

# 73rd Annual Meeting of the American Society of Brewing Chemists

*Part of Brewing Summit 2010*

**June 15–18, 2010**  
**Rhode Island Convention Center**  
**Providence, Rhode Island, U.S.A.**



**The Science of Beer**



AMERICAN SOCIETY OF  
Brewing Chemists



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# Catch the Buzz in Providence



Courtesy of the Providence Warwick CVB.

As we were reading about Providence, RI, we were struck by three things that make it special and also similar to the American Society of Brewing Chemists. First, Providence is a small, compact city; second, it's been nicknamed the "Beehive of Industry"; and, third, the city has recently rebranded itself as the "Creativity Capital" to emphasize its educational resources and arts community.

So, Providence seems to be the perfect venue for the 73rd Annual Meeting of the American Society of Brewing Chemists! Like Providence, ASBC is a small, close-knit community; so, too, is the ASBC meeting a beehive of activity, and ASBC has worked diligently during 2010 to communicate the message that ASBC embodies the "Science of Beer."

As in years past, the ASBC Program Committee has worked tirelessly to deliver an annual meeting program that provides value to our membership, promotes the "Science of Beer," and offers ample opportunities for discussion and networking. All this takes place in a casual, open format. For those of you who attended the 2009 ASBC Annual Meeting in Tucson, you may remember that several changes were made to the program format to adapt to the challenges in the brewing industry and the economy at that time. Because our members responded with such enthusiasm about many of the changes, we've taken the best of these and incorporated them into this year's annual meeting. We've also added a twist—the Brewing Summit, in which the Friday programming and the exhibition are shared with MBAA.

Needless to say, this year's ASBC Annual Meeting and Brewing Summit offer a wealth of opportunities to increase your brewing science knowledge. On Tuesday, there are three exciting pre-meeting short courses to start things off in good fashion! On Wednesday, Graham Stewart sets the tone of the meeting with a keynote address entitled "*Using Brewing Science to Make Good Beer Great.*" This is followed by 3 days filled to the brim with 11 technical sessions, 9 workshops, 18 technical subcommittee meetings, 3 poster and exhibit sessions, the hugely popular Pearls of Wisdom debate session, Beer and Food Pairing Recognition Luncheon, and What's the Buzz? open forum at the close of the day on Friday. All of this is like the city of Providence: compact, active, and creative!

On behalf of the ASBC Program Committee, we are pleased to have you join our community of brewing scientists for a few days of camaraderie, discussion, debate, and learning about the plethora of topics related to brewing science. To our perennial attendees, we welcome you to our reunion. To our international colleagues and friends, we thank you for making the journey. To new attendees, we welcome you to the Society with the hope that you feel the sense of community and catch the buzz generated at our annual meeting! We encourage all of you to contribute to the program by using every opportunity to network with new colleagues and old friends, share your expertise, and expand your technical knowledge by learning something new. By doing so, we all contribute to brewing science and to making good beer great!

Cindy-Lou Bell  
Jeff Cornell  
Program Committee Co-chairs

# ASBC Schedule at a Glance

Sessions and events all take place at the Rhode Island Convention Center unless otherwise indicated.

Tuesday, June 15			
8:00 a.m. – 5:00 p.m.	ASBC Board of Directors Meeting • <i>Westin: Executive Board Room</i>	<b>Short Course:</b> The Audacity of Hops * • 550 A/B	<b>Short Course:</b> Brewing Boot Camp * • 552 A/B
10:00 – 11:30 a.m.	ISIHAS Hop Subcommittee Meeting • 553 A/B		
1:00 – 5:00 p.m.	<b>Short Course:</b> Lessons Learned in Yeast Management * • 553 A/B		
6:30 – 7:00 p.m.	ASBC Meeting Orientation • 553 A/B		
Wednesday, June 16			
8:00 – 9:30 a.m.	Opening General Session and Keynote: Using Brewing Science to Make Good Beer Great • 555/556		
9:30 – 10:15 a.m.	Emerging Issues Forum • 555/556		
10:30 a.m. – 12:00 p.m.	<b>Technical Session:</b> Sensory • 555/556	<b>Technical Session:</b> Malt & Barley I • 551 A/B	
11:00 a.m. – 12:00 p.m.	Technical Subcommittees • <i>See program for details</i>		
12:00 – 1:45 p.m.	Food and Beer Pairing Recognition Lunch • <i>1st Floor Lobby</i>		
2:00 – 4:05 p.m.	<b>Workshop:</b> Beer and Wine Crossover • 552 A/B	<b>Technical Session:</b> Malt & Barley II • 551 A/B	
2:00 – 4:30 p.m.	<b>Technical Session:</b> Stability & Quality • 555/556		
4:15 – 5:30 p.m.	<b>Workshop:</b> Sensory: Understanding What Influences Beer Flavor * • 552 A/B	<b>Technical Session:</b> Yeast • 551 A/B	
5:45 – 6:30 p.m.	<b>Pearls of Wisdom:</b> Industry-Sponsored University Research Is Charity • 555/556		
7:00 – 9:30 p.m.	ASBC Welcome Reception • <i>Rotunda</i>		
Thursday, June 17			
8:00 – 10:05 a.m.	<b>Technical Session:</b> Flavor • 555/556		
8:30 – 10:00	<b>Workshop:</b> Barley to Malt to Beer: Context and Content • 552 A/B		
10:15 – 11:30 a.m.	<b>Technical Session:</b> Fermentation • 555/556	<b>Workshop:</b> Is Good Foam Just In the Eye of the Beholder? * • 552 A/B	
11:30 a.m. – 2:00 p.m.	Exhibits and Lunch • <i>Ballroom/Exhibit Area</i>		
11:30 a.m. – 2:00 p.m.	ASBC Posters ( <i>authors present 1:00 – 2:00 p.m.</i> ) • <i>West Pre-function</i>		
1:00 – 2:00 p.m.	Technical Subcommittees • <i>See program for details</i>		
2:15 – 4:00 p.m.	<b>Technical Session:</b> Health • 555/556	<b>Workshop:</b> An Inside Look at Craft Distilling * • 554 A/B	
4:15 – 6:00 p.m.	<b>Technical Session:</b> Hops • 555/556	<b>Workshop:</b> Rapid Methods in Brewing Microbiology • 552 A/B	
6:00 – 6:30 p.m.	MBAA Convention Orientation • 554 A/B		
Friday, June 18: Shared Day of Programming with MBAA			
8:30 – 9:30 a.m.	ASBC Technical Subcommittee Meeting – New and Alternate Methods of Analysis • 550 A		
8:30 – 10:00 a.m.	Exhibits • <i>Ballroom/Exhibit Area</i>		
8:30 – 10:00 a.m.	ASBC and MBAA Poster Viewing • <i>West &amp; East Pre-functions</i>		
10:15 – 11:30 a.m.	<b>ASBC-MBAA Workshop:</b> In-line/On-line Measurement • 555/556	<b>ASBC Technical Session:</b> Packaging • 551 A/B	<b>MBAA Technical Session:</b> Nutrition & Enzymes • 552 A/B
11:30 a.m. – 12:30 p.m.	ASBC Technical Subcommittees • <i>See program for details</i>		
11:30 a.m. – 2:00 p.m.	Exhibits and Lunch • <i>Ballroom/Exhibit Area</i>		
11:30 a.m. – 2:00 p.m.	ASBC and MBAA Posters ( <i>authors present 1:00 – 2:00 p.m.</i> ) • <i>West &amp; East Pre-functions</i>		
2:15 – 3:30 p.m.	<b>ASBC-MBAA Workshop:</b> Critical Quality Review: Culture, Communications, and Customers • 555/556	<b>ASBC Technical Session:</b> Innovation • 551 A/B	<b>MBAA Technical Session:</b> Yeast • 552 A/B
3:45 – 5:00 p.m.	<b>ASBC Closing Session:</b> What's the Buzz? • 555/556	<b>MBAA Workshop:</b> Practical Malt Quality • 551 A/B	<b>MBAA Technical Session:</b> Stability • 552 A/B
7:00 – 10:00 p.m.	Brewing Summit Social • <i>Offsite: Squantum Association</i>		
10:00 p.m.	After Glow Party • <i>Rotunda</i>		

\* Additional registration or ticket is required.

# ASBC Program

## Tuesday, June 15

8:00 a.m. – 5:00 p.m.	Board of Directors Meeting and Lunch	Westin: Executive Board Room
8:00 a.m. – 5:00 p.m.	The Audacity of Hops *	550 A/B
8:00 a.m. – 5:00 p.m.	Brewing Boot Camp I *	552 A/B
10:00 – 11:30 a.m.	ISIHAS Hop Subcommittee Meeting	553 A/B
1:00 – 5:00 p.m.	Lessons Learned in Yeast Management *	553 A/B
3:30 – 6:45 p.m.	Registration	5th Level Lobby
4:00 – 11:00 p.m.	Hospitality	Westin: Waterplace Board Room
6:30 – 7:00 p.m.	ASBC Meeting Orientation	553 A/B

\* Additional registration is required

## Tuesday Highlights

### The Audacity of Hops

*Charlie Bamforth, University of California, Davis; Brian Buffin, Kalsec, Inc.; David Grinnell, Boston Brewing Co.; Tim Kostelecky, Barth-Haas Group; Tom Shellhammer, Oregon State University; Mark Schulze, Kalsec*

This course entails a wide overview of hops including a review of their botany, horticulture, and the history of hops and use in brewing. Detailed presentations will be given on the various hop varieties, functionalities in beer, hop products, and applications, as well as strategies and methods to optimize hop efficiency and effectiveness in brewing and beer design. Hands-on demonstrations profiling various hop products and hop flavors in beer will be included.

### Brewing Boot Camp I

*Greg Casey, MillerCoors; Joe Dirksen, Ecolab; Mark Eurich, MillerCoors; Mary Jane Maurice, Malteurop North America; Dave Thomas, Ecolab; Sue Thompson, MillerCoors; Rob Tod, Allagash Brewing Co.*

This full-day course is designed for the employee new to brewing science or new to brewing in general. Attendees will come away from this course understanding why brewers gauge various parameters. Brewing science experts will define the what/why/how of quality and quality measures and will describe various methods used by brewers large and small that will demystify the science of brewing. Course topics include:

- Quality Building Blocks—Fundamentals of Quality at a Brewery
- Grains and Wort Construction—Good In, Great Out
- Cellar and Yeast Management—Keep It Alive and Happy
- CIP—Not Just for the End of the Day/Shift or Beer Style; It's an Always Proposition
- Sensory Assignment—A Matter of Taste
- Creative Brewing—The Process of Making Beers Unlike Anyone Else

### Lessons Learned in Yeast Management

*Greg Casey, MillerCoors and Graham G. Stewart, GGStewart Associates Panel Members: Joe Casey, Craft Brewers Alliance; Steven Pauwels, Boulevard Brewing Co.; Chris Powell, University of Nottingham; Derek Stepanski, St. Louis Brewery*

This discussion of yeast management includes a panel of individuals from small to large breweries wanting to share their experiences and expertise in this area. During this course, participants will have the opportunity to ask questions about yeast and yeast management as well as be provided with information and tools to help troubleshoot common yeast management issues. Greg Casey will also share some of his experience and knowledge through his cause-and-effect fishbone diagrams.

### Meeting Orientation

Grab a bottle of beer, meet other attendees, and learn what you can do at the ASBC Annual Meeting. Plan to attend if this is your first ASBC Annual Meeting or if you have not attended in the last few years.

## Wednesday, June 16

7:00 – 8:00 a.m.	Speaker Orientation and Breakfast: All presenters	Rotunda
7:30 a.m. – 5:00 p.m.	Registration	5th Level Lobby
7:30 a.m. – 5:00 p.m.	Silent Auction	5th Level Lobby
8:00 – 9:30 a.m.	Opening General Session and Keynote	555/556
9:30 – 10:15 a.m.	Emerging Issues Forum	555/556
10:30 a.m. – 12:00 p.m.	Technical Session: Sensory <i>Moderator: Sue Thompson, MillerCoors</i>	555/556
10:30 a.m.	O-1. Martina Gastl. Influence of matrix effects on body and mouthfeel of non-alcoholic beverages.	
10:55 a.m.	O-2. Thomas H. Shellhammer. Beer composition influences bitterness perception of isomerized hop acids.	
11:20 a.m.	O-3. Femke L. Sterckx. The influence of volatile monophenols on the flavor of Belgian beers.	
10:30 a.m. – 12:00 p.m.	Technical Session: Malt & Barley I <i>Moderator: Scott Heisel, American Malting Barley Assn.</i>	551 A/B
10:30 a.m.	O-4. Henrik Andren. High-speed single-seed sorting of malting barley, based on chemical composition, for producing a premium malt and a premium beer.	
10:55 a.m.	O-5. Stefan Kreis. Barley specifications for brewing technology based on unmalted barley.	
11:20 a.m.	O-6. Takashi Iimure. Development of DNA markers for the selection of beer foam stability in barley breeding.	
11:00 a.m. – 12:00 p.m.	Technical Subcommittee Meetings	
	• Total Carbohydrates by HPLC	550 A
	• IBU of Dry Hopped Beers	550 B
	• MOA Wort Review	553 A
	• SPME Fingerprint and GC-FID Analysis for Beer Volatiles	553 B
12:00 – 1:45 p.m.	Food and Beer Pairing Recognition Lunch	1st Floor Lobby
2:00 – 4:00 p.m.	Workshop: Beer and Wine Crossover	552 A/B
2:00 – 4:05 p.m.	Technical Session: Malt & Barley II <i>Moderator: Michael Davis, American Malting Barley Assn.</i>	551 A/B
2:00 p.m.	O-7. Yueshu Li. Study of the effects of pre-harvest sprouting on the storability and malting quality of three Canadian malting barley varieties.	
2:25 p.m.	O-8. David J. Cook. The time-course of color and flavor formation during malt roasting processes.	
2:50 p.m.	O-9. Mandeep Kaur. Microbial community structure changes during the malting of Australian barley.	
3:15 p.m.	O-10. Michael J. Edney. Using germination index and homogeneity to predict malt processing and quality.	
3:40 p.m.	O-11. Evan Evans. Impact of mashing-in temperature on extract and fermentability and the level of wort $\beta$ -glucan, soluble protein, free amino nitrogen (FAN), and lipids.	
2:00 – 4:30 p.m.	Technical Session: Stability & Quality <i>Moderator: Aaron Porter, Sierra Nevada Brewing Co.</i>	555/556
2:00 p.m.	O-12. Roland Folz. Consistently meeting the specifications—The rising importance of defined quality control for craft breweries.	
2:25 p.m.	O-13. Karl J. Siebert. Modeling beer foam behavior.	
2:50 p.m.	O-14. Frank-Jürgen Methner. The influence of specific Maillard reaction products in malt on oxidative beer stability—Pro- and antioxidative effects.	
3:15 p.m.	O-15. Michael Voetz. PYF analyses of commercial malt samples—Assessment of recent results.	
3:40 p.m.	O-16. Peter J. Rogers. Will the key anti-oxidant protein in beer please stand up.	
4:05 p.m.	O-17. Daan Saison. Improved flavor stability by aging beer in the presence of yeast.	
2:00 – 5:00 p.m.	ASBC Poster Set Up	West Pre-function
2:00 – 6:00 p.m.	Exhibit Set Up	Ballroom/Exhibit Area

4:00 – 11:00 p.m.	Hospitality (closed 7:00 – 9:30 p.m. during Welcome Reception)	Westin: Waterplace Ballroom
4:15 – 5:30 p.m.	Technical Session: Yeast <i>Moderator: Chris Powell, University of Nottingham</i>	551 A/B
4:15 p.m.	O-18. Hiroyuki Yoshimoto. Monitoring yeast physiological state during fermentation by quantitative cell morphogenesis analysis.	
4:40 p.m.	O-19. Katherine A. Smart. Key flocculation performance indicators during production-scale lager brewing fermentations.	
5:05 p.m.	O-20. Christopher A. Boulton. An exploration of the relationships between yeast physiology, brewery propagation, and fermentation performance.	
4:15 – 5:30 p.m.	Workshop: Sensory: Understanding What Influences Beer Flavor*	552 A/B
5:45 – 6:30 p.m.	Pearls of Wisdom	555/556
7:00 – 9:30 p.m.	ASBC Welcome Reception	Rotunda

## Wednesday Highlights

### Opening General Session and Keynote

Using Brewing Science to Make Good Beer Great  
*Graham G Stewart, GGStewart Associates*

Brewing is one of the oldest biotechnologies. Beer has always been considered to be a high-quality alcoholic beverage with an enhanced reputation compared to other alcoholic beverages. However, a focus on the science and technology of brewing has made good beer great! Our dependence on scientific and technical enhancement has resulted in greater process efficiency and reproducibility together with improved beer quality, integrity, stability, and product diversity. Allied with this tradition is a dependence on brewing education and training, ensuring a well-trained workforce and widespread dissemination of results and opinions that is a model for other industries.

