place through the different interactions between hops and yeast during fermentation, it is possible to drive to very different dry hop flavors.  This poster will provide a summary of the final volatile products created when different single hop varieties are added at three different points in fermentation with different yeast strains.  Fermentation performance (yeast viability, VDK reduction time, attenuation, gravity, alcohol and pH) was monitored throughout the study.  Hop volatile compounds were monitored using spme/headspace gas chromatography/mass spectrometry (SPME/HS GC/MS).  Yeast volatile compounds were measured at the beginning and end of fermentation by SPME/HS GC/MS.  Organic acids and iso-α-acids were measured by Ultra High Performance Liquid Chromatography (UPLC).  Multivariate analysis was used to determine correlations between the measured variables.  The results of this study suggest that dry hopping on yeast can be a powerful tool to obtain a desired flavor profile.

The instability of dry-hop flavor and aroma in beer.

Bradley Barnette and Thomas H. Shellhammer

Department of Food Science and Technology, Oregon State University

**Abstract:** It is generally recognized within the brewing industry that hop aroma/flavor in beer changes throughout distribution, and it is widely understood that dissolved oxygen has a negative impact on the flavor stability of beer, but there is little published work on oxygen’s impact on hop aroma stability. This study was designed to understand the impact of dissolved oxygen on the analytical and sensory profiles of hop aroma in dry-hopped beers during storage. Dry-hopped beer, procured from a local craft brewery, was prepared with low dissolved O2 throughout its production. This base beer was dosed with oxygen in a controlled fashion to create beers with a variety of dissolved oxygen levels via air injection during the transfer from 1/2 bbl to 1/6 bbl vessels, resulting in four levels: low (~50 ppb), moderate (~100 ppb), medium (~150 ppb), and high (>200 ppb). Treatments were stored at 2°C and 30°C for 2 weeks. A trained panel (10 participants) characterized the sensory changes using a projective mapping and flash profiling approach. Alongside the sensory analysis, non-volatile and volatile chemistry was assessed using HPLC and GC, respectively. Oxygen concentration had a significant impact on the extent of hop aroma degradation and overall beer flavor modification, with a loss of hoppy, herbal/spicy, and citrus aromas and an increase in stale/wet cardboard and malty aromas, plus an increase in sweet flavor. These changes were observed during storage at both temperatures, albeit to different degrees. Outcomes from this work provide guidance to brewers producing hop-forward beer styles.

Development of race specific molecular assay for detection of hop powdery mildew.

Mary H. Block, Michele Wiseman, David H. Gent

OSU Crop Science, OSU Botany/Plant Pathology, USDA-ARS, Corvallis, Oregon

**Abstract:** Hop powdery mildew (*Podosphaera macularis*) was confirmed in the Pacific Northwest in 1996. Prior to 2012, the most common race of *P. macularis* carried virulence genes that had overcome Rb, R3, and R5. Breeders had successfully introduced qualitative resistance into hop cultivars. Post 2012 a new race of *P. macularis* was discovered to have overcome R6-based resistance. Another race has been found to infect Cascade, a hop cultivar with known partial resistance to powdery mildew. Current methods for race determination for *P. macularis* are slow, costly, and labor intensive. This project intends to create a rapid and inexpensive race specific assay to detect strains of *P. macularis* that are R6-virulent. Forty-six isolates were collected from the PNW region and divided into three categories: Cascade-adapted, pre-2012 and R6-virulent. The transcriptome of the epiphytic mycelia and conidia was sequenced and loci that differentiated the three races were identified by the G'st statistic. Sixteen loci that differentiated R6-virulent isolates from the others were identified. Primers and probes will be designed from a 100 nucleotide region containing 5 single nucleotide polymorphisms. The primers will be tested against *P. macularis* isolates with known virulence. After confirming the efficacy of these primers against known isolates they will be tested against *P. macularis* collected from various hop fields across Oregon and Washington and phenotyped to confirm the accuracy of the assay. This poster will summarize research progress towards development and validation of the race-specific assay.

