

4-26-2019

Comparison of Hop Derived Humulone Constituents in Beer Using UV-Vis, HPLC, and LC-MS

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Recommended Citation

Boland, Aaron I.; Viriyasiri, Nicholas; Espinosa, Lorna; Campbell, Hunter; and Driesner, Kurt, "Comparison of Hop Derived Humulone Constituents in Beer Using UV-Vis, HPLC, and LC-MS" (2019). *Undergraduate Research Symposium*. 22.
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Introduction

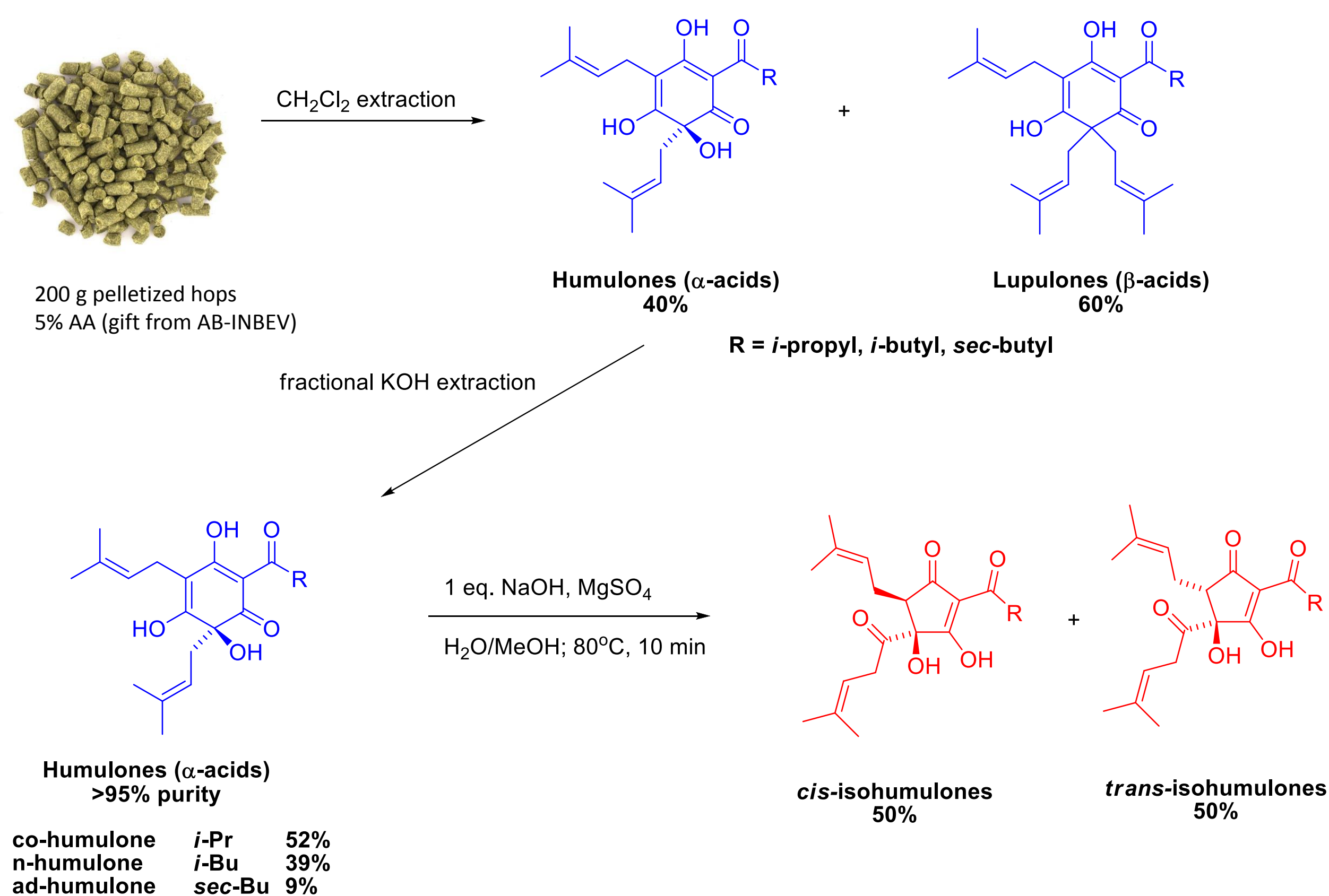
Humulones, or α -acids (AA), are significant components of the hop cones or strobile representing between 2-20% of the dried weight. During beer brewing, AA are extracted from the hops added during boiling and converted to the isomeric isohumulones, or α -isoacids (IAA). These IAAs contribute to beer bitterness and provide a balance to the malt flavors from the grains. Beer bitterness is often described as IBU (International Bittering Units) which is based on the UV absorption IAA at 275nm. Historically this approach worked well for traditional lagers and lightly hopped beers. However, with the advent of late hopping and dry hop routines, modern craft beers often contain significant amounts of AA and oxidized derivatives which also absorb in the UV region. As a result, the measurement of IBU by UV absorption can be significantly obscured by these additional, hop derived components. In this experiment, the use of UV spectroscopy, HPLC and selected ion LCMS methods are compared from profiling the concentrations of hop derived constituents and determining contributions to beer bitterness.