### Emerging Issues Forum

*Michael Brophy, Brewing Malting Barley Research Institute; Rebecca Newman, Consultant; Dana Sedin, MillerCoors; Fred Strachan, Sierra Nevada Brewing Co.*

*Moderator: Rebecca Newman, Consultant*

Join us for questions and concerns related to emerging issues of brewers. This year's forum will focus on all sizes of brewing operations as well as "radical brewing" companies. Ingredients are redefining beer and beer styles and are no longer as simple as malted barley, hops, and yeast. We will also take a look at what is happening in North America and Europe in regards to BPA inclusion affecting brewers using cans and plastic bottles.

### Technical Subcommittee Meetings

Each meeting is specific to a Technical Subcommittee run from 2009 to 2010 and will provide an overview of the results and recommendations. The meetings are open to all meeting attendees, and your feedback and participation in these meetings are essential to ensuring the quality of the methods being tested or reviewed.

### Food & Beer Pairing Recognition Lunch

Back by popular demand, our Food and Beer Pairing Recognition Lunch was created especially for ASBC. We start with the beer and the chef takes over from there. He uses the dominant flavors and characteristics of each beer and pairs it up with food stations that will provide a complementary and distinct dining experience.

\* Additional registration or ticket required

### Beer and Wine Crossover

*Jeff Biegert, New Belgium Brewing Co.; Greg Casey, MillerCoors; Annette Fritsch, Annette Fritsch Consulting; and Peter Rogers, Fosters (retired)*

*Moderator: Steven Pauwels, Boulevard Brewing Co.*

A panel of brewers and winemakers, varied and practical, compare and contrast the processes for production of beer and wine. Speakers will address the methods of measure, process flow, and regulatory issues that define each as unique, and they will then blur lines of distinction. The workshop will be moderated by brewmaster and winemaker Steven Pauwels of Boulevard Brewing. At the end of the session, author and professor Charlie Bamforth will be on hand to autograph copies of his book *Grape vs. Grain*. Save 10% when you buy a copy of the book at the meeting!

### Sensory: Understanding What Influences Beer Flavor\*

*Cathy Haddock, Sierra Nevada Brewing Co.; Francis Meehan, International Flavors & Fragrances; and Neva Parker, White Labs, Inc.*

*Moderator: Sue Thompson, MillerCoors*

Give your taste buds a workout during this two-hour sensory session. The four segments that will be addressed are beer basics, aging (in tank and in wood), bugs, and flavorants. This workshop will also provide you with an overview on your taste buds: what they are, how they work, and why different beers taste...well...different. You will also have the opportunity to go extreme with bugs! Space is limited.

### Pearls of Wisdom: Industry-Sponsored University Research Is Charity!

*Peter Rogers, Fosters (retired) and Tom Shellhammer, Oregon State University*

*Moderator: John Engel, MillerCoors*

Experience a point/counterpoint style debate at the Pearls of Wisdom, where controversial topics, outrageous points of view, and audience participation are guaranteed! Peter Rogers, recently retired Fosters Group Ltd., and Tom Shellhammer, Oregon State University, will square off to bring light and levity to what has occurred in research from the university bench to the brewer's bench. Then the floor is open for all to participate.

### Welcome Reception

Kick off the meeting with friends and colleagues! Come hungry and you will not be disappointed. The event will have a wonderful array of food from appetizers all the way through to dessert! And what would an ASBC event be without good beer to round out the evening.

## Thursday, June 17

7:30 a.m. – 6:00 p.m.	Registration	5th Level Lobby
7:30 a.m. – 6:00 p.m.	Silent Auction	5th Level Lobby
7:30 – 9:00 a.m.	Exhibit Set Up	Ballroom/Exhibit Area
8:00 – 9:00 a.m.	Poster Set Up	West Pre-function
8:00 – 10:05 a.m.	Technical Session: Flavor <i>Moderator: Sylvie Van Zandycke, Lallemand, Inc.</i>	555/556
8:00 a.m.	O-21. Takeshi Arai. Analysis of flavor compounds by yeasts in insufficient nutrition II—Studies on brewing yeast indole production and the tryptophan pathway.	
8:25 a.m.	O-22. Mustapha Nedjma. Improving beer flavor and fermentative capacity with selected hybrids <i>S. cerevisiae</i> and interspecific <i>S. uvarum</i> × <i>S. cerevisiae</i> produced on specific maltose medium.	
8:50 a.m.	O-23. Udo Kattein. About the different behavior of miscellaneous wort aroma compounds during wort boiling—Concentrations in kettle-full and –finished worts and condensed vapors.	
9:15 a.m.	O-24. Norihiko Kageyama. Studies for specific control of astringent substances in malt to improve beer aftertaste.	
9:40 a.m.	O-25. Leif A. Garbe. Rapid determination of the reactive (off) flavors 4,5-epoxy-2E-decenals in beer.	
8:30 – 10:00 a.m.	Workshop: Barley to Malt to Beer: Context and Content	552 A/B
10:15 – 11:30 a.m.	Technical Session: Fermentation <i>Moderator: Thomas Shellhammer, Oregon State University</i>	555/556
10:15 a.m.	O-26. Yuichi Nakamura. Impact of wort aeration period of multi-filling cylindroconical vessels.	
10:40 a.m.	O-27. Pieter J. Verbelen. The importance of wort composition in yeast metabolism during accelerated fermentations.	
11:05 a.m.	O-28. Graeme M. Walker. Impact of zinc on brewing yeast fermentation performance and beer quality.	
10:15 a.m. – 12:00 p.m.	Workshop: Is Good Foam Just In the Eye of the Beholder?*	552 A/B
11:30 a.m. – 2:00 p.m.	Exhibits and Lunch	Ballroom/Exhibit Area
11:30 a.m. – 2:00 p.m.	ASBC Poster Viewing (authors present 1:00 – 2:00 p.m.)	West Pre-function
1:00 – 2:00 p.m.	Technical Subcommittee Meetings	
	• Sensory	550 A
	• Malt 8 Protein, Malt 4 Extract, EMAST Standard	553 A
	• Miniature Fermentation Assay, ATP, and MOA Review Micro	553 B
2:15 – 4:00 p.m.	Technical Session: Health <i>Moderator: David Maradyn, Novozymes North America, Inc.</i>	555/556
2:15 p.m.	O-30. Moeko Ozaki. Tetra-hydro-iso- $\alpha$ -acid (THIAA), a reduced compound derived from hop bitter acid, contributes to the prevention of osteoporosis.	
2:40 p.m.	O-29. Hiroshi Hirata. Xanthohumol, the main prenylflavonoid in hops, inhibits cholesteryl ester transfer protein activity and improves dyslipidemia.	
3:05 p.m.	O-31. Moritz Krahl. The glycemic index of beverages—A critical review.	
3:30 p.m.	O-32. Charles W. Bamforth. Antioxidants in beer: Better than those from wine?	
2:15 – 4:00 p.m.	Workshop: An Inside Look at Craft Distilling*	554 A/B
4:00 – 11:00 p.m.	Hospitality	Westin: Waterplace Ballroom
4:15 – 6:00 p.m.	Technical Session: Hops <i>Moderator: Tim Kostecky, John I Haas, Inc.</i>	555/556
4:15 p.m.	O-33. Paul S. Hughes. Kinetic modeling of hop acid isomerization: Insight for the enhancement of bitterness yields during wort boiling.	
4:40 p.m.	O-34. Patrick L. Ting. Resolution and properties of isomerized ( $\pm$ )-tetrahydrocohumulone.	
5:05 p.m.	O-35. Stefan Hanke. Influence of hopping technology on harmony of bitterness.	
5:30 p.m.	O-36. Sebastian Kappler. Influence of hop pre-treatment before dosage on the yield of isohumulones and resulting beer quality.	
4:15 – 6:00 p.m.	Workshop: Rapid Methods in Brewing Microbiology	552 A/B
6:00 – 6:30 p.m.	MBAA Convention Orientation	554 A/B

\* Additional registration or ticket required

## Thursday Highlights

### Barley to Malt to Beer: Context and Content

*Michael Brophy, Brewing Malting Barley Research Institute; Mary-Jane Maurice, Malteurop North America, Inc.  
Moderator: Mary-Jane Maurice, Malteurop North America, Inc.*

Barley availability has a profound effect on the economics and operation of breweries. Current supply and demand for North American barley will be presented, as well as topics concerning barley storability and the potential for non-malt barley beers. We promise there will be plenty of opportunities to bring our ideas together for a robust discussion!

### Is Good Foam Just In the Eye of the Beholder?\*

*Charlie Bamforth, University of California, Davis; Florian Kuplent, Anheuser-Busch InBev; Evan Evans, University of Tasmania; Katie McGivney, New Belgium Brewing Co.  
Moderator: Rebecca Newman, Consultant*

The workshop will commence with a discussion of the theory of what makes good foam and how this can be measured reliably for quality control. The theory will be extended into practical options that brewers have for optimizing the foam head on their established brands and factors to consider when developing new beer brands. The theory will subsequently be put into practice by the guided pouring of a series of beers into glasses that have been designed to accentuate the noble qualities of these beers. The workshop will conclude with a panel discussion. Space is limited.

### Technical Subcommittee Meetings

Each meeting is specific to a Technical Subcommittee run from 2009 to 2010 and will provide an overview of the results and recommendations. The meetings are open to all meeting attendees, and your feedback and participation in these meetings are essential to ensuring the quality of the methods being tested or reviewed.

### An Inside Look at Craft Distilling\*

*Steve Wright, Spiritech Solutions*

Take a look at the growth of small distillers in the United States. We will discuss who is distilling, where they are located, and what they are producing. The presentation will then be turned over to Rhode Island distillers to share their stories; offer their personal perspectives; and discuss their successes and challenges, including funding, profitability, distribution, and sales. Attendees will have a chance to enjoy samples. Space is limited. *This workshop requires preregistration along with a minimal fee for materials.*

### Rapid Methods in Brewing Microbiology

*Moderator: Kelly Tretter, New Belgium Brewing Co.*

Faster is better! Don't we all wish we could have our micro results yesterday? Well, we may not be able to achieve yesterday, but there are faster tests available than what you are currently using. Discover rapid methods for every budget, from what to use for a start-up brewery to flow cytometry, fluorescent microscopy, and PCR.

*\* Additional registration or ticket required*



**Blake Layfield**  
North Carolina State University  
2009 Student Travel Award Recipient

## Your Bid Makes a Difference ASBC Foundation Silent Auction

*Silent Auction proceeds help student presenters attend the annual meeting.*

### Hours:

Wednesday, June 16	7:30 a.m. – 5:00 p.m.
Thursday, June 17	7:30 a.m. – 6:00 p.m.
Friday, June 18	8:00 a.m. – 1:30 p.m.

### Location:

Outside of the ballroom

**We'll See You at the Auction!**



## Friday, June 18—Shared Day of Programming with MBAA

8:00 – 9:30 a.m.	ASBC Program Committee Meeting and Breakfast	550 B
8:00 a.m. – 1:30 p.m.	Silent Auction	5th Level Lobby
8:00 a.m. – 4:00 p.m.	Registration	5th Level Lobby
8:30 – 9:30 a.m.	ASBC Technical Subcommittee Meeting – New and Alternate Methods of Analysis	550 A
8:30 – 10:00 a.m.	Exhibits	Ballroom/Exhibit Area
8:30 – 10:00 a.m.	ASBC and MBAA Poster Viewing	West & East Pre-functions
10:15 – 11:30 a.m.	ASBC Technical Session: Packaging <i>Moderator: Kathy Kinton, MillerCoors</i>	551 A/B
10:15 a.m.	O-37. Roland Folz. Microbiological QA—Classification perplexity with modern packaging.	
10:40 a.m.	O-38. Lorinda (Lori) Yoder. Technologies, tools, and challenges for packaging beer in PET.	
11:05 a.m.	O-39. Johann Angres. Beverage and package quality—Two inseparable key parameters in the modern quality control of bottled beverages.	
10:15 – 11:30 a.m.	MBAA Technical Session: Nutrition & Enzymes <i>Moderator: Jens Voigt, Technical Univ Munich Weihenstephan</i> <i>Abstracts found in the MBAA section.</i>	552 A/B
10:15 a.m.	O-1 Moritz Krahl. Innovative concepts for the production of non-alcoholic malt-based beverages.	
10:40 a.m.	O-2 Charles Bamforth. Mindfulness: What happened?	
11:05 a.m.	O-3 Elisabeth Steiner. A comparison of beer quality attributes between 100% barley malt and barley adjunct beer, with a focus on changes in the protein composition.	
10:15 – 11:30 a.m.	ASBC–MBAA Workshop: In-line/On-line Measurement	555/556
11:30 a.m. – 12:30 p.m.	ASBC Technical Subcommittees	
	• Malt 2 B Sortimat, Malt 13 DON, and Malt 7 Alpha Amylase	551 A/B
	• Craft Brewers	553 B
11:30 a.m. – 1:15 p.m.	ASBC Publications Committee Meeting and Lunch	550 B
11:30 a.m. – 2:00 p.m.	Exhibits and Lunch	Ballroom/Exhibit Area
11:30 a.m. – 2:00 p.m.	ASBC and MBAA Poster Viewing (authors present 1:00 – 2:00 p.m.)	West & East Pre-functions
2:15 – 3:30 p.m.	ASBC Technical Session: Innovation <i>Moderator: Fred Strachan, Sierra Nevada Brewing Co.</i>	551 A/B
2:15 p.m.	O-40. Zhumao Jiang. A novel approach to brew alcohol-free beer.	
2:40 p.m.	O-41. Annika Wilhelmson. Ingredients and energy from brewer's spent grain.	
3:05 p.m.	O-42. Zhumao Jiang. Recycling and refining of alcohol from waste beer.	
2:15 – 3:30 p.m.	MBAA Technical Session: Yeast <i>Moderator: Roland Folz, VLB Berlin</i> <i>Abstracts found in the MBAA section.</i>	552 A/B
2:15 p.m.	O-4 Chris Powell. Profiling a lager fermentation completed using active dried yeast.	
2:40 p.m.	O-5 Michael Bradley. Yeast activity monitoring.	
3:05 p.m.	O-6 Eric Samp. Possible roles of the mitochondria in sulfur dioxide production by lager yeast.	
2:15 – 3:30 p.m.	ASBC–MBAA Workshop: Critical Quality Review: Culture, Communications, and Customers	555/556
3:45 – 5:00 p.m.	ASBC Closing Session: What's the Buzz?	555/556
3:45 – 5:00 p.m.	MBAA Technical Session: Stability <i>Moderator: Daniel Carey, New Glarus Brewing Co.</i> <i>Abstracts found in the MBAA section.</i>	552 A/B
3:45 p.m.	O-7 Alastair Pringle. A fresh look at beer flavor stability.	
4:10 p.m.	O-8 Thomas Kunz. The influence of unmalted barley on the oxidative stability of wort and the final beer.	
4:35 p.m.	O-9 Karl Siebert. The role of polyphenols in beer haze and astringency.	
3:45 – 5:00 p.m.	MBAA Workshop: Practical Malt Quality	551 A/B
4:00 – 7:00 p.m.	Hospitality/Bierstube	Westin: Waterplace Ballroom
4:45 – 5:30 p.m.	ASBC Poster Take Down	West Pre-function
7:00 – 10:00 p.m.	Brewing Summit Social	Offsite: Squantum Assoc.
10:00 p.m.	After Glow Party	Rotunda
10:00 – 11:30 p.m.	Hospitality/Bierstube	Westin: Waterplace Ballroom

## Friday Highlights

### **In-line/On-line Measurement**

*Jeff DeVoy, Heuft U.S.A., Inc.; Patrick Mazzeo, Hach Co.; Al Worley, optek-Danulat, Inc.*

*Moderators: Jeff Cornell, MillerCoors; Horace Cunningham, Terrapin Beer Co.*

Speakers will discuss in-line measurement technologies across a wide range of brewing and packaging applications. The focus will be placed on the theory and principles of the various technologies as well as the advantages and potential pitfalls in real-world applications. Attendees should come away with a good sense of the measurement science and how these various technologies are best applied to process measurements, enabling improved quality and/or throughput. Specific topics include applied photometry for brewing applications, in-line package inspection, and applied technologies for dissolved oxygen measurement. The workshop will conclude with questions from the audience and an interactive discussion.

### **Technical Subcommittee Meetings**

Each meeting is specific to a Technical Subcommittee run from 2009 to 2010 and will provide an overview of the results and recommendations. The meetings are open to all meeting attendees, and your feedback and participation in these meetings are essential to ensuring the quality of the methods being tested or reviewed.