Hops variety trials in Maryland: The first three years.

Peter Coffey, Andrew Kness, and Bryan Butler

University of Maryland Extension

**Abstract:** In an effort to support Maryland’s new and rapidly growing brewing industry, the University of Maryland Extension, in partnership with Flying Dog Brewery, established a ½ acre hop yard at the Agricultural Experiment Station in Keedysville, Maryland. This planting contains 24 varieties selected in consultation with local growers and brewers. All varieties are replicated 3 times, and are managed intensively with regard to fertility and irrigation, as well as insect, disease, and weed management using IPM principles. The goal of this trial is to determine what varieties may be best suited for production in Maryland, and what challenges and opportunities Maryland growers may encounter along the entire production chain from growing to pelletization to final product. Compared with current American areas of high hops production, Maryland is hotter, more humid, at a lower latitude, and has a large variety of insect pests and soil types.

 Current varieties in the trial;

1. Planted May *2016*: Alpharoma, Cascade, Centennial, Chinook, Crystal, Mt. Hood, Mt. Rainier, Nugget, Sorachi Ace, Southern Cross, Tahoma, and Ultra.
2. Planted in *2017*: Canadian Red Vine, Galena, Glacier, Amallia, Neo 1, New Port, Multi Head, Southern Brewer, Teamaker VF, Willamette, Vojvodina, and Zeus.
3. Multi Head and Neo 1 have been removed and will be replaced for the 2019 season by plants believed to be historic Maryland cultivars.

Identification of Powdery Mildew resistance in a hop (*Humulus lupulus* L.) germplasm collection.

J. S. Havilla, T. B. Morcolb, E. J. Kennellyb, P. D. Matthewsc, and G. J. Muehlbauera

a Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108

b Department of Biological Sciences, Lehman College and The Graduate Center, The City University of New York, Bronx, NY 10468

c Hopsteiner, S. S. Steiner Inc., New York, NY 10065

**Abstract:** Commercial hop (*Humulus lupulus* L.) production is commonly limited by fungal diseases. Hop powdery mildew, caused by *Podosphaera macularis* (Wallr.) Braun & Takam., occurs in major hop production regions in the Northern Hemisphere and causes significant economic losses. Control of this disease is accomplished using combinations of fungicides and host resistance. *R* genes have been identified in a number of hop varieties though some may no longer be effective depending on pathogen strains present in a production region. New sources of resistance would provide hop breeders and growers with the tools necessary to help reduce damage caused by hop powdery mildew. Upon screening a wild germplasm collection consisting of 6036 seedlings and clones, collected from 209 locations, we identified 38 populations possessing individuals that were completely immune following inoculation with an isolate commonly found in major production regions. We screened an additional subset of cultivated hop varieties in a similar manner which represented materials from 10 different hop breeding and improvement programs. Assessments were made on single plants using a five-step ordinal scale representing different disease classes used to reflect the qualitative or quantitative nature of disease present on each genotype. Further examination is required to determine if these are novel resistance loci and determination of their underlying genetic architecture.

Impact of harvest maturity on the aroma characteristics and chemistry of Cascade hops.

Authors/Collaborators: Scott Lafontaine\*1, Scott Varnum2, Aurélie Roland3, Stéphane Delpech3, Laurent Dagan3, Daniel Vollmer1 and Thomas Shellhammer1

1. Department of Food Science and Technology, Oregon State University, 100 Wiegand Hall, Corvallis, OR
2. John I. Haas Inc., 1600 River Rd, Yakima, WA, USA
3. Nyseos, 53 Rue Claude François, Parc 2000, 34080, Montpellier, France