Methods

Beer samples were obtained from CHEM 1021: Beer Brewing – Chemical & Biochemical Principles. Beers were analyzed by the industry standard method, 'Beer-23', for determination of IBU by UV spectroscopy. Extracts obtained by this method were analyzed by HPLC and LCMS. For comparison, beer samples were also analyzed chromatographically by direct injection.

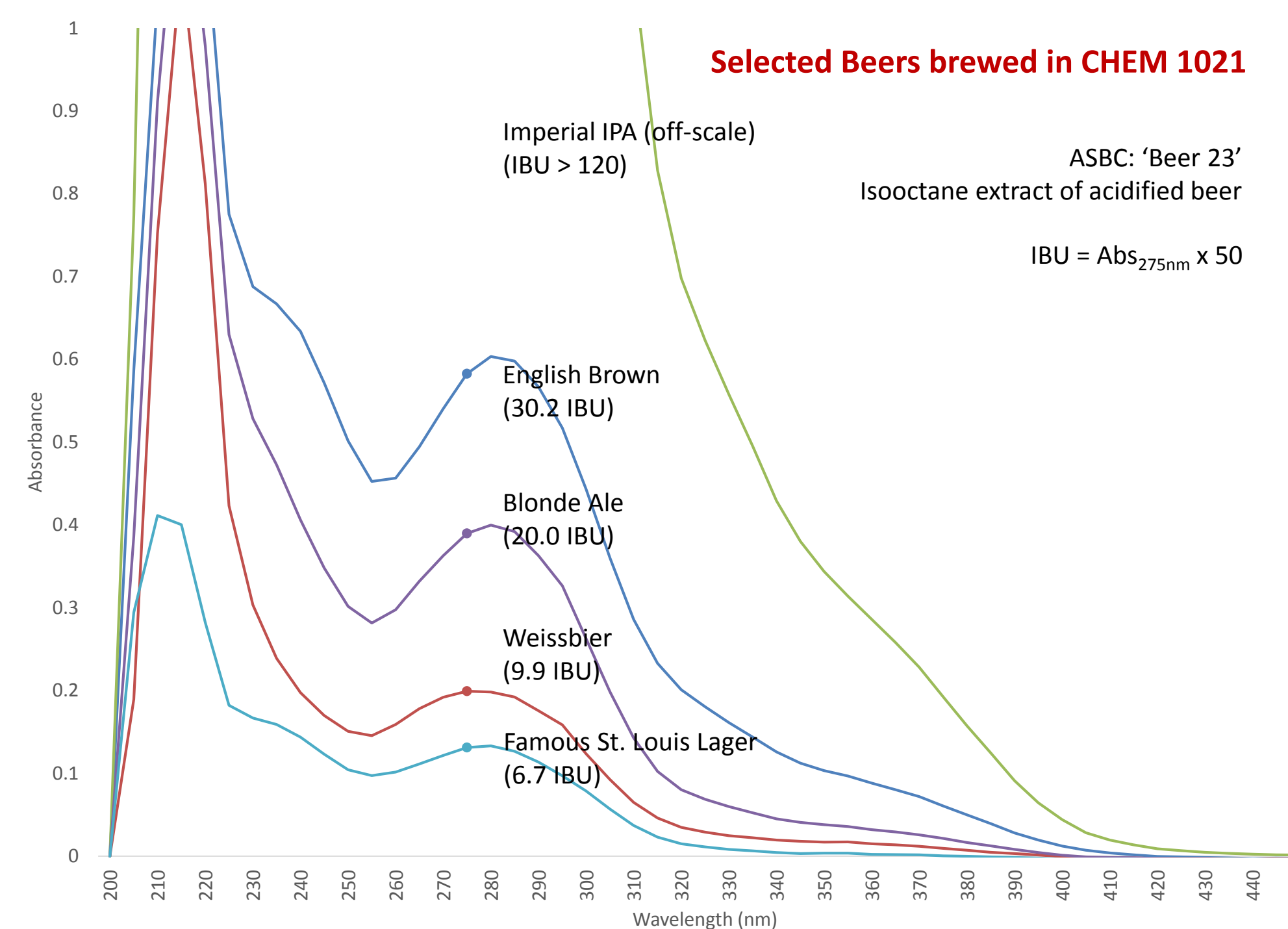
Results

Isolation of Standards



Results (cont'd.)