### **Critical Quality Review: Culture, Communications, and Customers**

*Stu Oliver, MillerCoors; Dan Carey, New Glarus Brewing Co.; Jason Perkins, Allagash Brewing Co.; Paul Pettinger, New Belgium Brewing Co.*

*Moderators: Rebecca Newman, Consultant; Mary Pellettieri, MillerCoors*

This workshop targets expanding breweries that are seeking to understand and grow the maturity of their quality efforts. Representatives from micro-breweries to macro-breweries will be present to speak about the quality journey. The objective is to provide context and direction around creating culture of quality that fosters internal and external customer relationships.

### **ASBC Closing Session: What's the Buzz?**

The Closing Session is an excellent capstone to the ASBC Annual Meeting. This interactive session will provide you with a recap of the entire annual meeting. The floor will then be opened for you to voice your thoughts about ASBC and discuss your experiences from the past three days. This session was new in 2009 and the feedback was outstanding. Make plans to join us for a great end-of-the-meeting synopsis.

### **Practical Malt Quality**

*Nigel Davies, Muntons PLC; Bob Hansen, Briess Malt*  
*Moderator: Susan Welch, Malteurop North America Inc.*

This workshop will include a panel of international brewers and maltsters who will present overviews and lead discussions on topics related to malt color and beer, such as: a comparison of how brewers and maltsters view color as a quality parameter, how malt formula is determined from beer color and how beer color is predicted by malt formula, variability in base malt color, variability in the analysis of base malt color, and a brewer's perspective on color specifications and beer.

### **Brewing Summit Social**

Join your colleagues as ASBC brings their annual meeting to a close and MBAA kicks off their convention. The social will take place at Squantum Association where we will mix and mingle in the historic Main Club House and the Bakehouse that is built out over the rocky coastline. Attendees will also enjoy the beautiful manicured gardens and dramatic views of the Providence River and Narragansett Bay. A wide selection of appetizers, dinner fare, desserts, and drinks, along with the ambiance of the waterfront and the historic surroundings, will make the evening complete. Shuttle service will be available from the Westin to Squantum Association. Shuttles to the social will run from the Westin Providence from 6:00 to 7:00 p.m. and return service will be available from 9:00 to 10:30 p.m.

### **After Glow Party**

After the Brewing Summit Social, join us for a relaxing night of Irish coffee, cocktails, and networking. The After Glow Party is sponsored by Malteurop North America, Inc.

# WBC

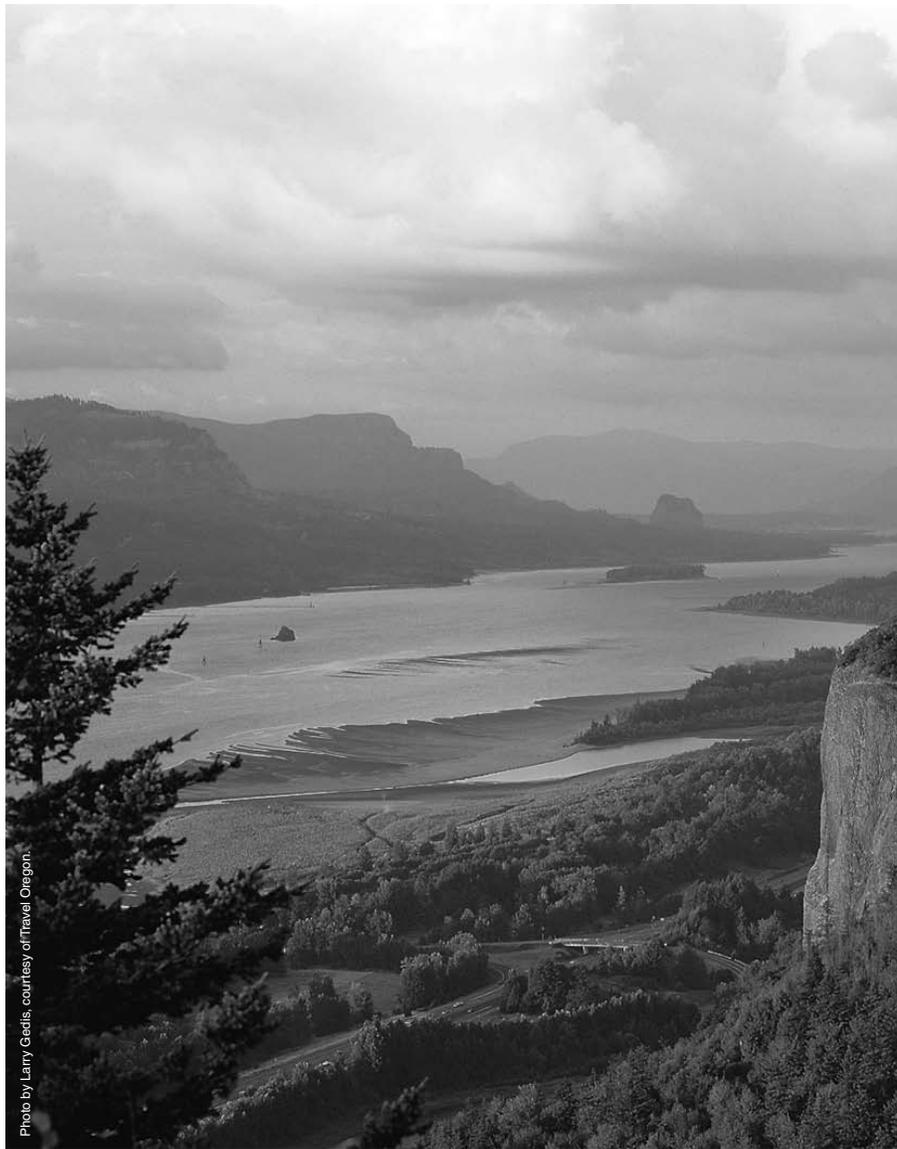


Photo by Larry Geddis, courtesy of Travel Oregon.

## Save the Date

July 28–August 1  
Oregon Convention Center  
Portland, OR, U.S.A.

# 2012

[www.worldbrewingcongress.org](http://www.worldbrewingcongress.org)

**Hosted by:**

American Society of Brewing Chemists  
Master Brewers Association of the Americas

**With active participation by:**

Brewery Convention of Japan  
European Brewery Convention  
Institute of Brewing & Distilling



# ASBC Annual Meeting Abstracts

## O-1

### **Influence of matrix effects on body and mouthfeel of non-alcoholic beverages**

MARTINA GASTL (1), Anke Kiesslich (1), Alexander Quadt (2), Joachim Tretzel (1)

(1) Technische Universität München; (2) Döhler GmbH

Next to olfactory and gustatory effects, the importance of somatosensory impressions in the oral cavity has gained lots of interest in recent years. Nevertheless little is known about the characteristics and perception of body and mouthfeel. Therefore the exact description and differentiation of these sensations is necessary. According to the International Organization for Standardization, mouthfeel comprises tactile sensations perceived at the lining of the mouth, including the tongue, gums, and teeth (ISO 5492:1990). The first step was to characterize the term mouthfeel. In the course of this specification, a taste panel (expert panel) was built up. During the training, 20 descriptors with appendant references and precise descriptions were defined. These results formed the background for further estimations and enabled the panel to evaluate a high number of different non-alcoholic beverages. The trained panel then was deployed to evaluate the term body. In particular, the target was to identify the qualities that influence perception in a positive or negative direction. Under the assumption that body can be considered as a part of mouthfeel, 14 descriptors were defined. The created body and mouthfeel wheel was presented at 2008 WBC in Honolulu, HI (poster 188) and at the 2009 EBC Congress in Hamburg, Germany. In this project, correlations (matrix effects) between different beverage ingredients (e.g., sugar, sweetener content, acidity) were worked out systematically to describe their effect on body and mouthfeel. Following 32 samples, which varied in sugar or rather sweetener content, acidity and malt-extract level were analyzed using the conventional method of profiling and examined by different statistical methods. By means of correlation and principal component analyses the data were linked to each other to define dependencies. This revealed, for example, that viscosity is not the only important property affecting the perception of body. There exist further influencing factors that have a nearly equal effect on body and mouthfeel perception. In conclusion, this study exemplifies how body and mouthfeel can be described, evaluated, and examined. Furthermore the results demonstrate the descriptors and properties or beverage ingredients that need to be influenced to optimize the body and mouthfeel characteristics of non-alcoholic beverages.

*Martina Gastl apprenticed as a brewer and maltster from 1994 to 1996 in Klosterbrauerei Andechs, Germany. She studied brewing and beverage technology at the Technische Universität München-Weihenstephan, Germany. She graduated as an engineer in 2002. From 2002 to 2006 she completed her Ph.D. degree on the "Technological Influence on Lipid Degradation in Terms of Improvement of Beer Flavour Stability." After graduation in 2002 she worked as a scientific employee and head of the GC/HPLC laboratory at Lehrstuhl für Technologie der Brauerei I (TU München-Weihenstephan) for two years, following the head of the malt laboratory. She is currently assistant professor and head of the raw material research group at the Lehrstuhl für Brau- und Getränketechnologie in Weihenstephan. Since 2008 she has been working on her postdoctoral lecture qualification; her research interest involves "Characterization and Interaction of Flavour Active Taste Compounds in Cereal based Beverages Influencing Beverage Harmony."*

## O-2

### **Beer composition influences bitterness perception of isomerized hop acids**

THOMAS H. SHELLHAMMER (1), Jeffrey E. Clawson (1), Timothy J. Kostecky (2)

(1) Oregon State University, Corvallis, OR; (2) John I. Haas, Yakima, WA

The relative bitterness of iso-alpha-acids (Iso) and tetrahydro-iso-alpha-acids (Tetra) is a topic of considerable debate. Comparisons of these two isomerized hop acids have found Iso to be equibitter to Tetra in some studies, while in others Tetra has been found to be more bitter than Iso. Factors leading to this discrepancy could likely be beer medium differences. In this study, we investigated how residual extract (RE), ethanol concentration, acidity, and residual sweetness influenced the perception of bitterness intensity and quality in a lager beer medium. In one trial, residual extract and ethanol were examined together by preparing a pale-malt wort using exogenous enzymes for low RE (1.3% w/w) and a high-temperature mashing regime for high RE (5.7% w/w). These worts were fermented using lager yeast, and the resultant beers had a base ethanol level of 3% w/w. For the high ethanol (6% w/w) treatment, food-grade ethanol was added. Separately, Iso or Tetra was added to the four beer treatments, the beers were filtered, and the final concentrations were adjusted to 15 mg/L of Iso or Tetra. In a second trial, wort was prepared, resulting in a more typical beer (3.5% w/w RE, 3.3% w/w ethanol, 4.0 pH) to which lactic acid was added to achieve a low pH treatment (pH 3.5) and/or maltose was added to achieve a high sweetness treatment (1.5% w/w). Again, Iso and Tetra were dosed individually to reach 15 mg/L. Twelve trained bitterness panelists evaluated all eight treatments in each of the two experiments using a randomized block format. In the presence of high ethanol and high RE, Tetra and Iso were similar in bitterness intensities; however, in the presence of low extract and regardless of the ethanol level, Tetra was significantly more bitter than Iso. In the presence of acid, Tetra was significantly more bitter than Iso, with or without added sweetness. Increased sweetness resulted in decreased bitterness for all treatments. Qualitatively, the presence of the higher level of ethanol or acid resulted in increased medicinal-like attributes. In contrast, the presence of sweetness subdued these attributes and resulted in greater similarity between the two hop acids. Differences in residual extract, ethanol, sweetness, and acidity, common variable components in beer, had a significant effect on the perception of bitterness for Iso versus Tetra.

*Thomas Shellhammer is the Nor'Wester Professor of Fermentation Science and associate professor in the Department of Food Science and Technology at Oregon State University, where he leads the brewing science education and research programs. His brewing research examines the role of hops in beer quality. Thomas received his Ph.D. degree in food engineering from the University of California, Davis, in 1996 and worked as assistant professor of food science at The Ohio State University for four years prior to his move to Oregon in 2001. During the 2008–2009 academic year he worked at the Technische Universität Berlin and the Versuchs- und Lehranstalt für Brauerei (VLB) as a Fulbright Senior Scholar and Alexander von Humboldt Research Fellow.*

## O-3

### **The influence of volatile monophenols on the flavor of Belgian beers**

FEMKE L. STERCKX (1), Daan Saison (1), Freddy R. Delvaux (1)

(1) Centre for Malting and Brewing Science, Leuven, Belgium

Beer flavor is the result of a complex interaction between hundreds of chemical compounds and even more taste and olfactory receptors. Although much research has been published on different aspects of beer

flavor, there are still many aspects to be unraveled. One such aspect is the influence of volatile monophenols on beer flavor. In wines and spirits many monophenols, such as vanillin, eugenol, syringaldehyde, guaiacol, 4-ethylphenol, and 4-vinylguaiacol, are reported as important flavor compounds, often held responsible for certain spicy, smoky, vanilla-like flavors or phenolic off-flavors. For beer, the focus has mainly been on 4-vinylguaiacol, a phenol contributing to the overall flavor of numerous beers and, when present in high concentrations, causing off-flavors. Yet, several other monophenols have been identified in various Belgian beers, and their impact on beer flavor is not known, although certain phenolic, spicy, vanilla-like notes are ascribed to these Belgian beers during sensory evaluation. To elucidate the influence of monophenols on beer flavor, we determined the individual and mixture flavor thresholds of these monophenols in beer, and we compared analytical data on the monophenol content of Belgian beers with sensory evaluation of these beers. Since the individual sensitivity of tasters for these compounds varied strongly, we assessed group thresholds, as well as frequency distributions among the taste panels. Individual thresholds give a good indication of the flavor impact of particular compounds, but it would be too simplified to consider beer flavor as the sum of contributions made by each individual compound. Several combinations of monophenols showed strong synergistic effects, which indicates that interactions between monophenols are likely to influence the overall effect of monophenols on beer flavor. By quantifying monophenols in various Belgian beer styles and comparing these results with sensory evaluation we could form a tentative image of how the monophenols influence the flavor of the studied beer samples. Finally, to validate these results, we added mixtures of monophenols to lager beer at concentrations at which they occur in the Belgian specialty beers, and these simulations were compared sensorially to the original beers. Despite the complexity of flavor interactions, this study confirms the influence of monophenols on the overall flavor of numerous beers and its importance.

*Femke Sterckx was born in 1984 in Herentals, Belgium. In 2007, she graduated with an M.S. degree in applied biological sciences and engineering from K.U. Leuven, Belgium. She carried out her master's thesis on the influence of *Saccharomyces cerevisiae* and *Brettanomyces custersii* on glycosidically bound flavor compounds in hops and sour cherries at the Centre for Malting and Brewing Science at K.U. Leuven. After graduation she started a Ph.D. program at the Centre for Malting and Brewing Science. Her work is focused on the identification of flavor-active monophenols in beer and their influence on beer flavor. For this research, IWT Vlaanderen grants her financial support.*

#### O-4

##### **High-speed single-seed sorting of malting barley, based on chemical composition, for producing a premium malt and a premium beer**

HENRIK ANDREN (1), Bo Lofqvist (1)  
(1) BoMill, Lund, Sweden

There is a huge variation in quality from one single kernel to the other, even in the most homogenous populations of malting barley that presently are available to maltsters. This variation can be registered for almost any quality parameter and certainly has a large impact on the performance of the grain in the malt house and/or the malt in the brewhouse. A new, unique sorting technology—TriQ sorting technology—for sorting of single seeds according to their quality at a speed of 20,000–200,000 kernels per second has made it possible not only to produce more homogenous lots of malting barley than has ever been seen for micro-scale as well as full-scale malting production, but also offers sorters with a capacity of 2–20 t/hr to the industry. The purpose of this paper is to examine the impact of improved homogeneity on malting and the quality of malt. Special emphasis will be given to the effects of improved homogeneity with regard to protein content in the malting barley, yield of extract, and  $\beta$ -glucan in the wort, as well as haze (EBC) and *Fusarium*.

