

Application Of Metabolomics To Understand Barley Variety And Its Contribution To Beer Flavor

1: Department of Food Science & Human Nutrition, Colorado State University
 2: Horticulture & Landscape Architecture Facility, Colorado State University
 3: Proteomics & Metabolomics Facility, Colorado State University
 4: New Belgium Brewing Company, Inc., Fort Collins, CO
 5: Department of Biochemistry & Molecular Biology, Colorado State University

Harmonie M. Akers¹, Jacqueline M. Chaparro^{2,3}, Corey D. Broeckling^{2,3}, Dana Sedin⁴, Christian Holbrook⁴, Lindsay Guerdrum⁴, Jessica E. Prenni^{3,5}, Adam L. Heuberger²

1 Introduction

Background:

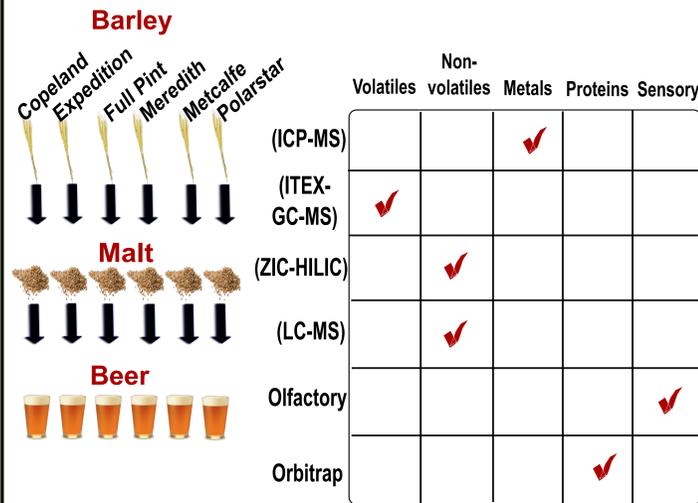
- Quality control is a vital component of a rapidly-growing beer industry.
- Identifying superior ingredients that provide distinct flavors (e.g. taste, aroma) is an important area of research.
- Brewers seek to identify raw ingredients to improve the stability of flavors during storage.

Rationale:

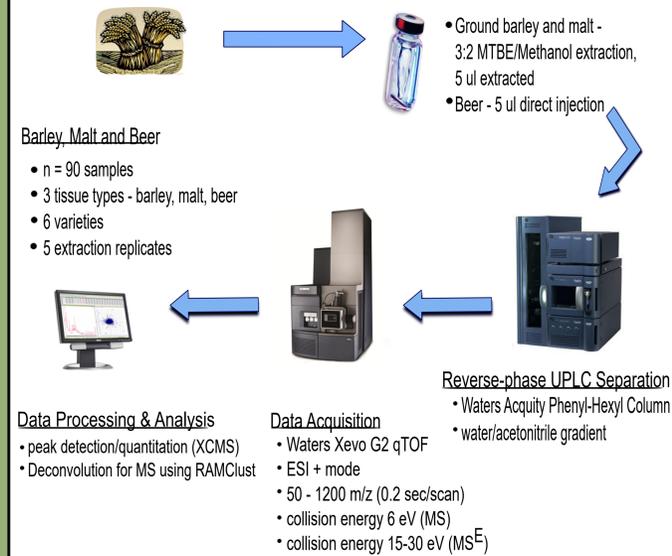
- It is currently unclear if different varieties of barley contribute to different flavors and flavor stability characteristics of the beer.
- Here, we used a metabolomics approach to characterize the chemical content of six barley grains and the corresponding malt and beer to determine if variation in the chemical content of the barley grain results in the differences in flavor and flavor stability in the beer.
- Flavor** is a sensation produced by the combination of aroma, taste, texture and temperature involving olfactory, gustatory, and tactile sense organs.
- Flavor stability** is the ability for a beer to maintain its true-to-brand flavor qualities after long-term storage.

2 Experimental Design

- An experiment was designed to profile metabolites and sensory traits for six varieties of barley used to produce six different malts and beers.