Spectroscopic Determination of Bitterness

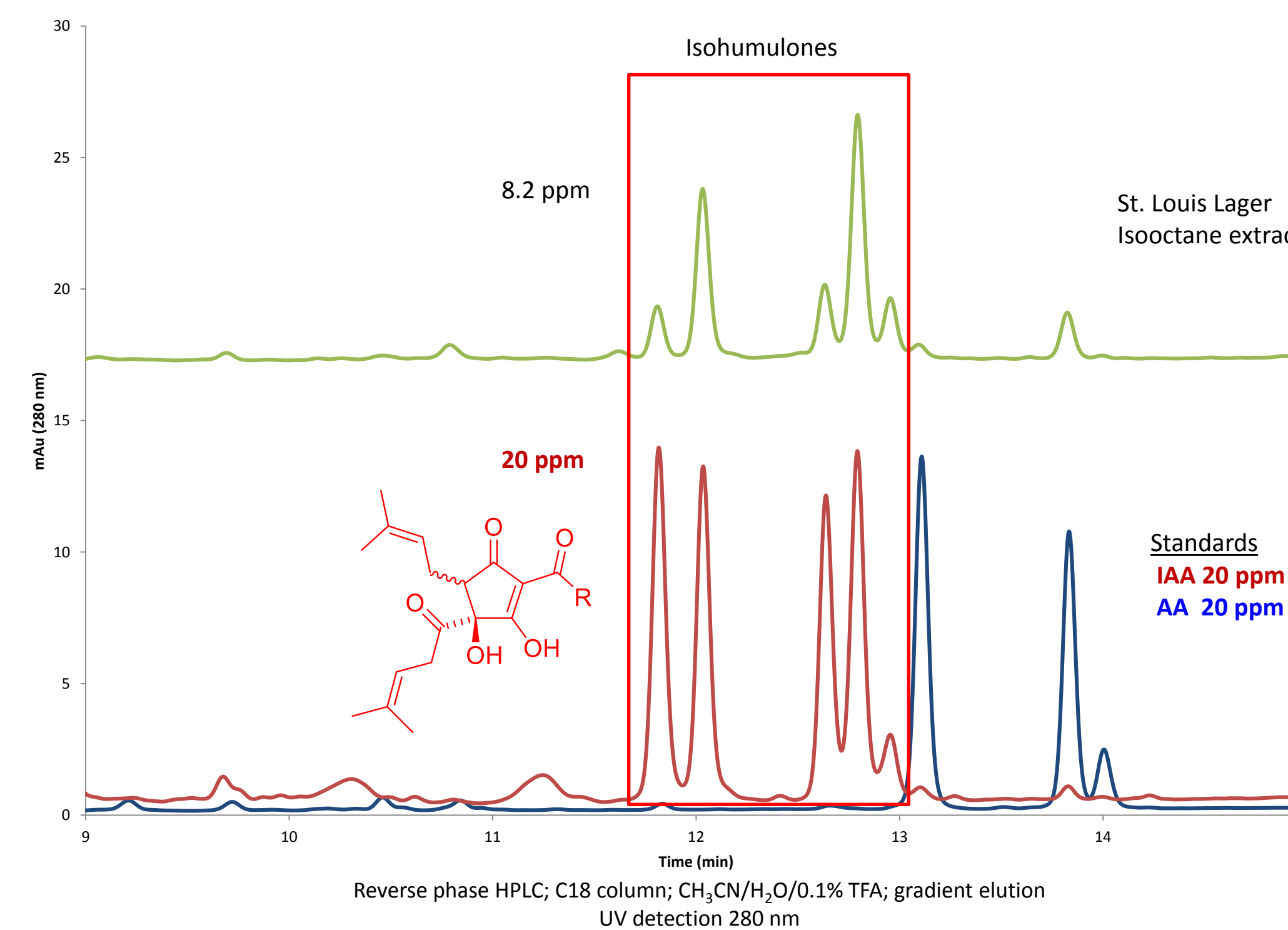


Comparison of IBU and PPM

Beer	Calc IBU	IBU (UV)	IAA ppm (HPLC)	AA ppm (HPLC)
Imperial IPA	56	120	15	24
English Brown	31	30	8	9