*Henrik Andren has been working for almost 25 years within the grain-related business, with responsibility for the grain business within the FOSS group. Recently Henrik became the managing director for BoMill AB. BoMill has developed a unique high-speed sorting machine that can sort individual malting barley kernels based on their composition.*

#### O-5

##### **Barley specifications for brewing technology based on unmalted barley**

STEFAN KREISZ (1), Niels Elvig (1), Hans-Peter Heldt-Hansen (1)  
(1) Novozymes A/S, Copenhagen, Denmark

The full or partial replacement of malt as brewing raw material is becoming more and more standard in brewing recipes worldwide. One possible replacement option for malt is unmalted barley. Barley has the advantage of providing the same basic composition as malt, and if hydrolyzed with a suitable enzyme blend, it can deliver nearly the same wort quality as malt. Barley supplies not only fermentable sugars like other starch sources, but in combination with exogenous enzymes it also delivers sufficient free amino nitrogen and comparable aroma and taste profiles. These properties place barley in its own category as a brewing raw material, and therefore, the specifications, as well as the breeding goals, established for malting barley have to be challenged. Reviewing common malting barley specifications, the food safety requirements, as well as the general involvement of quality control of the supply chain, will stay unchanged, whereas some quality parameters designed for malting properties like cytolytic abilities, e.g., friability or beta-glucan content, are not relevant. This opens up the specifications for unused barley sources like naked barley and allows brewers to utilize local raw materials in regions where barley is planted but malting is not established. In 2008 and 2009 we analyzed over 80 barley samples from all over the world with a “modified Congress mash” system. The barley was milled with a disc mill (0.2 mm gap) and mashed adding a newly developed enzyme system (Ondea Pro) with a 2 kg/t dosage. The mashing profile was adapted using an infusion mashing regime with rests at 54°C, 64°C, and 80°C. We measured lauter performance, wort turbidity, extract, FAN, sugar and amino acid profile, viscosity, and ADF. All analyses were made according to *Analytica EBC* or *MEBAK*. Twelve barley samples that showed major differences during the first trials were chosen for intensified research, including beta-amylase and limit dextrinase activity, gelatinization temperature, and germination capacity measurement. Finally, to simulate lauter tun grist composition, analyses with coarse grist (0.8 mm gap) were added. The results give a unique overview of the contribution of unmalted barley to wort quality. The barley range from two-row European spring barley to local barley from India or Russia shows the full variation in barley qualities used for brewing today. The statistical evaluation allows suggestions to be made concerning brewing barley specifications for brews with high or full replacement of malt by barley. Furthermore it shows that breeding programs designed for unmalted brewing barley may focus more on yield, disease resistance, and extract than on malting properties and, therefore, opens the range of suitable brewing barleys.

*Stefan Kreis� studied brewing and beverage technology at the Technischen Universität München-Weihenstephan, Germany (1991–1997). He graduated with an engineering degree in 1997. From 1997 until 2002 he completed his doctoral thesis, concerning the filterability of wort and beer, at the Institute for Brewing Technology I in Weihenstephan. From 2000 until 2002 he worked as a scientific employee and assistant at the malt laboratory at the Institute for Brewing Technology I. From 2002 until 2007 he was an assistant professor and head of the malt laboratory. His main research interest has been cereals and malting technology and beer filtration. He also has worked as a consultant for malt houses and breweries and has presented several papers at ASBC meetings and EBC congresses. Since May 2007 he has been working as a science manager for Novozymes A/S in the Department for Brewing and Alcoholic Beverages in Copenhagen, Denmark.*

## O-6

### Development of DNA markers for the selection of beer foam stability in barley breeding

TAKASHI IIMURE (1), Makoto Kihara (1), Kazutoshi Ito (2), Seiichiro Ichikawa (1), Kazuhiro Sato (3), Kazuyoshi Takeda (3)  
(1) Sapporo Breweries Ltd., Ota, Japan; (2) Sapporo Breweries Ltd., Yaizu, Japan; (3) Okayama University, Okayama, Japan

In malting barley breeding, brewing quality traits are evaluated and selected only in later generations, which usually requires more than 10 years after initial crosses. A limited number of barley lines can be evaluated for brewing quality trials, because a certain amount of grain sample is required for brewing tests. DNA markers related to brewing traits may significantly reduce the efforts to select for these traits. In this study, efficient DNA markers for the selection of beer foam stability were developed. Both protein Z4 and protein Z7 were the candidate proteins controlling beer foam stability. To confirm the relationship between the levels of protein concentrations and foam stability, 24 beer samples were brewed from each malt of 10 barley cultivars. Regression analyses suggested that protein Z4 and protein Z7 could be positive and negative markers for beer foam stability, respectively. To develop DNA markers associated with protein Z4 and protein Z7 contents, their nucleotide polymorphisms in the barley cultivars tested were compared in the upstream region of the translation initiation codon, where the promoter region might be located. As a result, 4 and 24 polymorphisms were detected in protein Z4 and protein Z7 nucleotide sequences, respectively. By using these polymorphisms, cleaved amplified polymorphic sequence (CAPS) markers were developed. The CAPS markers for protein Z4 and protein Z7 classified 23 barley cultivars into 2 (Ryofu and Kendall types) and 3 (Ryofu, Kendall and Barke types) genotypes, respectively. Barley cultivars with the Kendall genotype in protein Z4 were significantly

higher in grain protein Z4 contents. Also, barley cultivars with Kendall and Barke genotypes were significantly lower in grain protein Z7 contents. Beer foam stability in the cultivars with Kendall genotypes in both protein Z4 and protein Z7 were significantly higher than those with Ryofu genotypes in both alleles. The results indicate that these CAPS markers provide an efficient selection tool for beer foam stability in barley breeding programs.

*Takashi Iimure received a master's degree in molecular chemistry from Hokkaido University, Sapporo, Japan, in 2001 and defended his Ph.D. thesis in March 2010 from Okayama University, Okayama, Japan. He has worked in the Bioresources Research and Development Department (formerly Bioresources Research and Development Laboratories), Sapporo Breweries Ltd., since 2004. His career at Sapporo has concentrated on the improvement of storage substances in barley seeds by means of protein and DNA techniques. He is currently a researcher in the R&D Support Center at Gunma, Japan.*

## O-7

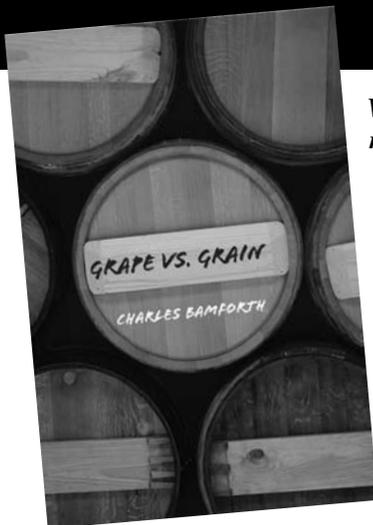
### Study of the effects of pre-harvest sprouting on the storability and malting quality of three Canadian malting barley varieties

YUESHU LI (1), Rob McCaig (1)  
(1) Canadian Malting Barley Technical Centre, Winnipeg, MB, Canada

Variations in germination and malting performance during storage for Canadian AC Metcalfe, CDC Kendall, and CDC Copeland barley samples were examined. These barley samples were collected from three successive crop years and had suffered different degrees of pre-harvest sprouting damage (PHSD) at harvest. During the course of this study these barley samples were stored under three different storage conditions to duplicate conditions seen in commercial shipping and handling of Canadian malting barley. During storage, PHSD barley samples showed

## Dr. Charles Bamforth Book Signing Event

12:00 p.m., Wednesday, June 16



*Why is wine considered sophisticated and beer is not even though the brewing process is more technologically complex?*

*Why is wine touted for its health benefits when beer has more nutritional value?*

*Why does wine conjure up images of staid dinner parties while beer denotes screaming young partiers?*

In his fascinating book, Charles Bamforth explores paradoxes involving beer and wine, paying special attention to the culture surrounding each. He argues that beer can be just as grown-up and worldly as wine and be part of a healthy, mature lifestyle.

**Purchase Grape vs. Grain at a 10% discount and have it signed by Dr. Bamforth on Wednesday, June 16 at 12 noon.**



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**Charles Bamforth** is Chair of the Department of Food Science and Technology and Anheuser-Busch Endowed Professor of Malting and Brewing Sciences at the University of California, Davis. He is Editor-in-Chief of the *Journal of the American Society of Brewing Chemists* and a member of the editorial boards of the *Master Brewers Association of the Americas Technical Quarterly*, the *Journal of the Science of Food and Agriculture*, and the *Journal of the Institute of Brewing*.

significant changes in germination energy and water sensitivity compared with barley with no PHSD. The higher the degree of PHSD the more variations in germination energy and water sensitivity were recorded. Barley samples stored at higher temperature showed more variation than samples stored at a lower temperature. Varietal difference in germination energy and water sensitivity and their interactions with storage conditions were also observed. In micro-malting trials, at steep the barley samples with higher degrees of PHSD showed faster water uptake and lower chitting rate than the barley samples with no pre-harvest sprouting damage. As with germination energy, barley samples with a high degree of PHSD stored at higher temperatures exhibited lower chitting rates. Some varietal differences in water uptake and chitting variations were also recorded. The trial results suggested that germination energy, chitting rate, friability, soluble protein, enzyme levels, and beta-glucan content were all sensitive to PHSD and storage conditions. Changes in malt quality could be related to the quality of the barley samples prior to storage, the subsequent barley quality change during storage, and the interactions between pre-storage barley quality and storage conditions. Both barley pre-harvest condition and storage conditions largely determine the changes in germination and overall maltability, as well as the resultant malt quality of the barley.

*Yueshu Li joined the Canadian Malting Barley Technical Centre in August 2000 and is the centre's director of malting technology. Previously, he was senior technical consultant for Malting Barley in the Market Development Department of the Canadian Wheat Board. Yueshu has held several senior research and management positions in the malting industry in both North America and China, including Prairie Malt Limited, Canada; Schreier Malting, USA; and CUC Nanjing Malt Limited, PRC. Yueshu completed his B.S. and M.S. degrees in China and holds a Ph.D. degree in plant physiology and ecology from the University of Saskatchewan.*

#### **O-8**

##### **The time-course of color and flavor formation during malt roasting processes**

Hafiza Yahya (1), DAVID J. COOK (1)

(1) University of Nottingham, Loughborough, U.K.

Roasted malt products can be used to add color and flavor in the brewing process. Flavor control in these products is principally exerted by “cooking to color” or utilizing specified operating conditions, but either may result in batch to batch differences, which can cause issues for brewers when swapping over from one batch of roasted malt to another. This study investigated the time course of flavor generation during malt roasting operations on commercial (2 tonne) and pilot (2 kg) scales. The aim was to better understand the underlying chemistry of flavor generation and, hence, to improve process control or open up routes to new, more energy-efficient products. Malt samples were taken at frequent intervals during the production of roasted barley, crystal, and black malts on both commercial and pilot scales (two replicates of each manufacturing process). In excess of 17 time-point samples were taken during each run, with the majority of samples taken during phases of rapid heating. Each time-point sample was analyzed for moisture content and color using the tristimulus CIE L\*a\*b\* system. Flavor volatiles in each sample were extracted (methanol) and analyzed quantitatively by GC-MS (three replicates per time-point sample), enabling average time-course data to be plotted for the generation of 15 flavor compounds. Results will be discussed in relation to the changing flavor composition during production of individual malts and comparisons will be made that illustrate the impacts of raw materials (i.e., malted versus non-malted) and process (e.g., impact of “stewing” on the flavor profile of crystal malts). Roasted barley product, for example, was characterized by very high levels of hydroxymethylfurfural (HMF), as opposed to black malt product, which contained high levels of maltol and 2-furaldehyde and minor amounts of HMF. Spraying water into the drum during roasting had

a significant impact on flavor formation pathways (stimulating some and depressing others), while the rate at which flavor composition changed close to the end point suggested that more accurate control in this area would be desirable to better control batch to batch flavor differences. CIE L\*, a\*, b\* data allow separate visualization of changes in malt color with respect to lightness and darkness (L\*), red (-yellow) hue (a\*), and green (-blue) hue (b\*). While it is possible to obtain finished roasted malt products with similar EBC color values at different production time points (since EBC color exhibits a peak), the use of tristimulus data allows these products to be differentiated with respect to the associated red or blue tints. Results indicated that for roasted barley and black malt products in particular, product tristimulus color characteristics changed relatively rapidly toward the end of processing, indicating that, as with flavor, end-point control significantly influences product color characteristics.

*David Cook received a B.S. (honors) degree in chemistry and food science from the University of Reading, U.K. He subsequently worked in both industry and academia prior to studying for a Ph.D. degree (2003) in flavor technology (University of Nottingham, U.K.). In 2006 David was appointed as lecturer in brewing science at the University of Nottingham and is course director for its innovative e-learning-based postgraduate courses for brewers. He is currently engaged in research across the malting and brewing fields. His specific interests include on-line analysis of flavor formation during thermal processing of malts (using mass spectrometry); genetic regulation of germination and dormancy in barley; the mechanism of malt-induced PYP; pretreatment of lignocellulosic materials to optimize bioethanol fermentations; and multisensory aspects of beer flavor.*

#### **O-9**

##### **Microbial community structure changes during the malting of Australian barley**

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Barley malt is a complex, dynamic ecosystem consisting of two constantly interacting components—microbes and germinating barley grain. The microbes contribute enzymes and other factors that can either favorably (i.e., proteases, cell wall-degrading enzymes) or unfavorably (i.e., premature yeast flocculation [PYF]) impact malt quality. This novel study compared the microbial population ecology (both bacteria and fungi) of Australian barley samples and the matched commercial malts from different growing and malting plant locations using molecular microbiology techniques such as terminal restriction fragment length polymorphism (TRFLP) and cloning and sequencing analyses. Statistical analyses of bacterial and fungal TRFLP data showed significant differences in both bacterial and fungal community structures of barley and their corresponding malts. These differences included the combined effect of both qualitative (type of bacteria/fungi) as well as quantitative (relative abundance of different bacteria/fungi) dissimilarities. The location of the barley and malt was also found to significantly influence the microbial community association with grain. The differences were greater for fungi compared with bacteria and for malts compared with barleys. Cloning and sequencing analyses of rRNA genes showed greater diversity in barley malt-associated bacterial and fungal community structure than previously observed.

*Mandeep Kaur received her Ph.D. degree in molecular microbiology from the University of Tasmania in Australia, with support from the Australian Research Council and Joe White Maltings. She is currently a post-doctoral research fellow at the University of Tasmania. She is researching the microbial population ecology of barley and malt, especially malts associated with premature yeast flocculation (PYF). Mandeep is supported by the Australian Grain Research and Development Corporation and Joe White Maltings.*

## O-10

### Using germination index and homogeneity to predict malt processing and quality

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Maltsters have always required knowledge of a barley's germination potential to assist in producing quality malt. Germination energy has been the method of choice for predicting how well a batch of barley can be expected to germinate during the malting process. This 3-day method provides ideal conditions, 100 kernels in 4 mL of water at room temperature, to induce germination of non-dormant barley. ASBC, IOB, and EBC all have official methods for this test, although details do differ. Additional information on the speed of germination, which can also affect malt quality, can be attained from the method by counting the germinated kernels every 24 hr as described in EBC's official germination index method. Reports in the literature also use the raw data to calculate germination homogeneity, a measurement of the uniformity of germination. The present study investigated how agronomic factors affect germination energy, germination index, and germination homogeneity and, in turn, how these germination parameters predict malt processing and malt quality. The experiment involved two varieties, AC Metcalfe and CDC Copeland, two seeding rates, and five nitrogen levels. The tested varieties were grown over multiple years and at a range of western Canadian locations. Agronomic factors were found to significantly affect germination index and homogeneity but not germination energy. Higher seeding rates produced barley with better germination indices and homogeneities, while high nitrogen rates tended to produce barley with poorer germination homogeneity. Varieties showed significant differences in germination properties. Barley with a higher germination index produced higher steep-out moistures but lower malt yields. Germination index and homogeneity correlated positively with endosperm modification (Calcofluor) and its homogeneity. In summary germination index and homogeneity were found to provide significantly more information, than germination energy, on a barley's malting potential and could be used as a tool for altering processing conditions in a predictable manner. Results support adaptation of the methods by ASBC, which would require simple additions to ASBC's standard Barley 3C method.

*Michael Edney received a Ph.D. degree from Weihenstephan, Technical University of Munich and an M.S. degree from the University of Saskatchewan. In 1988 he joined the Grain Research Laboratory of the Canadian Grain Commission, where he continues to research the quality of malting barley and its measurement. He has played a major role in the development and quality evaluation of new barley varieties in western Canada. He holds positions on the quality evaluation team of the Prairie Recommending Committee for Oat and Barley, the Western Grains Research Foundation Barley Advisory Committee, the Technical Committee of the Brewing and Malting Barley Research Institute, and the Executive of the Barley Development Council. Michael is the author of more than 50 research papers in scientific journals and articles in industry publications, as well as 7 book chapters.*

## O-11

### Impact of mashing-in temperature on extract and fermentability and the level of wort $\beta$ -glucan, soluble protein, free amino nitrogen (FAN), and lipids

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The quality of wort produced from malt during mashing has a substantial impact on determining the final beer quality. Wort quality in terms of

fermentable sugars, FAN, soluble protein, and viscosity-causing factors (e.g.,  $\beta$ -glucan) has been extensively investigated and is generally well understood in terms of the controlling malt and brewing factors, and how these impact beer quality. However, the interaction between barley variety, malting, and brewing processes on lipids in beer has not been widely investigated. This is despite the impact of lipids on beer quality in terms of yeast nutrition, flavor, and foam stability. The effect of mashing-in temperature (62.5–75.0°C) was compared on a series of eight varietal malts using small-scale brewing trials. It was found that increasing mashing-in temperatures from 62.5 to 75°C resulted in a slow but progressive loss of extract for all eight malt samples used. The optimal mashing-in temperature for maximal AAL (apparent attenuation limit) generation was 65°C with Sd2H varieties (high  $\beta$ -amylase thermostability), producing noticeably increased AAL at higher mashing in temperatures than the Sd1 and Sd2L varieties (intermediate and low  $\beta$ -amylase thermostability, respectively). With respect to lipids, mashing-in temperatures above 67.5°C, and particularly above 70°C, resulted in substantially higher levels of lipids in the wort (sample averages: 13.8 mg/L at 62.5°C, 12.5 mg/L at 65°C, 36.5 mg/L at 67.5°C, 116.9 mg/L at 70°C, 205.8 mg/L at 72.5°C, and 321.1 mg/L at 75°C). In addition, at each temperature point the amount of lipid extracted into wort varied significantly between the eight different malt samples, potentially indicating a varietal and/or sample effect. A relationship between the level of wort lipid and the speed of lautering was also observed, with the higher level of lipids giving poorer lautering performance. These results further demonstrate the importance of brewing conditions and malt quality on resultant wort quality, which is expected to be fundamental for final beer quality and downstream process efficiency.