**Abstract:** Cascade is the main hop variety used by U.S. craft brewers, especially for late and dry-hopping. How harvest maturity impacts the aroma potential and chemistry development of this variety is not well understood. Working with a commercial hop farm in Washington, a unique harvest maturity sampling protocol was performed to evaluate the impact on Cascade dry-hop aroma intensity and quality. This sampling yielded 5-6 weekly time points for each of 2014, 2015 and 2016 harvest years. We observed that Overall Hop Aroma Intensity (OHAI) and Citrus quality increased as a function of harvest date. That is, later picked Cascade hops tended to produce beers with higher overall hop aroma intensities that were primarily citrusy in quality. Total oil content displayed a significant positive trend with the harvest date. At an individual component level, a number of different hop volatiles correlated with the harvest date. Most notably geraniol concentrations increased significantly with harvest maturity, and hops harvested late had ~2x, ~12x, and ~ 4x more geraniol than the early harvested samples in 2014, 2015, and 2016 harvests, respectively. Three years of data from this plot indicates that later picked Cascade hops had higher total oils, higher geraniol concentrations, and a more intense dry-hop aroma than earlier picked hops.

Application of metabolomics to discover influence of genetic and environmental factors affecting expression in hops.

Authors/Collaborators: Katie J. Nasiatka1, Scott Dorsch3, Paul D. Matthews4, Harmonie M. Bettenhausen2, Corey D. Broeckling2,3, Adam L. Heuberger2

1. Odell Brewing Company, Fort Collins, CO
2. Horticulture & Landscape Architecture, Colorado State University, Fort Collins, CO
3. Proteomics & Metabolomics Facility, Colorado State University, Fort Collins, CO

**Abstract:** In the U.S., the expectations for quality and unique organoleptic properties of hops (*Humulus lupulus)* is growing. The contribution of chemical compounds such as α-acids are well-understood, but it is unclear how chemical variation among different hop cultivars plays a role in sensory. This preliminary study characterized chemical variation in hops gathered from farms in both Yakima and Willamette Valley. The goals of this study are to determine if chemical variation related to *(i)* genetics, *(ii)* environment (*i.e.* different growing locations), *(iii*) and processing, is seen among hop cultivars, and *(iv)* how observed differences in chemical profiles impact the sensory attributes of the final hop product. For this preliminary study, a total of five hop samples (two different cultivars, growing locations, and processing conditions) were utilized. Non-targeted metabolomics was performed via Rapid Evaporative Ionization Mass Spectrometry (REIMS®) leading to the identification of distinct chemical composition for each hop sample. The analysis detected a total of 800 compounds that were evaluated for chemical variation using principal component analysis (PCA) and analysis of variance (ANOVA). Hop samples were evaluated for sensory attributes utilizing the American Society of Brewing Chemists sensory methods (see references) to ascertain if samples are different or similar at 0, 2 and 5 weeks. Work related to sensory will be discussed. Taken together, these data support that REIMS technology can be a tool to evaluate the chemical profile of hops and provide valuable information regarding hop metabolome, as it relates to the organoleptic properties of hops.

References:

HOP GRIND SENSORY EVALUATION METHOD. (2018). https://doi.org/10.1094/ASBCMOA-Sensory

TRIANGLE TEST (International Method). (2011). <https://doi.org/10.1094/ASBCMOA-Sensory-7>

Assessing haplotype variation and visualizing synteny in the diploid genome assembly of hop (*H. lupulus*)

Lillian Padgitt-Cobb, Sarah Kingan, Daniel Moore, Gregory Concepcion, Paul Peluso, David Rank, John Henning, and David Hendrix