Blonde Ale	15	20	14	3
Weissbier	10	10	8	9
St. Louis Lager	12*	7	8	0

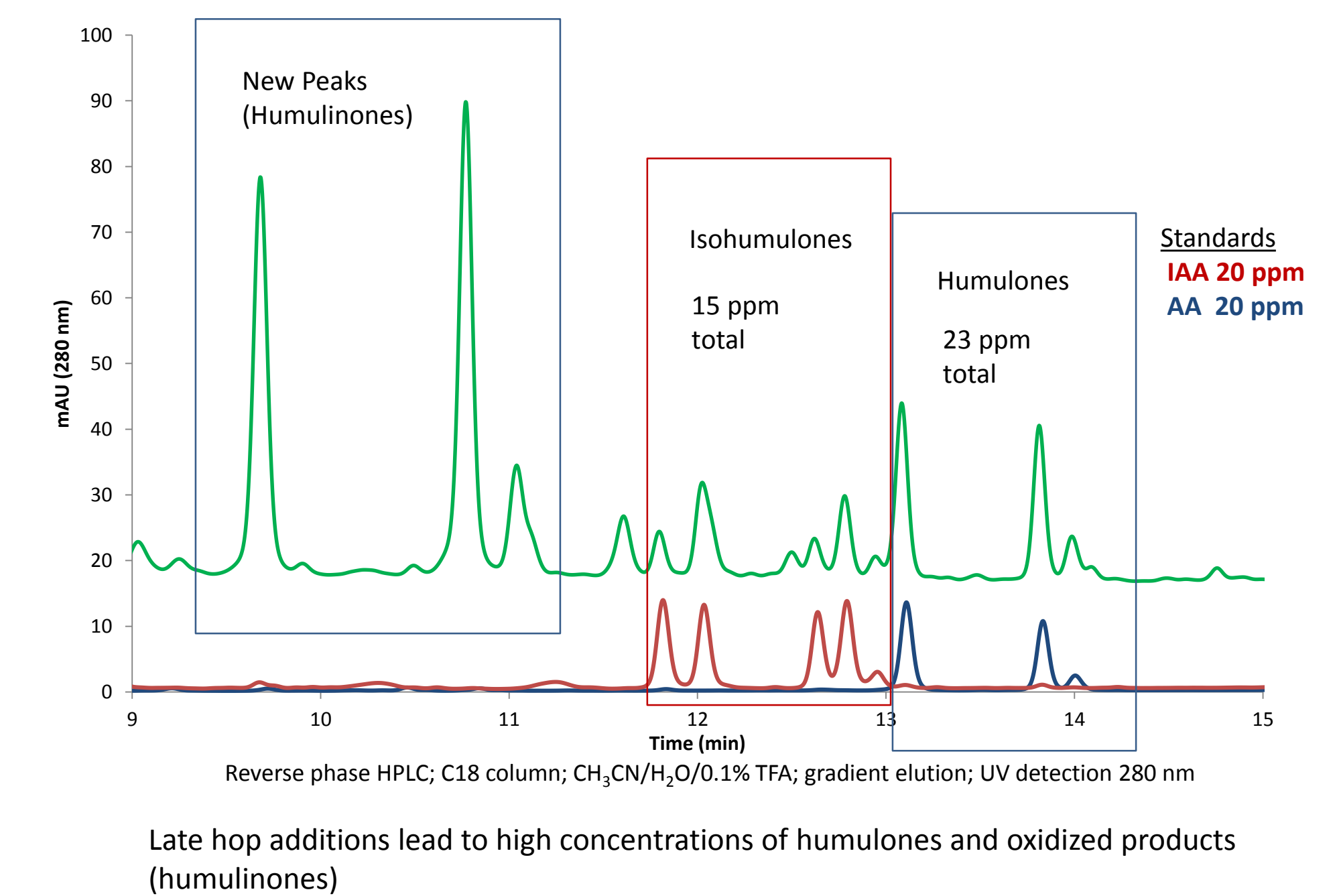
* Value obtained online: <https://untappd.com/b/anheuser-busch-budweiser/3783/variants>