*Evan Evans graduated with a B.Agr.Sc. (honors) degree in 1986, followed by a Ph.D. degree in 1990, both at the University of Melbourne. In 1992, he joined the University of Adelaide, where he developed his interest in malting barley and brewing. Recently he relocated to the University of Tasmania, where his brewing research interests continue to be in improving malt quality to improve beer quality and the efficiency of the brewing process. Evan is currently serving on the IBD Awards Committee and is a member of the editorial board for the ASBC Journal. In 2005, Evan was made a Fellow of the Institute of Brewing and Distilling. In 2009, Evan was an inaugural debater for ASBC's Pearls of Wisdom session on beer foam quality.*

## O-12

### Consistently meeting the specifications—The rising importance of defined quality control for craft breweries

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(1) VLB-Berlin

The craft brewers scene has come a long way in the United States—the production sites are not only rising in numbers and volume, but also in professionalism. A sign of professionalism for a quality beer is, as stated by Michael J. Lewis, simply “a beer that consistently meets specification.” Consistency is becoming more essential to production, and a control plan including methods of analysis is needed to help craft brewers ensure quality throughout production. Keeping records is the foundation of a quality control program, so that up or down trends can be detected rapidly. Upper and lower limits of acceptability should be established as guidelines for corrective actions. A simple statistical method for setting up plots will be presented. Once the proper specification measurements and consistency have been achieved through the use of concepts and methodologies craft brewers can then focus on the art of brewing. State-of-the-art methods named in a control sheet cover malt, hops, wort, beer, filter aids, and packaging from the chemical-technical, as well as microbiological and sensorial, side. A designed control plan provides the backbone to consistent brewing quality, with selected methods important to all growing craft brewers. A control plan set up will help craft brewers avoid quality assurance gaps that can result in a significant

cost impact, even within a small-batch processing environment. The idea of consistency immediately requires a system of people, plants, and processes that is able to repeat exact procedures. The lecture, therefore, will also address the questions of what annual barrel production, what suggested lab equipment, and which tests is it necessary to carry out at the craft brewery versus the idea of addressing third-party laboratories for analytical results. QA programs are essential to success in a brewery enterprise. The lecture provides basic techniques for rigorous quality testing that will help craft brewers repeatedly produce excellent specialty beers to meet the high standards of their market.

*Roland Folz apprenticed as a brewer and maltster at the Beck's brewery in Bremen, Germany. After working another year for the Beck's brewery, he started his studies in Berlin and received a diploma engineer degree in brewing technology from the Technical University, Berlin. After graduation, he worked as head of the Technical Department/Production at the Preussen Pils brewery in Pritzwalk, Germany, for two years. In October 2006, he started at VLB-Berlin as a global consultant for brewing technology, he worked for the Engineering and Packaging Department as the specialist for filling, packaging, and PET topics. In addition to his consulting practice, he is involved in teaching and research projects and manages the internationalization of VLB. Since autumn 2008, Roland has been the head of the Brewing & Beverage Technology and Applications Department at VLB-Berlin. This department includes the education and teaching part of VLB, as well as the research activities regarding technological topics, global consulting, analytics, and services.*

#### **O-13** **Modeling beer foam behavior**

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Many researchers have related beer foam properties to various beer constituents. Most of these studies used measurements of a number of beer properties and one or more foam assessments, both made on the same set of beers. Since both types of observations are actually dependent variables of the way the beers were made, correlations between them show associations, but cannot demonstrate cause-and-effect relationships. Further, simple correlations only test for linear (positive or negative) behavior, not curvature, and are mathematically incapable of demonstrating interactions between factors. A statistical experiment design (central composite face-centered with added interior points) was used to choose 41 combinations of 4 factors: protein (50–350 mg/L of ovalbumin), iso-alpha-acid (0–40 BU), pH (3.8–4.6), and ethanol (0–10% [v/v]). Samples were prepared at each design condition in 0.02M acetate buffer in 20 mm OD × 125 mm screw-cap tubes. After mixing and allowing time for equilibration, the samples were shaken by hand. Foam height was measured after 30 min. The collected data were modeled as a function of first-, second-, and third-degree polynomial and interaction terms using partial least squares regression. After removing several outliers and terms with minimal predictive value, an equation that predicted foam as a function of the remaining factors was obtained. This had a good fit ( $R^2 = 0.876$ ) to the observations. The model was used to predict results within the range covered in the experiment and revealed both curvature in and interactions between several factors. Foam height was positively affected by protein. Increasing BU from 0 to near 15 produced a sizable increase in foam but relatively little change between 15 and 30 BU (at constant protein levels). An additional foam increase in the vicinity of 35 BU was noted. Surprisingly, the greatest foam height was seen at low pH (near 3.8) with moderate ethanol levels (4–5% [v/v]). Both higher and lower ethanol led to reduced foam, and increasing pH decreased foam. The implications of these results will be discussed.

*Karl Siebert received a Ph.D. degree in biochemistry from Penn State in 1970. He then spent 18 years at the Stroh Brewery Company in Detroit,*

*where he held positions from research associate to director of research. In 1990, Karl joined Cornell University as professor of biochemistry in the Department of Food Science and Technology. He served five years as department chair and now has a predominantly research commitment. Karl is active as a consultant in beverage technology and chemometrics. He twice received MBAA Presidential Awards for papers he presented, and he and his colleague Penny Lynn have received the ASBC Eric Kneen Memorial Award (for the best paper published in the Journal of the American Society of Brewing Chemists in the prior year) three times. Karl received the ASBC Award of Distinction in 1999. He is a member of the ASBC Journal Editorial Board and the ASBC Foundation Board. Karl's research interests involve foam and haze in beverages, astringency and other flavor perceptions, the application of chemometric methods in food science, and assessment of microbiological risk.*

#### **O-14** **The influence of specific Maillard reaction products in malt on oxidative beer stability—Pro- and antioxidative effects**

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The influence of the Maillard reaction products in specialty malt on oxidative flavor stability is discussed. Prior investigations have shown that the development of stable organic radicals in malt during malting is directly related to the content of Maillard reaction products, color, and temperature during kilning. It was established that a higher concentration of organic radicals and Maillard reaction products resulted in a lower antioxidative potential of the wort and final beer. In further trials the influence of colored malt on oxidative stability was investigated using 10% different specialty malts and 90% pilsner malt in the brewing process. In relation to this the influence of adding malt extracts or artificial caramel color to pilsner beer compared with the influence of reductones was investigated. For the determination of endogenous antioxidative potential (EAP) and radical generation in wort and beer EPR spectroscopy was used (EAP-determination, T-400 and T-600 value). Simultaneously, the concentration of dicarbonyl compounds in wort was determined using HPLC methods. In direct comparison with the EPR methods, traditional methods were used according to MEBAK and Chapon to determine reduction power. Chapon's method provides information about the content of reductive substances and their ability to reduce  $Fe^{+3}$ , which is one of the Fenton reaction products. The MEBAK reducing power method indicates the quantity of fast-reacting reducing substances. The results show a direct correlation between the content of Maillard reaction products in malt and lower oxidative stability (EAP) in wort and final beer. This negative effect of specialty malts is based on a more rapid consumption of  $SO_2$  during the brewing process and in the finished beer. In contrast, the same specialty malts led to a higher reducing power when measured by the Chapon or MEBAK methods. These contradictory results seem to be one reason why the influence of specialty malts on beer stability has been a matter of discussion in the literature. The explanation of this indirect correlation arises from the strong reduction properties of specific Maillard intermediate reaction products such as reductones or other dicarbonyl compounds that can rapidly reduce oxidized metal ions like  $Fe^{+3}$  and consequently intensify the Fenton and Haber-Weiss reaction system. Based on the acceleration of the Fenton and Haber-Weiss reaction system, a stronger radical generation of very reactive radicals (e.g.,  $OH\cdot$ ) can be observed in the wort and beer matrix. This leads to the observed faster consumption of specific antioxidative substances such as  $SO_2$ . The results present the pro- and antioxidative effects of the Maillard reaction products and can explain the influence of the reductone compounds in beer caused by the addition of specific malts or ascorbic acid (reductone).

*Frank-Jürgen Methner studied brewing science at Berlin University of Technology (TU Berlin) from 1975 to 1981. After finishing a Dipl.-Ing.*

degree, he worked as an operating supervisor at the Schlösser Brauerei. From 1982 to 1986 Frank was a scientific assistant with teaching duties at the Research Institute for Brewing and Malting Technology of VLB in Berlin. For 18 years, starting in 1987, Frank-Jürgen has held a leading position as a director at the Bitburger Brauerei, Bitburg, Germany, with responsibilities in fields such as technology and quality assurance. Beginning with the winter semester of 2004–2005, he took over the Chair of Brewing Science at TU Berlin.

## O-15

### **PYF analyses of commercial malt samples—Assessment of recent results**

MICHAEL VOETZ (1), Heiko Woest (1)

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In 2009 VLB Berlin analyzed 233 malt samples originating from different malt source plants for PYF activity. The fermentation protocol applied is based on the PYF test published by Asahi Brewing Co., which uses a malt-derived Congress wort. According to this protocol the flocculation yeast ratio (FYR: 40 hr/24 hr) serves as measure for the risk of PYF in industrial-scale fermentations. Almost a fifth of all malts tested in that period revealed flocculation ratios of less than 60%, which can be defined as a critical value. Surprisingly, 23 samples exhibited the PYF phenomenon with less than 50% FYR. In these cases PYF also could be assessed visually. A statistical overview will be presented completed by the data generated until early summer 2010. We found that different portions of a 1-kg malt sample revealed different behavior with respect to PYF. The content of removed husk material within the malt sample analyzed was responsible for this effect. The data underline the importance of sampling in the malt house as well as in the lab. A water-soluble PYF factor was postulated by different researchers to be responsible for the phenomenon. In addition to the data published by A. Speers and coworkers we could identify the factor present in a fraction representing molecules with a molecular mass of 100–300 kDa. According to S. van Nierop there is a link between fungi on barley, the release of high molecular weight polysaccharides serving as PYF factor, and the occurrence of PYF. The gushing phenomenon also was induced by different fungi. Since VLB possesses hundreds of well-characterized samples with high gushing risk, we checked for a correlation between the two phenomena. For this purpose small-scale fermentations were established that enabled us to analyze a large number of relevant samples. After analyzing a first set of 50 malt samples with different gushing risk values a correlation between gushing and PYF could not be observed.

*Michael Voetz, born in 1964, is head of the Biological Laboratory at VLB Berlin. He earned a Ph.D. degree in plant molecular biology from the University of Cologne/Max-Planck-Institute for Breeding Research in 1995. From 1995 to 2000, he was a scientific collaborator in the Research Department of the Weissheimer Malzfabrik in Andernach, working in the field of barley biotechnology. From 2000 to 2007 he was head of the biotechnology/PCR laboratory at the Research Institute for Raw Materials within VLB Berlin.*

## O-16

### **Will the key anti-oxidant protein in beer please stand up**

PETER J. ROGERS (1), Vincent Higgins (2), Ming J. Wu (2), Frank M. Clarke (1)

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When it comes to the quality ledger, beer protein has an ambivalent record: it can be a root cause of physical stability and even, some say, flavor instability. On the positive side protein is an important contributor to mouthfeel and likewise to foam formation and lacing, although yeast proteinase, a protein, breaks down foam-stabilizing proteins in some brews. Still, we have not wavered from a belief that some beer proteins are functional and may improve flavor stability. There is a causal link

between protein levels in beer and flavor stability. In particular to a protein subgroup around 12-kDa molecular mass. This fraction is redox active, i.e., the thiol/disulfide (PrSH/Pr-S-S-Pr) ratio changes reversibly during fermentation. And, during storage protein thiol levels mirror beer flavor. This redox protein may have peroxidase activity directing reducing equivalents to destroy peroxides. There are precedents; a yeast thioredoxin-dependent peroxidase uses the reducing power of two thioredoxin thiols. The active (reduced) thioredoxin is regenerated in ferments by another enzyme system dependent on metabolism. In beer there is a significant pool of reactive polyphenols and low molecular weight molecules—cysteine, dicarboxylic acids, aldehydes, etc.—that might act as electron donors, like the yeast system. This would generate a much larger pool of reductants than is commonly accepted. One way to explore this possibility is using a reconstituted model system. The antioxidant activity of the 10-kDa protein fraction was screened against six oxidants using a *Saccharomyces cerevisiae*-based assay. This fraction counteracted all six oxidants; yeast growth was 4- to 5-fold higher than the controls in the presence of H<sub>2</sub>O<sub>2</sub>, LAH (linoleic hydroperoxide), peroxyxynitrite, or diamide. We have now isolated and identified the redox-active, low-MW protein from beer. The 10-kDa band was excised after SDS-PAGE, trypsinized, and sequenced by ESI-MS/MS. The protein was identified by matching sequenced peptide in protein databases. This analysis showed that the protein is barley LTP1 (lipid transfer protein). Barley LTP1 has the same free-radical scavenging and antioxidant capacity as the starting 10-kDa protein fraction. Barley LTP1 with free-thiol groups on its cysteine residues is abundant in some beers, in particular kettle-hopped beers. It is absent in aged beer with stale flavor. This suggests a role for LTP1 as a diagnostic for quality. But more than that, we are poised to develop a model antioxidant combination that can be realized in commercial beers.

*Until 2010 Peter Rogers was the national manager of research within the Supply section of Carlton and United Breweries (CUB). He joined CUB's Brewtech technical arm in 1997. A chemistry graduate, he completed his Ph.D. degree in yeast genetics at the Australian National University, worked at Germany's Max Planck Institute for Experimental Medicine in the 1970s before moving to Griffith University in Brisbane. He was part of the Queensland push into beef coproduct recovery with regional producers to make high-end products for pharma and food use. He served on the international advisory group for the European Brewery Convention driven by the Brewers of Europe. He is a past president of the former Australian Biotechnology Association. He received the Presidential Award from the Master Brewers Association of the Americas in 2001 and the Eric Kneen Memorial Award from the American Society of Brewing Chemists in 2005. He is an adjunct professor at Griffith University. He has experienced first-hand the barrage of industry mergers and acquisitions over the past 12 years, the marriage of beer and wine and beverage, and the reshaping (reinvention might be a better term) of the international technical communities.*

## O-17

### **Improved flavor stability by aging beer in the presence of yeast**

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Several quality aspects of beer are subject to change during storage. Alteration of the flavor profile in particular is of great concern to brewers, as flavor is considered the main quality parameter. During beer aging, fresh flavor notes diminish, and several typical aged flavors appear as a result of numerous chemical reactions. Despite extensive research, it remains very difficult to understand and control flavor stability. In previous studies, carbonyl compounds appeared to be the main contributors to aged flavor. Furthermore, yeast appeared to have high reducing activity on these compounds during fermentation and refermentation processes. As a result, high concentrations of aging

compounds can be removed almost completely from wort or beer. These results are promising for applying yeast in order to obtain better flavor stability. In this study, a fresh lager beer was refermented with a bottom-fermenting and a top-fermenting yeast (1 million cells/mL). The refermentation was performed with or without the addition of fermentable extract to the bottle. After the addition, the beers were aged for 6 months at 20°C, and the flavor was evaluated by sensory analysis and the determination of aging compounds using headspace solid-phase microextraction coupled to gas chromatography and mass spectrometry. Through sensory analysis, it became evident that the beers to which yeast was added were substantially less aged, irrespective of the addition of fermentable extract. In these beers, only traces of aged flavor notes were perceived after aging, while beers that were aged without yeast had a strong aged flavor. Hence, the activity of both the bottom- and top-fermenting yeasts resulted in lower aged flavor. These sensory results were very much reflected in the concentrations of the aging compounds. Carbonyl compounds that were previously shown to be reducible by yeast, such as 3-methylbutanal, phenyl acetaldehyde, methional, 5-hydroxymethylfurfural, and diacetyl, especially were lower in the beers that were aged with yeast. Further experiments were set up by adding varying amounts of yeast, without fermentable extract, to lager beer. After aging, similar results were obtained, and it became evident that yeast could diminish aged flavor, even when yeast cell concentrations of only 10,000 cells/mL were applied. Again, the concentrations of aging compounds corresponded very well with the sensory perception of aged-beer flavor. Moreover, at these low yeast cell concentrations, only limited haze was observed in the beers. In conclusion, it can be stated that the flavor stability of beer can be improved substantially by adding yeast cells to the bottle, even at very low concentrations.