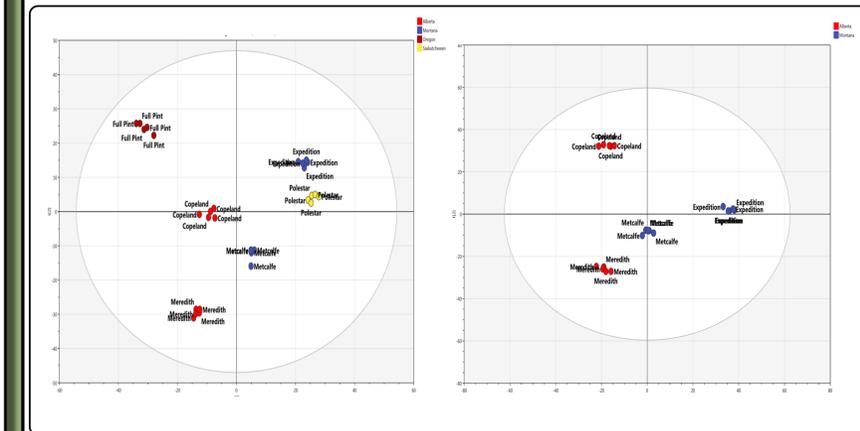


3 Non-targeted RP-UPLC-MS Metabolomics Workflow



4 Contribution of Barley to Non-Volatile Metabolites in Beer (RP-UPLC-MS)

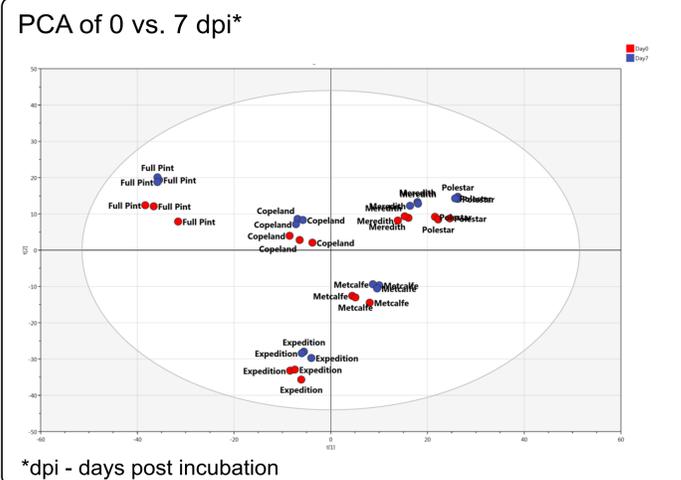
- UPLC-MS analysis detected over 1,650 molecular features in beer.
- PCA resulted in 5 PCs that explained 75% of variation.
- Principal component analysis (PCA) showed chemical variation in beer was influenced by barley variety (labels), and not maltster (colors).



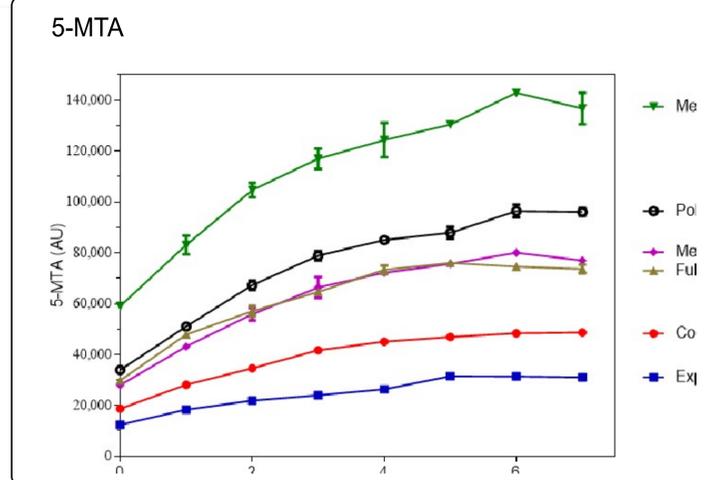
These data support the hypothesis that different barley varieties contribute to the non-volatile metabolite fingerprint of beer, and this may affect the flavor profile.