HPLC – Famous St. Louis Lager

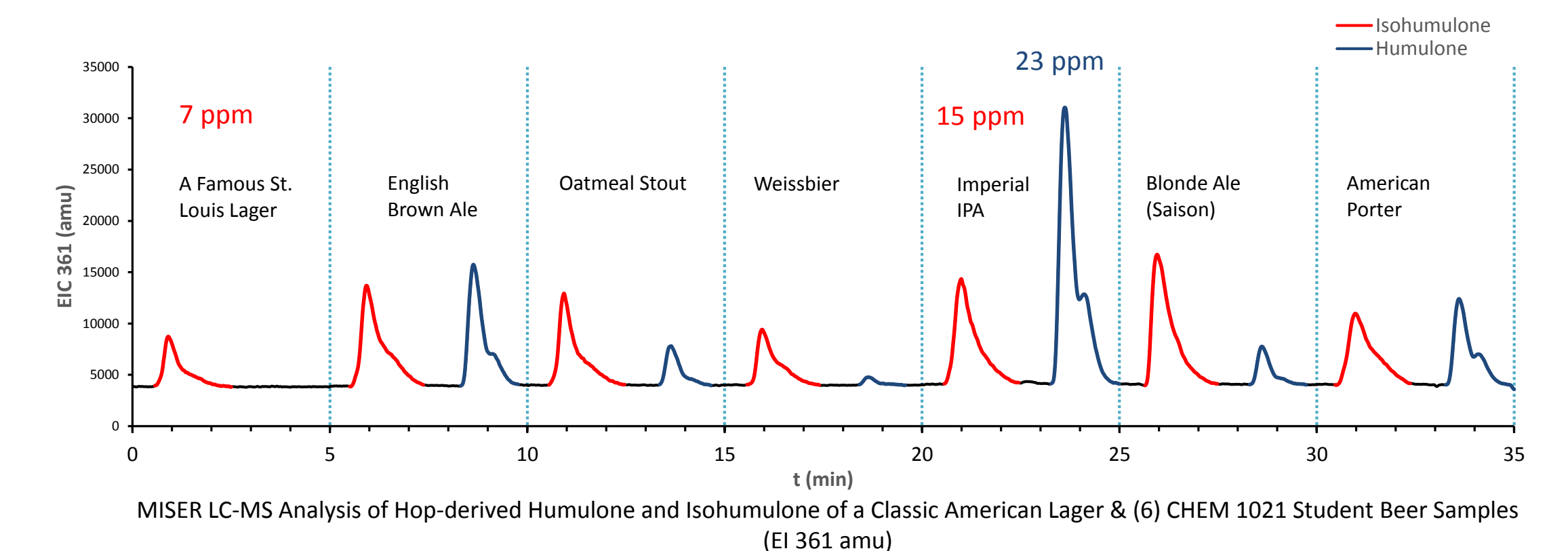


Results (cont'd.)

HPLC – Imperial IPA



Isohumulone / Humulone Comparison of CHEM 1021 Beer Samples



Discussion and Conclusions

- The 'Beer-23' UV spectroscopic method for measurement of IBU is limited in scope to traditional lager beer styles.
- Hop additions late in the boil or post-boil result in significant deviation from IBU or concentration of IAA as determined by HPLC, extracted ion LCMS or sensory profiling.
- Knowing possible interferences is important to brewers from a reproducibility perspective and also in the growing marketplace demand for product transparency.

References:

Hamper, B. C.; Zawatzky, K.; Zhang, V.; Welch, C. J.; *Rapid Determination of Humulones and Isohumulones in Beers Using MISER LC-MS Analysis*. J. Am. Soc. Brew. Chem. 75(4), 2017, 333-338.

Peacock, V. "The International Bitterness Unit (IBU), its creation and what it measures. In: *Hops Flavor and Aroma, Proceedings of the 1st International Brewers Symposium*. Aug. 9, 2007.

American Society of Brewing Chemists. *Methods of Analysis*, 14th ed. Beer-23 Beer bitterness; American Society of Brewing Chemists, Saint Paul, MN.