*Daan Saison graduated as a bioengineer in food chemistry and technology from the Catholic University of Leuven in 2005. He carried out his master's thesis research on glycoside hydrolase activity in brewer's yeast and its influence on hop glycosides. Afterward, he started a Ph.D. program at the Centre for Malting and Brewing Science at K.U.Leuven (Belgium). He presented a part of his work at World Brewing Congress 2008 in Honolulu, HI, and finished his Ph.D. thesis, entitled "Role of Carbonyl Compounds and the Impact of Yeast Reducing Activity," in September 2009. He is now working as a post-doctoral researcher at the Centre for Malting and Brewing Science in Leuven.*

#### **O-18** **Monitoring yeast physiological state during fermentation by quantitative cell morphogenesis analysis**

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Yeast cell morphology is highly specialized and strictly controlled under fermentation conditions. Although many differences in yeast cellular morphology during fermentation have been observed in microscope images of yeast cells, it is difficult to analyze the morphology quantitatively. Thus, the relationship between cellular morphology during fermentation and the physiological state of yeast remains unknown. Recently, an image-processing program that recognizes yeast cell morphology was developed. This program, named CalMorph, can extract quantitative morphological data directly from microscope images to allow quantitative analysis of yeast cell morphology using 501 parameters that give information on cell shape, actin, and nuclei. This technique was applied to bottom-fermenting *Saccharomyces pastorianus* KBY011 yeast cells under fermentation conditions. Cells were incubated at 20°C for 96 hr in YPD10 medium (1% yeast extract, 2% peptone, and 10% glucose) under anaerobic conditions with sampling every 24 hr. Yeast cells were stained with fluorescein isothiocyanate-concanavalin A (FITC-ConA)

for cell wall identification, with DAPI to localize nuclei and rhodamine-phalloidin (Rh-ph) to visualize actin distribution. After taking pictures, CalMorph analysis was performed to quantify the brightness of all pixels in each digital cell image and to recognize yeast cell shape and the shapes and locations of stained cell components. The quantitative data for at least 200 cells of each sample analyzed to quantify 501 morphological parameters using CalMorph were investigated to develop profiles of time-course morphological changes during fermentation. These data were applied to average-linkage hierarchical clustering analysis, and time-course specific morphological parameters were identified. These results clearly indicate that there are morphological differences in cells during fermentation and that these occur at different stages of the cell cycle and reflect various cellular characteristics. Here, we demonstrated that CalMorph is a powerful tool for monitoring the yeast physiological state during fermentation. This approach offers a possible way to monitor and optimize yeast performance during fermentation.

*HiroYuki Yoshimoto received a Ph.D. degree in engineering from Hiroshima University, Japan, in March 1992. He was employed in April 1992 at the Central Laboratories for Key Technology, Kirin Brewery Company, to conduct research on yeast. He also studied yeast technology at Stanford University, California, from June 1999 to September 2001. Since March 2007, he has been working in the Research Laboratories for Brewing, Kirin Brewery Company, Limited.*

#### **O-19** **Key flocculation performance indicators during production-scale lager brewing fermentations**

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Flocculation is a key performance indicator for brewing fermentations. Inconsistencies frequently occur either because of the genetic instability of the yeast genome, changes in wort composition, or changes in process conditions. Achieving consistency even when major problems do not occur remains a challenge. To address this omission the current study (conducted in last 12 months and not previously reported or presented) focused on examining flocculation in lager full production-scale (2,697 hL) fermentations. Changes in viability and budding index were monitored. The fermentation profile was typical for the lager strain assessed. Fermentable consumption was also monitored because fermentation sugars inhibit flocculation either directly by binding to the lectins that enable cell wall interactions to occur and/or indirectly because flocculation onset is triggered by a starvation response. Interestingly, a significant reduction in cell density after 60 hr into the fermentation corresponded to the complete utilization of glucose, fructose, maltose, and maltotriose, as detected by HPLC analysis. This was, however, 20 hr prior to flocculation onset. Cell surface properties indicated a gradual increase in both flocculation and hydrophobicity profiles of the yeast cells, whereas cell surface charge remained relatively unchanged. Furthermore, genome-wide microarray data permitted the correlation of wall-related gene expression levels to changes in environmental and cell surface properties. These observations were been supported by parallel investigations in the laboratory, in which the genetically pliable laboratory strain, BY4741, was used as a model to investigate the impact of individual mannoprotein-encoding genes on both flocculation potential and hydrophobicity in environments designed to mimic fermentation conditions.

*Katherine Smart completed a B.S. (honors) degree in biological sciences at Nottingham University in 1987 and was awarded the Rainbow Research Scholarship to complete a Ph.D. degree in brewing yeast and fermentation at Bass Brewers, Burton-on-Trent. She then moved to Cambridge University to take up an appointment as research fellow in the Department of Plant Sciences, where she worked on bioactive surfaces, biofouling, and bacterial contamination of beverages in collaboration with the beverage packaging company Elopak. In 1992,*

*Katherine became a lecturer in microbiology and fermentation at Oxford Brookes University. By 2000, she had been appointed to Scottish Courage Reader in Brewing Science and became the youngest Fellow of the Institute and Guild of Brewing. In 2005 Katherine moved to the University of Nottingham, where she became the SABMiller Professor in Brewing Science. Katherine has received several awards for her research, including the Institute of Brewing and Distilling Cambridge Prize (1999), the prestigious Royal Society Industrial Fellowship (2001–2003), an Enterprise Fellowship (2002), and the Save British Science Award at the Houses of Parliament in the United Kingdom (2003). She has also recently commenced patent filing for a novel PCR technology. Her core research interests are yeast cell biology, fermentation, and stress responses in yeast.*

#### **O-20**

##### **An exploration of the relationships between yeast physiology, brewery propagation, and fermentation performance**

CHRISTOPHER A. BOULTON (1), Katherine Miller (1), Katherine Smart (1)

(1) University of Nottingham, Loughborough, U.K.

The periodic introduction of new yeast cultures into the brewery via propagation is an essential prerequisite for ensuring consistent fermentation performance. Strategies that aim to intensify fermentation productivity, such as the use of large-batch, high-gravity worts and elevated fermentation temperatures, impose increased stress on brewing yeast and, consequently, restrict the number of times it may be re-pitched and, therefore, lead to an increase in the frequency of propagation. In order to satisfy the requirements for yeast supply in large-capacity breweries, modern propagation plants must have a high yield and short cycle times. Generally this is achieved by cultivation of yeast under continuous aerobic conditions and at relatively high temperatures. The use of wort as a feedstock ensures that the yeast retains a repressed physiology similar to that of conventional pitching yeast cropped from fermentation. High temperatures favor rapid growth rates, and aerobiosis promotes high yeast yields. Propagators of this type yield in excess of 200 million cells/mL within a total cycle time of less than 48 hr. In theory such yeast should contain relatively high concentrations of sterols and unsaturated fatty acids and, by inference, should produce vigorous fermentations when pitched into first-generation worts. However, it is usually observed that first- and sometimes second-generation fermentations are slower than standard, and the subsequent beers require blending. Since the maximum number of permissible fermentations before the culture requires replenishment is commonly no more than 5–10, this non-ideal behavior can pose a significant problem. Here the design and operation of a modern aerobic brewery propagator is described. Results of trials that have sought to probe aspects of the physiology and genetics of yeast during propagation and throughout subsequent generations of fermentations are presented. Evidence is provided that suggests that the conditions at the end of propagation exert an influence on yeast, which in turn has an impact on fermentation performance. Potential methods for controlling the end point of propagation in order to ensure consistent fermentation performance are suggested.

*Chris Boulton received his first and second degrees in the Department of Biochemistry, University of Hull. He joined the R&D Department Bass Brewers as a fermentation microbiologist in 1984. Since that time he has had a variety of roles within the same company and latterly with Coors Brewers, where he has maintained and developed an interest in studying the relationships between yeast physiology and fermentation performance. In particular, he has developed a passion for unraveling the responses of yeast to the production environment, especially during fermentations using very large batch sizes. He is currently employed as a freelance brewing consultant and as a teaching fellow in brewing science at the University of Nottingham. He is the author of more than*

*70 research papers and review articles and is a coauthor of two text books, Brewing Yeast and Fermentation, with David Quain, and Brewing Science and Practice, with Dennis Briggs, Peter Brookes, and Roger Stevens.*

#### **O-21**

##### **Analysis of flavor compounds by yeasts in insufficient nutrition II—Studies on brewing yeast indole production and the tryptophan pathway**

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In brewing, it is important to control the content of off-flavors in final products because of their unpleasant character. The production of off-flavors is dependent on the brewing conditions. In particular, yeast metabolism plays a key role and tends to produce off-flavors under conditions of insufficient nutrition. Recently in Japan, beer-flavored beverages have been brewed with no or less malt compared with regular beers. However, the raw materials used in these beer-flavored beverages contain less nutrition than malt, which sometimes leads to sluggish fermentation and the production of off-flavors. In order to deal with these issues, we are studying the relationship between the shortage of various nutrients in wort and the flavor profile of the final product. At the 2009 ASBC Annual Meeting, we reported that the lack of vitamin B<sub>6</sub> and tryptophan in wort causes the production of off-flavors by brewing yeast, and we identified it as indole, which is an intermediate of the tryptophan synthesis pathway. We also confirmed that adding vitamin B<sub>6</sub> and tryptophan to wort repressed the production of indole by yeast. During further studies on the mechanism of indole production in brewing yeast, we were interested in the relationship between the behavior of indole content and oxygen. Indole content increased in the presence of oxygen, for example, in the early stage of primary fermentation or when using a doubling fermentation method. On the other hand, indole content decreased in the late stage of primary fermentation, which contains little oxygen. As previously described, indole is produced by the tryptophan synthesis pathway and metabolized into tryptophan by tryptophan synthase, which requires vitamin B<sub>6</sub> as a cofactor. Tryptophan is further metabolized into nicotinamide adenine dinucleotide (NAD). In this step, oxygen is required. Niacin, glutamate, and aspartate are also precursors of NAD independent of oxygen. We hypothesized that if indole is produced to synthesize NAD, external addition of niacin would repress the production of NAD from tryptophan, and similarly, the production of indole would be repressed even under a shortage of vitamin B<sub>6</sub> and tryptophan in wort. To verify this hypothesis, we examined whether the production of indole is repressed by adding niacin to the wort. As expected, we found that the addition of niacin had a significant effect on lowering the indole content. This result indicates that in addition to vitamin B<sub>6</sub> and tryptophan, the production of indole is repressed by niacin, which also suggests that the production of indole is relevant to NAD synthesis.

*Takeshi Arai received an M.S. degree from the Department of Agricultural and Environmental Biology, University of Tokyo. He found employment with Sapporo Breweries, Ltd. in April 2007 as a microbiologist in the Frontier Laboratories of Value Creation.*

#### **O-22**

##### **Improving beer flavor and fermentative capacity with selected hybrids *S. cerevisiae* and interspecific *S. uvarum* × *S. cerevisiae* produced on specific maltose medium**

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(1) Spindal, Gretz-Armainvillier, France; (2) INRA, Thiverval-Grignon, France

The development and production of selected beer yeast for a fast and complete metabolization of maltose, maltotriose, and glucose, the three main fermentable sugars in wort, has been considered. Maltose, maltotriose, and glucose are the most abundant fermentable sugars in wort; in the case of incomplete fermentation, maltotriose can cause a range of qualitative problems in beer and ethanol loss. Furthermore, yeast that comes pre-grown on glucose biomass cannot fully adapt itself during beer fermentation. Using the fact that the medium constitution for the biomass production of the selected yeast strain is a key factor for gene expression, the same strain produced on maltose medium has a greater impact on maltose and maltotriose metabolization. To exploit the potential of the selected yeast, an interesting solution was its hybridization with *S. cerevisiae* or *S. uvarum* by crossing of spores or cells from selected strains of our collection, which showed interesting characteristics. This hybrid, thanks to the union of the two genomes, permitted increased enzymatic expression. The performance of fermentation was followed through the optimization of the culture medium, reproducing accurately the wort composition by monitoring yeast growth, ethanol synthesis, original gravity, attenuation, and sugar consumption during the fermentative process. Beer flavor was evaluated through the content of higher alcohols, volatile esters, and aroma compounds. In this study, we investigated the influence of medium composition on the overexpression of different genes encoding for maltose and maltotriose permease especially and the impact on yeast metabolism and correlated these with beer aroma profile. This optimized medium for yeast biomass, yeast strain selection, and breeding displayed an improved fermentative capacity and improved maltose and maltotriose conversion, resistance to ethanol, and temperature choc. The equilibrium and reproducibility of the aromatic profiles have also been analyzed and compared with traditional yeasts after successive inoculation for mutation, membrane permeability, and permease.

*Mustapha Nedjma is the director of the Research and Development Department within AEB Group, has been in charge of biotechnologies since 1997 in the facility based in Île de France, close to Paris. He received two post-doctoral positions specializing in microbiology, enzymology, and fermentation. He has published several papers, reviews, and patents for the beverage industry, especially in the beer, wine, and juice fields. His current research activities include beer fermentation and production of enzymes under solid-state fermentation (SSF).*

#### O-23

##### **About the different behavior of miscellaneous wort aroma compounds during wort boiling—Concentrations in kettle-full and -finished worts and condensed vapors**

UDO KATTEIN (1), Stefan Hanke (1)  
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The wort-boiling process is one of the most important steps during beer production because it has an enormous influence on flavor and flavor stability. Due to extreme conditions such as high temperatures and times up to 90 min, the complex mixture of reaction partners provides an excellent environment for thermal-induced chemical reactions, i.e., the release of aroma compounds. In addition to these formations it must be considered that a lot of similar or identical compounds also can be detected in the kettle-full worts. During the boiling process there is an interaction between formation and evaporation of volatiles. The behavior of the compounds is not equal but depends on a lot of physical and chemical characteristics, e.g., different volatilities and reaction rates. Until today, no fundamental approach could be found with regard to the ratios between release rate and evaporation of aroma compounds during wort boiling at an industrial scale. The reasons are obvious: for exact determination of these connections it is necessary to condense, collect, and analyze the vapors. In commercial brewhouses these facilities simply cannot be installed. The Weihenstephan research brewery has a 8-hL scale finished wort, modern boiling systems, and a self-constructed vapor condenser. This unit enables the total precipitation of vapors up to

kettle evaporation rates of 10%; the cooled condensate is at 6°C. With this equipment it was possible to perform screening tests on the interplay between formation and evaporation of miscellaneous volatiles during the wort-boiling process at a semi-industrial scale. The brews were performed with our standard Stromboli boiling system. With regard to the volatiles, it was suggested these aroma compounds be divided into three groups: high, medium, and low volatile components. We monitored substances from each group: the high volatiles DMS and a summary of strecker aldehydes; the medium volatile phenylacetaldehyde; and the low volatile 2-furfural. The paper shows details about the condensing device and gives an overview about the trials and the results obtained.

*Udo Kattein received a diploma engineer degree from the Technical University of Munich – Weihenstephan in 1972; afterward, he performed an economic study at the University of Munich, finishing a diploma merchant degree in 1976. At that time he started work on his doctoral thesis and employment at the TU Munich. He was in charge of the technical leadership of the Trial and Research Brewery Weihenstephan. He served as head brewer and was responsible for production of commercially sold malts and top-fermented beers. In addition to theses, tasks he was involved in the development of new beer types and training students. In 1984 he received a Ph.D. degree in engineering sciences, with a thesis on investigations of sulfur compounds in malt, wort, and beer. Since 2002 he has been responsible for the construction of the new malting and brewing facilities of the research brewery, which began in 2005.*

#### O-24

##### **Studies for specific control of astringent substances in malt to improve beer aftertaste**

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(1) Suntory Liquors Limited, Osaka, Japan

We have already reported that the malt acrospire contains ingredients that could aggravate the aftertaste of beer. Using nuclear magnetic resonance and mass spectral analysis, the chemical structures of those components were determined to be derivatives of hordatine. We have also demonstrated that the removal of acrospires from malt using the malt fractionation technique is a good way to reduce astringent substances in beer. In the beer using malt without acrospires, however, the value of total nitrogen, free amino nitrogen, and total polyphenols were also reduced, and the character of beer was changed to clean and mild. It was thought that the physical removal of malt tissues by this method would not provide specific enough targeting of the astringent substances. At the 2009 ASBC Annual Meeting, we reported that malt astringent substances could be degraded by treatment with sub-critical water. The reacted solution was composed of lots of degraded products, which revealed that the degradation was not simply carried out but complicatedly hydrolyzed and thermally decomposed. In this presentation, we will discuss which moieties of the astringent substances would cause astringency in beer flavor based on analysis of the chemical structure. We studied whether the flavor of the astringent substance could be changed to calm by targeting a change in some astringent moieties in the molecule, which could be possible to improve beer flavor by specifically reducing the astringent substances.