5 Contribution of Barley to Stability of Non-Volatile Metabolites in Beer

- An experiment was designed to test the flavor stability of the 6 beers in an accelerated assay.



- PCA shows variation in metabolite profile due to high temperature at 7 days post incubation (dpi).
- Accelerated aging resulted in increased 5-MTA (5-methylthioadenosine), a marker for stability.

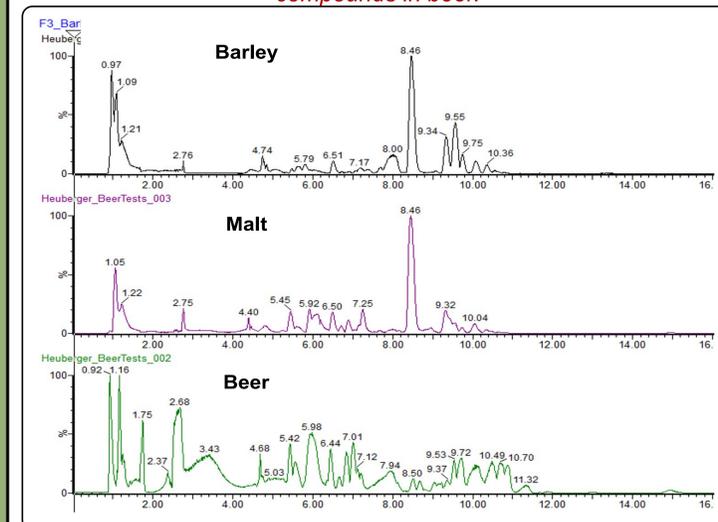


- Trends:**
- Beer differed in initial amounts of 5-MTA, and 5-MTA increased at different rates.

These data support that barley variety influences the chemical stability of beer, and this may affect the flavor profile after storage.

6 Detection of Polar Compounds Using ZIC-HILIC-UPLC-MS

- Zwitterionic Hydrophilic Interaction Chromatography (ZIC-HILIC) detects polar compounds and can be a valuable method to detect carbohydrates, amino acids, and organic acids in beer.
 - 3 ul of barley, malt extract or beer was injected into a Waters Acquity UPLC coupled to a Waters Xevo G2 ESI-TOF MS acquired in negative MS^E mode
 - Mobile phase: Acetonitrile (Solvent A); 10mM Ammonium bicarbonate (Solvent B). Column: Merck EMD Millipore ZIC-pHILIC (5 ul polymer) 150 x 2 mm
- These data support ZIC-HILIC as a robust method to detect polar compounds in beer.*



7 Conclusions

- Barley variety contributes to the non-volatile metabolite profile of beer.
- Barley variety contributes to several non-volatile metabolites associated with flavor stability.

Future Directions

- Determine if barley variety contributes to profiles of volatile and non-volatile polar metabolites.
- Quantification of elements (e.g. iron, copper, sulfur) using ICP-MS ionomics.
- Assess barley and malt metabolite associations to beer quality using standard metrics, such as appearance, aroma, flavor, and storage
- Integrate data with sensory on the beer to determine effect on flavor.
- Investigation of 5-MTA contribution to flavor and flavor stability.

Acknowledgements:

This work was supported by the Brewers Association and New Belgium Brewing Company

References:

Heuberger, Adam L., Broeckling, Corey D., Lewis, Matthew R., Salazar, Lauren, Bouckaert, Peter, Prenni, Jessica E. "Metabolomic profiling of beer reveals effect of temperature on non-volatile small molecules during short-term storage." Food Chemistry, Vol. 135, Issue 3, (2012): 1284-1289.

Prenni, Jessica, Adam Heuberger, and Dana Sedin. "Nontargeted Metabolite Profiling by UPLC-MS: A Case Study in Beer Reveals Effect of Temperature on Nonvolatile Small Molecules During Storage." LC GC NORTH AMERICA 31.4 (2013): 306-+.