Department of Biochemistry & Biophysics, Oregon State University

**Abstract:** Hop (*Humulus lupulus*) is a diploid, dioecious plant with a history of cultivation spanning thousands of years. Around the world, hop has been used in traditional medicine to treat a variety of ailments, including anxiety, insomnia, and pain. Modern research has revealed compounds of medicinal interest featuring anticancer, antimicrobial, and phytoestrogen properties. Large size (2.8Gb), heterozygosity, and abundant repeats have challenged draft assemblies of the hop genome. Using PacBio long-read sequencing and FALCON-Unzip for assembly, resolution of the diploid genome has become achievable. A more-complete, diploid assembly, in which both haplotypes are assembled separately, provides a foundation for identifying genes of interest involved in disease resistance and metabolism. The diploid assembly allows for studying variation between haplotypes, including rates of mutation, gaps, gap lengths, and selection. Exploring differences between protein-coding sequences of the haplotypes lends insight into the evolution, function, and selective pressures that have influenced the organism's history. Calculation of the non-synonymous to synonymous substitution rate ratio (dN/dS) provides a method to quantify selection within protein-coding regions, while Kimura distance provides an estimate of the rate of transitions (purine-purine or pyrimidine-pyrimidine) and transversions (purine-pyrimidine) that give rise to amino acid substitutions. We present novel approaches to analyze a diploid genome, including visualization of both haplotypes simultaneously on a synteny browser using GBrowse\_syn. These approaches serve to deepen our understanding of gene content and haplotype variation in hop.

Quantitative PCR assay development for field differentiation and dispersal patterns of hop powdery mildew.

Cameron Ross, David Gent

Oregon State Department of Botany and Plant Pathology, USDA-ARS, Corvallis, Oregon

**Abstract:** *Podosphaera macularis*, the causal agent of powdery mildew of hop (*Humulus lupulus*), causes severe economic damage annually across the Pacific Northwest through expensive fungicide applications, hop cone degradation, and quality defects. The fungusemerges as flag shoots focally located within hop yards in the spring and spreads throughout the season. Determining the late season dispersal patterns of powdery mildew between yards may improve management practices for this potentially devastating disease. Conclusive detection of *P. macularis* within a field through spore traps has proven difficult because both *P. macularis* and *P. clandestina,* powdery mildew of cherry,share sequence similarity in the internal transcribed spacer (ITS) region and therefore assays intending to differentiate these species have been unsuccessful.   In this project, the intergenic spacer region (IGS) will be used to develop an assay capable of detecting and differentiating *P. macularis* from other fungi.  Rotorod spore traps were placed adjacent to cv. Nugget yards across Oregon in order to collect conidia for quantifying patterns of dispersal. Ideally with the development of a quantitative PCR assay, differentiation and detection within fields can be confirmed with only femtograms of DNA. This could enable growers to better time fungicide applications and potentially avoid nonessential late season applications.

What weather factors drive early season release of overwintering spores of the hop powdery mildew pathogen.

Bill Weldon and David M. Gadoury

Section of Plant Pathology and Plant-Microbe Biology, Cornell University, Geneva, NY

**Abstract:** Disease management expenditures due to the hop powdery mildew pathogen *Podosphaera macularis* can top $500/hectare, yet the disease still causes appreciable direct yield loss each season. Satisfactory protection of hop cones is largely correlated with efficient early season disease management. The pathogen can overwinter and cause early season infection in two ways: (1) by living within semi-dormant buds as fungal mycelium that gives rise to shoots heavily coated with asexual spores (conidia) upon emergence in the spring, and (2) as chasmothecia; microscopic structures that discharge sexually produced spores (ascospores) during spring rains, infecting nearby newly emerged hop tissue. Since ascospores are formed through a sexual reproduction process, they have the potential to rapidly distribute advantageous fungicide and varietal resistance traits throughout hop powdery mildew populations where both mating types exist. The relative contribution to early season infection by the sexual phase of the pathogen, as well as the primary epidemiological factors that induce ascospore release are insufficiently understood. My research has focused on clarifying the early season threat posed by hop powdery mildew ascospores through modeling (1) the interaction between rain duration and temperature in promoting ascospore release, and (2) the seasonal maturation and release of *P. macularis* ascospore populations in Geneva, NY. These models are intended to provide hop growers with a functional tool to predict risk periods of ascospore release in the early growing season, which eventually may be incorporated into early season disease management decisions regarding more precise pruning or fungicide application timepoints.