*Norihiko Kageyama received his M.S. degree in chemistry from Osaka University in 1998. He joined the Institute for Fundamental Research, Suntory Ltd. in 1998 as a chemist researching natural products. Prior to developing brewing technologies in the Beer Development Department, he worked in the Process Development Department and developed material processing technologies mainly based on subcritical fluid technology for development of new food and beer items. He was also actively involved with studies on identification of malt astringent substances and development of malt fractionation technology. He received the JB Award for his excellent article published in the Journal of Biochemistry by the Japanese Biochemical Society in 2000. He received The SCEJ Technology Award from the Society of Chemical Engineers,*

Japan, for his contribution on the development of subcritical fluid technology in 2005.

#### O-25

##### **Rapid determination of the reactive (off) flavors 4,5-epoxy-2E-decenals in beer**

LEIF A. GARBE (1), Konrad Neumann (1)  
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The flavor-active compounds *cis*- and *trans*-4,5-epoxy-2E-decenals result from linolenic acid peroxidation and cleavage. These flavor-active compounds have odor thresholds of approx. 0.01 ppb retro nasal in water. Epoxydecenals have a reactive epoxy group, conjugated double bond, and aldehyde function; therefore, their accurate analysis requires isotope standards and a rapid workup procedure. The chemical synthesis of deuterated and C-13 labeled standards has been demonstrated previously. The beer workup was performed with RP-18 solid-phase extraction (SPE) in five parallel samples of 50-mL each. No further workup procedure was necessary. Analysis was performed by GC-MS in the negative chemical ionization (NCI) and selected ion monitoring (SIM) mode. A simple method for the synthesis of high reactive *cis*- and *trans*-4,5-epoxy-2E-decenals was established and used to achieve isotope standards. The beer workup procedure was optimized, and the SPE technique provided the capability of automatization and fast analysis compared with lq.-lq. extraction/SAFE sample preparation. GC-MS analysis with NCI enabled limits of detection for epoxydecenals of <1 ng/L (0.001 ppb) in beer. The content of epoxydecenals in beer (fresh) varied from 0.01 ppb (fresh beer) to 0.05 ppb (40°C, 68 hr). The exposure of fresh beer to air at room temperature revealed 0.006 ppb for the *cis*- and 0.005 ppb for the *trans*-isomers. Exposure at 7°C led to concentrations of 0.003 ppb for the *cis*- and 0.002 ppb for the *trans*-isomers in beer. The synthesis and usage of epoxydecenal isotope standards enables accurate analysis of epoxydecenals in the sub-parts per billion range in beer. An optimized sample workup, especially SPE and subsequent GC-NCI-MS allows fast and reliable epoxydecenal quantification at ultra-trace amounts (ppt) in beer.

*Leif-Alexander Garbe is a professor and the head of the TU Berlin Chair for Bio-analysis/Molecular Analysis and, since 2002, also the head of the VLB Research Institute for Spezialanalytik. Leif graduated from TU Berlin (TUB), Germany, with a diploma in chemistry in 1996. He then worked at the Research and Teaching Institute for Brewing in Berlin (VLB). From 1997 to 2002 he worked on his Ph.D. thesis and received his degree in April 2002 from the Institute of Biotechnology, TUB. His work included supervision of students in biotechnology and brewery. In 2002 he established a new research group at TUB that focuses on trace analysis, biotransformations, and biocatalysis. In July 2008 he became the head of the TUB Department for Bio-analysis/Molecular Analysis.*

#### O-26

##### **Impact of wort aeration period of multi-filling cylindroconical vessels**

YUICHI NAKAMURA (1), Taku Irie (1), Minoru Kobayashi (1), Miho Shikata (1)  
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We analyzed fermenting beer and yeast from five main breweries (5,000-hL cylindroconical fermenting tanks, filled with four 1,250-hL batches of wort) of Asahi Breweries. We measured dissolved oxygen of fermentation vessels, budding index, fatty acids of fermenting yeasts, yeast gene expression, esters, higher alcohols of beer, and amino acids. These results showed that the length of the aeration time period for the initial stage of fermentation—flotation process, aeration process, and filling up the fermenter—is important for brewing beer. When the oxygen was supplied to the wort for a prolonged time, intracellular fatty acids were excessively unsaturated, esters were decreased by the unsaturated fatty acids, brewing yeast cell cycle was desynchronized, abnormal budding yeasts increased, and nitrogen metabolism of fermenting yeast was depressed. We pitched

the same amount of yeast into the four batches of wort. This means the first and second pitched yeasts had excessive contact with oxygen. We examined a modified pitching method in a 50-hL pilot plant that was filled with two batches of wort. We compared late pitching (yeast pitched for the second wort only) with early pitching (yeast pitched for the first one only). The late-pitching beer had increased esters and decreased unsaturated fatty acids, ergosterol, and budding index. Conversely, early pitching increased unsaturated fatty acids, ergosterol, budding index, number of abnormal yeasts, and amino acid metabolism. We believe that the fermentation with late pitching occurred under anoxic conditions; in contrast, with early pitching fermentation occurred under hyperoxic conditions. These results suggested that the time for wort aeration and exposure to oxygen for yeast are meaningful for breweries using multi-filling cylindroconical vessels.

*Yuichi Nakamura received an M.S. degree in agricultural chemistry from Tokyo University, Japan. He began employment with Asahi Breweries, Ltd. in April 1993. After working as a researcher in the laboratory, he was transferred to the brewing section of Ibaraki brewery. He studied brewing technology at TU Muenchen-Weihenstephan in Germany for one year (2001–2002) and returned to the Nagoya brewery. He has been working in the Production Technology Center, Asahi Breweries, Ltd. since 2005.*

#### O-27

##### **The importance of wort composition in yeast metabolism during accelerated fermentations**

PIETER J. VERBELEN (1), Filip Delvaux (1), Freddy R. Delvaux (1)  
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In previous studies, it was demonstrated that high cell density (HCD) fermentations can drastically increase the speed of fermentation and have great potential in the brewing industry. Next to this, high-gravity brewing (HGB) is already a well-established strategy to increase brewing capacity, and most of today's lager beer is produced with this method. However, it is known that both HGB and HCD fermentations have a considerable influence on yeast metabolism and physiology. In this study, the emphasis was placed on the effect of wort composition, in particular the C/N ratio and the relative concentration of sucrose and maltose, on yeast stress response and flavor formation in HCD fermentations of high-gravity wort. Six different worts (15°P all-malt reference, 18°P all-malt, 14+4°P maltose or sucrose, 10+8°P maltose or sucrose) were fermented in 2-L tall tubes at a pitching rate of 80 million cells/mL and examined in duplicate. Fermenting medium parameters (density, cell count, amino acid content) were measured together with the assessment of yeast viability, gene expression profiles, and glycogen and trehalose contents during fermentation. In this study, it was observed that high-gravity worts exerted an extra source of stress on yeast cells in HCD fermentations, since both osmotic stress and ethanol stress induced the expression of stress-related genes. Additionally, higher trehalose levels were triggered, which ultimately led to decreased yeast viability. More than the wort density, the use of sucrose, an easily fermentable sugar, had a drastic impact on yeast fermentation performance compared with maltose. Sucrose stimulated yeast growth and fermentation productivity, but its consumption was associated by repressed subsequent uptake of maltose and maltotriose, strongly decreasing fermentation power in the second part of fermentation. Worts with high amounts of sucrose strongly enhanced the uptake of branched-chain amino acids and influenced trehalose and glycogen metabolism and the expression of stress-related genes. It seems that the use of sucrose, which fully activates the cAMP/PKA pathway, has a deleterious influence on yeast health. This could limit the application of HCD fermentations. Finally, flavor formation during HCD fermentation was strongly dependent on the wort specification (density, C/N ratio, and type of sugar used). This could be explained by the differences in yeast growth, altered uptake rates of amino acids, and changes in gene expression levels (BAP2, ATF1, ILV2, and ILV5). Hence, it is important

to fine-tune the wort composition in order to obtain the desired flavor profile.

*Pieter Verbelen graduated in 2005 as a bioscience engineer in chemistry, option food technology and industrial microbiology, from the University of Louvain. For his M.S. thesis, he joined the Centre for Malting and Brewing Science (CMBS) to study continuous primary fermentation with immobilized yeast in a two-step system at pilot scale. After graduation, he started his Ph.D. project at CMBS under the supervision of Professor Freddy Delvaux, on "Feasibility of High Cell Density Fermentations for the Accelerated Production of Beer." He presented a part of his Ph.D. work at WBC 2008 in Honolulu, HI, and in September 2009, he graduated as a doctor in bioscience engineering. At present, he is working as a post-doctoral researcher at CMBS.*

## O-28

### **Impact of zinc on brewing yeast fermentation performance and beer quality**

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Zinc is an absolutely essential trace element for growth and fermentative metabolism in industrial yeast strains (De Nicola et al., *J. Inst. Brew.* 115:265-251, 2009; De Nicola et al., *Int. J. Wine Res.* 1:85-94, 2009; De Nicola and Walker, *Enzyme Microb. Technol.* 44:210-216, 2009). In malt wort, zinc is required at appropriate concentrations not only to allow good fermentation progress but also flavor congener formation by brewing yeast. Occasionally, wort zinc bioavailability can be compromised, and this negatively impacts yeast physiology, beer fermentation, and product quality. We studied zinc uptake, fermentation performance, and flavor congener formation under conditions simulating lager beer fermentations in laboratory-scale conical fermenters and in a brewery pilot plant. Zinc was rapidly (and completely) accumulated by yeast cells when pitched into wort with Zn concentrations up to 10 ppm. The best fermentations were observed when levels of zinc in malt wort ranged from 0.48 to 1.07 ppm. In pilot-plant fermentation experiments, Zn-enriched (or preconditioned) pitching yeast successfully fermented zinc-deficient worts. Additionally, Zn-preconditioned yeast appeared quite tolerant to stress imposed by elevated temperature and ethanol. Surprisingly, high concentrations of zinc (10 ppm) were not detrimental for the progress of fermentations in terms of ethanol yield and production kinetics, but these levels did augment the synthesis of higher alcohols and esters such as ethyl caproate and isoamyl acetate. We consider that the control of zinc bioavailability through intelligent Zn-supplementation strategies (either directly to wort or to yeast cells via preconditioning) plays an important role in dictating brewing yeast fermentation performance and product quality.

*Graeme Walker graduated with a B.S. degree in brewing and biochemistry in 1975 and completed his Ph.D. degree in yeast physiology (1978), both from Heriot Watt University, Edinburgh. His professional career has included Royal Society/NATO Postdoctoral Fellow at the Carlsberg Foundation, Copenhagen; lecturer (biochemistry) at Otago University, New Zealand; lecturer (biotechnology) at Dublin City University; visiting researcher at Case Western Reserve University in Cleveland, OH; senior lecturer (microbiology) at Dundee Institute of Technology; and reader (biotechnology) at the University of Abertay Dundee, Scotland. He is currently professor and divisional leader for biotechnology and forensic science at Abertay University, where he directs a yeast research group investigating growth, metabolism, and stress in industrial yeasts. He is an active member of the Institute of Brewing & Distilling and the American Society of Brewing Chemists. Graeme has published more than 100 articles in journals, books, and conference proceedings and has also authored the textbook *Yeast Physiology and Biotechnology* published by J. Wiley (1998). He acts in a consulting capacity for international brewing and biotechnology companies.*

## O-29

### **Xanthohumol, the main prenylflavonoid in hops, inhibits cholesteryl ester transfer protein activity and improves dyslipidemia**

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Hops (*Humulus lupulus* L.) have a variety of functions: antibacterial, hyperglycemic, antiallergenic properties, etc. Among these functions, we are now focusing on dyslipidemia, which is a major social issue. Cardiovascular risk, in particular, accounts for the major cause of death, and dyslipidemia is mainly responsible for cardiovascular risk compared with other risk factors such as hypertension and hyperglycemia. The inhibition of cholesteryl ester transfer protein (CETP) is expected as an anti-dyslipidemia factor, because high-density lipoprotein (HDL) is elevated in the inhibition of CETP. Thus, we searched compounds that inhibit CETP in hop extracts. As a result, we identified a unique CETP-inhibition compound, xanthohumol (XN). XN inhibits CETP activity dose dependently at a rate of 50% in 40 ppm XN. Also, XN is the strongest inhibitor in most other major hop compounds, humulone, and lupulone, and their derivatives. Moreover, XN shows a higher inhibition level than the compounds of naringenin chalcone, naringenin, isoxanthohumol, and *p*-coumaric acid, similar to XN in the XN biosynthetic pathway. These results suggest that XN inhibits CETP in a specific manner. After that we verified whether XN improves dyslipidemia in vivo. Rodents, mouse and rat, do not have CETP, but hamsters do, and simultaneously, they have a lipoprotein profile similar to humans. Therefore, we administered XN to hamsters via a high-fat diet. As a result, HDL cholesterol was elevated, and low-density lipoprotein (LDL) cholesterol was decreased. Moreover the atherogenic index (AI) drastically decreased. Lipoprotein containing apolipoproteinB, chylomicron, very low-density lipoprotein (VLDL) cholesterol, and LDL all were reduced. These results suggest XN improves the lipoprotein profile, and this leads to anti-atherosclerosis. We also searched other anti-dyslipidemia functions of XN. XN inhibited the synthesis of apolipoproteinB in the HepG2 cell line, human liver cells. And, XN suppressed the elevation of triacylglycerol in mice post-feeding. All this evidence suggests the hop compound XN is an anti-dyslipidemia agent.

*Hiroshi Hirata received an M.S. degree from the Department of Agricultural Applied Biochemistry, Shizuoka University. He assumed employment with Sapporo Breweries, Ltd. in April 2007 as a biological chemist in the Frontier Laboratories of Value Creation.*

## O-30

### **Tetra-hydro-iso- $\alpha$ -acid (THIAA), a reduced compound derived from hop bitter acid, contributes to the prevention of osteoporosis**

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Hop (*Humulus lupulus* L.), an essential ingredient of beer that provides the typical bitter taste, has a variety of functions: antibacterial, hyperglycemic, antiallergenic properties etc. Hop bitter acids such as alpha-acids are also reported to exert a wide range of effects on health function, both in vitro and in vivo, including the aforementioned mechanisms. In this study, we examined the effect of THIAA (tetra-hydro-iso-alpha-acid) on bone metabolism. THIAA is a light, stable reductant of iso-alpha-acid that is used as the bittering agent for food. Many developed countries have an aging population. As these populations age, the number of patients with osteoporosis is rising. Bedridden patients are at high risk of osteoporosis. The prevention of osteoporosis incurs many important problems. Osteoporosis develops

when the balance between bone formation and bone resorption is disturbed, and consequently, it is considered feasible to prevent osteoporosis by promoting bone formation and/or inhibiting bone resorption. We researched the inhibition of osteoclastic bone resorption. We used rat primary osteoclast cells and assayed the pit formation, which was quantified by scanning electron microscopy coupled with image analysis. Results revealed that THIAA decreased the assimilation trace number and areas. This indicates that THIAA performed a drastic dose-dependent decrease in the bone resorption pit at concentrations of 0.1, 1, and 10 nmol/L. Next, we researched the influence of THIAA on senile osteoporosis. We utilized a well-established murine model of senile osteoporosis, the P6 strain of senescence-accelerated mice (SAMP6), to investigate fracture healing in aged osteoporotic bone. The experimental mice were given a diet containing 0.05, 0.2, and 0.5% THIAA for 8 weeks (12-20 week old mice). Food intakes did not differ among the groups throughout the experiment. On the other hand, THIAA intakes significantly improved the bone mineral density (BMD) of the distal femur compared with the control. These results show that THIAA is effective in the prevention of senile osteoporosis, and one of its mechanisms inhibits bone resorption.

*Moeko Ozaki received an M.S. degree from the Department of Pharmacy, Hiroshima University. She found employment with Sapporo Breweries, Ltd. in April 2008 as a pharmaceutical scientist in the Frontier Laboratories of Value Creation.*

#### O-31

##### **The glyceamic index of beverages—A critical review**

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Jenkins introduced the concept of the glyceamic index in the 1970s. In the following years diseases caused by poor or unbalanced diets have developed into severe problems in the Western world. The growing number of patients affected by diabetes mellitus type II especially seems to be directly related to the amount and type of carbohydrates consumed in the daily diet. The glyceamic index classifies carbohydrates with regard to their individual resorption time from a consumed food. The resulting postprandial glucose levels are compared to those measured after consumption of a reference food, notably glucose or white bread. The glyceamic index of glucose is set at 100, the index of sucrose is set at 66, and the index of fructose is set at only 20. In this paper the concept of the glyceamic index is explained. Measured glyceamic index values for different types of beverages, both alcoholic and non-alcoholic, are compared with the amounts of sugar present in these beverages. From the presented values a mathematical possibility for calculating the glyceamic index of a beverage is shown, and the significance of the glyceamic index for alternative malt-based beverages is discussed. The glyceamic index of beer is often said to be 110, and thus, one of the highest present in food. This statement can be found on a lot of nutrition-related web pages. The origin of these statements is described, and more realistic data will be provided. The glyceamic index of beers seems to be more likely around 70. Additionally the glyceamic load, an index comparing the glyceamic index of a food's carbohydrates with the amount of sugar present in a portion, will be explained. Using the glyceamic load the effect of beer and, thus, of the carbohydrates present in beer can be shown to be of no great importance in a balanced diet.

*Moritz Krahl was born in 1980 in Schwetzingen, Germany. In 2000 he started his education in brewing and beverage science at the Technische Universität München in Germany. In 2004 he graduated with a B.S. degree and in 2005 with a Dipl. Ing. degree. Since 2005 he has been working as a Ph.D. student at the Institute for Brewing and Beverage Technology in Weihenstephan.*

#### O-32

##### **Antioxidants in beer: Better than those from wine?**

CHARLES W. BAMFORTH (1), Michael Dipietro (1)  
(1) University of California

The red wine lobby has widely touted the polyphenols in their product, claiming remarkable benefits for the drinker arising from their very high levels and very high potency in this beverage. The question is: do they actually reach the parts that matter? Might there even be too much? The story of antioxidants and the body will be reviewed here, with recent data from this laboratory that suggests that, molecule-by-molecule, the antioxidants from beer may actually be more useful than those from red wine.

*Charlie Bamforth is the Anheuser-Busch Endowed Professor of Malting & Brewing Sciences at UCD. He has been part of the brewing industry for more than 31 years. He is the former deputy director-general of Brewing Research International and research manager and quality assurance manager of Bass Brewers. He is a special professor in the School of Biosciences at the University of Nottingham, England, and was previously visiting professor of brewing at Heriot-Watt University in Scotland. Charlie is a Fellow of the Institute of Brewing & Distilling, Society of Biology, and International Academy of Food Science and Technology. Charlie is editor in chief of the Journal of the American Society of Brewing Chemists, is on the editorial boards of several other journals, and has published innumerable papers, articles, and books on beer and brewing, as well as written prolifically on soccer.*

#### O-33

##### **Kinetic modeling of hop acid isomerization: Insight for the enhancement of bitterness yields during wort boiling**

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(1) International Centre for Brewing & Distilling, Edinburgh, U.K.

The alpha-acid isomerization yields during wort boiling rarely exceed 50%, resulting in a substantial loss of bittering potential. These losses have been variously attributed to precipitation of alpha-acids in the trub, with or without the mediation of metal cations. Alternatively, there can be challenges in terms of the solubilization and dispersion of the alpha-acids after hop addition due to their limited solubility in acidic media. In this study, a general scheme defining the various pathways that may impact isomerization was defined, and a range of parameters was estimated for all of the reaction paths. Subsequent systematic kinetic analysis of these pathways, using power law analysis and simulation, clearly indicated that both the relative rates both of dissolution and of precipitation of the alpha-acids dominated the final yields of iso-alpha-acids, and many of the models replicated the typical isomerization yields observed in practice. Based on simulations of varying complexity, several options for enhancing isomerization yields presented themselves, not the least of which is the possibility of making multiple additions of smaller quantities of alpha-acids throughout the boil to overcome limiting solubility in acidic, boiling wort. These simulations also highlighted the need for more detailed determination of metal cation-alpha-acid complex dissociation constants and the enhancement of isomerization rates in the presence of metal cations.

*Paul Hughes is a chemist by training, and after a short spell with the Health and Safety Executive, he joined the Brewing Research Foundation in 1990. After nine years in various roles, Paul moved to Heineken in the Netherlands, coordinating a range of projects on beer quality and safety/integrity. In 2005 he moved to Heriot-Watt University to take up the position of professor of brewing and assumed responsibility for the International Centre for Brewing and Distilling in 2006. Paul has a wide range of research interests, with a focus on consumer-product interaction, hop chemistry, and deriving value from waste streams. He is also becoming increasingly involved in research focusing on whisky maturation, blending, and sustainability issues. In addition to a Ph.D. degree in chemistry, Paul holds an MBA and diploma of brewing from the*

*Institute of Brewing and Distilling. He is also a past winner of the IBD Cambridge Prize and the ASBC Eric Kneen Memorial Award. He is the author of various papers, patents, and book chapters and is currently working on his second book.*

#### O-34

##### **Resolution and properties of isomerized ( $\pm$ )-tetrahydrocolumulone**

PATRICK L. TING (1), Jason Pratt (1), David Ryder (1)

(1) MillerCoors

Tetrahydroiso- $\alpha$ -acids have been demonstrated to have more benefits in brewing than their analogous iso- $\alpha$ -acids,  $\rho$ -iso- $\alpha$ -acids, and hexahydroiso- $\alpha$ -acids when comparing bitter intensity, light stability, flavor stability, foam, and antimicrobial activity. Tetrahydroiso- $\alpha$ -acids can be prepared from either  $\alpha$ -acids or  $\beta$ -acids. Both methods produce identical molecules of tetrahydroiso- $\alpha$ -acids that only differ in their stereoisomers. Tetrahydroiso- $\alpha$ -acids prepared from  $\alpha$ -acids are optically active compounds due to the natural structure of  $\alpha$ -acids (asymmetric molecules). However, tetrahydroiso- $\alpha$ -acids prepared from  $\beta$ -acids (disymmetric molecules) are a racemic mixture (pairs of mirror-image molecules or equal enantiomers) with no optical activity. Their stereochemistry and physiological properties (chiral recognition) is important because of their potential flavor contribution in beer. A variety of optical isomers have been described as having different odor qualities and/or different odor intensities. Such considerations prompted us to initiate this study, which aims to resolve the gram quantity of a pair of ( $\pm$ )-tetrahydrocolumulone (one representative of tetrahydro- $\alpha$ -acids) and to assess each isomerized enantiomer for bitter quality perception, foam, and antimicrobial activity.

*Patrick L. Ting received his Ph.D. degree in organic chemistry from the University of North Texas and was a post-doctoral fellow at Southwestern Medical School, University of Texas, Dallas. He started his hop research at Miller Brewing Company (now MillerCoors) in Milwaukee, WI, in 1978. At Miller, Patrick touched all aspects of hop chemistry in his 31-year career, including novel preparation and purification of hop bittering compounds, the mechanism of hop flavor formation, and identification of hop compounds for improving flavor stability in beer. He and his colleagues developed all of the hop processes for the Watertown Hops Company. Patrick was awarded 17 U.S. patents and also won the 2009 American Chemical Society-Milwaukee Section Award and 2008 ASBC Eric Kneen Memorial Award for his work. Patrick is an active member of ACS, ASBC, the U.S. Hop Research Council (HRC), and the International Subcommittee for Hop Standards.*

#### O-35

##### **Influence of hopping technology on harmony of bitterness**

STEFAN HANKE (1), Thomas Becker (1), Werner Back (1), Martin Krottenthaler (1)

(1) Lehrstuhl fuer Brau- und Getraenketechologie, Freising, Germany

Hop-derived bitterness is one of the most distinct sensory properties of beer. Bitterness and harmony are also important for drinkability. Some brewers often see hops only as a provider of  $\alpha$ -acids, and the usage of downstream products is very popular in the international brewing business. However, besides  $\alpha$ -acids, hops also contain other constituents that can contribute to the taste of a beer. The input of these substances can be controlled by the hop product used. Different hopping technologies were applied to test the influence of hop product and time of hop addition on the harmony of beer. The applied hop products were a CO<sub>2</sub> hop extract of Hallertau Taurus and pellets of several German aroma hop varieties. These hop products were used as single and combined bitterness providers at different stages of wort boiling. In these trials it could be shown that the use of aroma hop pellets positively impacted the harmony of beer. A dosage of 50% of aroma pellets at the beginning of wort boiling increased the harmony significantly. A shift in the aroma pellet dosage to the middle of boiling resulted in a slightly lower evaluation of

harmony. If an additional hop dosage is applied at the end of wort boiling, the harmony would be improved compared to pure CO<sub>2</sub> extract and a hop addition at the middle of boiling. The evaluation of the data showed that the residual extract of beer positively impacted harmonic bitterness perception. The role of linalool as an indicator substance for hop aroma was confirmed. Addition of hop pellets leads to an input of non-specific resins and polyphenols in addition to bitter and aroma compounds. It could be shown that adapted hopping technology can create a pleasant bitterness and desired hoppy flavor that is a great contribution to the harmony of bitterness and the drinkability of beer.

*Stefan Hanke was born in 1980. From November 1999 to July 2004, he studied brewing science and beverage technology at Munich Technical University (Weihenstephan), graduating as an engineer with a Dipl.-Ing. degree. In 2010 he finished his Ph.D. degree, which dealt with the influence of hopping technology on the harmony of beer. During his studies, he worked for and received practical training at several German brewing and malting companies. Since September 2004 he has been a scientific employee at the Lehrstuhl fuer Technologie der Brauerei I, Freising-Weihenstephan, Germany (Professor Back). From December 2006 until May 2007 he headed the institute's Small Scale and Pilot Scale Brewery Department. Since May 2007 he has been responsible for the GC/HPLC Laboratory of the institute. His main research topics are the influence of hops on beer drinkability and the influence of beer matrix on bitter taste. Since May 2009 he has been the head of the GC/HPLC Laboratory of the Chair for Brewing and Beverage Technology (Professor Becker).*

#### O-36

##### **Influence of hop pre-treatment before dosage on the yield of isohumulones and resulting beer quality**

SEBASTIAN KAPPLER (1), Udo Kattein (1), Thomas Becker (1), Martin Krottenthaler (1)

(1) Technische Universitaet Muenchen, Freising-Weihenstephan, Germany

Iso-alpha-acids are the major contributor to the bitter perception in beer. They contribute to over 85% to the overall bitterness of traditional beers. In the brewing process, however, only about 30% of the alpha-acids present in hops are isomerized and transferred into the finished beer. Although hop products are relatively inexpensive, many breweries try to reduce costs through savings in hopping technology. One way to reduce costs by increasing the yield of isohumulones is a pre-treatment of the hop product before dosing it into boiling wort. Pre-treating with high temperature or pH can result in several problems. The bitterness of the resulting beer often is described as harsh with a long-lasting aftertaste. This phenomenon may be caused by a large amount of unspecific degradation products of alpha-, beta-, and iso-alpha-acids in the resulting beer. By varying the parameters of hop pre-treatment the amounts of these degradation products can be reduced, and thereby, the quality of beer bitterness can be increased. In this work the influence of various technologies and parameters for treatment of hop products were evaluated. Pilot-scale trials were done to evaluate the influence of various treatment technologies on sensorial and analytical attributes, as well as on the behavior during beer aging. Particular attention is paid to the bitterness profiles of fresh and forced-aged beers. All trials were done in comparison with common brewed beers. The results presented in this paper provide a better understanding of the conversions taking place during the brewing process and their influence on beer quality. Suitable approaches toward an improved yield of bitter acids and improved bitter quality are shown!

*Sebastian Kappler received a Dipl.-Ing. degree in brewing and beverage technology from Technische Universitaet Muenchen in 2008. He began his employment with the Augustiner-Wagner brewery in Munich as an apprentice to a brewer and maltster in 2000. After achieving the position of assistant, he started his studies on brewing science at the Technische*

Universitaet Muenchen. Since May 2008 he has been working as a scientific employee at the Chair for Brewing and Beverage Technology in Weihenstephan. The topic for his doctoral thesis is the evaluation of the factors affecting the yield of isohumulones during preparation of wort.

**O-37**

**Microbiological QA—Classification perplexity with modern packaging**

ROLAND FOLZ (1)

(1) VLB-Berlin, Berlin, Germany

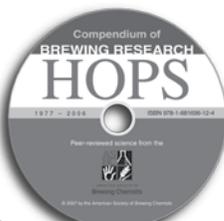
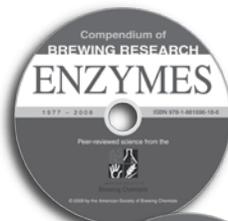
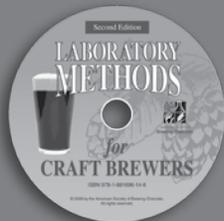
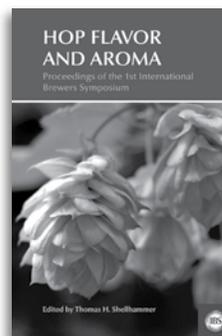
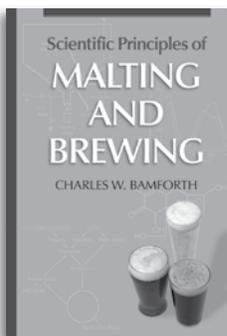
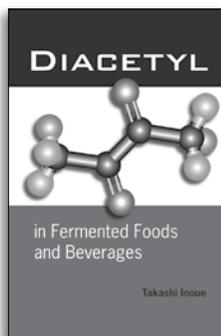
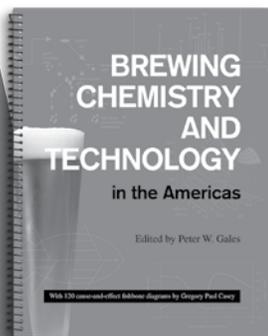
A study to investigate the growth behavior of different microorganisms in beer-filled PET bottles was carried out at VLB Berlin. Five microorganisms with different properties were chosen and used for standardized artificial contamination of beer. Growth behavior was tested in PET bottles with varying barrier properties that showed different behaviors concerning the permeation of gases. This procedure was supposed to test not only the effect on obligate and potential beer spoilage bacteria, but possibly even on harmless flora usually not monitored by QA. As a reference, glass bottles with crown corks were used. The level of carbon dioxide retention and especially oxygen ingress had a significant influence on the multiplication rate of most of the tested microorganisms. *Saccharomyces diastaticus*, an obligate beer spoilage organism, was able to reproduce faster the higher the oxygen uptake was. With a certain amount of oxygen present in the fluid, oxygen-dependent organisms such as *Gluconobacter oxydans* and *Micrococcus kristinae* were able to reproduce in beer, and therefore, microorganisms that have been referred to as being harmless turned into product-spoiling microorganisms. However, these microorganisms usually are not monitored in microbiological QA. Connected to the

growth of microorganisms and the uptake of oxygen, a faster increase in turbidity levels could be observed as well. The results showed a good relationship, with the possibility of making use of turbidity measurement in the microbiological shelf-life examination. Packaging-wise, the study showed that for microbiological stability of beer in innovative packaging, permeation issues affected by a well-working oxygen barrier can be of high importance. With the help of the barrier-enhancing technologies available on the market today, it is possible to achieve a suppression of microbiological growth comparable to glass bottles closed with crown corks. On the side of microbiological QA, the study shows that the industry is facing new demands with the rising diversity of packaging being made available to the customer that make it impossible to rely on traditional classification methods.

*Roland Folz apprenticed as a brewer and maltster at the Beck's brewery in Bremen, Germany. After working another year for the Beck's brewery, he started his studies in Berlin and received a diploma engineer degree in brewing technology from the Technical University, Berlin. After graduation, he worked as head of the Technical Department/Production at the Preussen Pils brewery in Pritzwalk, Germany, for two years. In October 2006, he started at VLB-Berlin as a global consultant for brewing technology, he worked for the Engineering and Packaging Department as the specialist for filling, packaging, and PET topics. In addition to his consulting practice, he is involved in teaching and research projects and manages the internationalization of VLB. Since autumn 2008, Roland has been the head of the Brewing & Beverage Technology and Applications Department at VLB-Berlin. This department includes the education and teaching part of VLB, as well as the research activities regarding technological topics, global consulting, analytics, and services.*

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## O-38

### Technologies, tools, and challenges for packaging beer in PET

LORINDA (LORI) Y. YODER (1)

(1) Plastic Technologies, Inc., Holland, OH

Although more than 50 barrier solutions are commercially available for plastic bottles through passive and active barrier blends, multilayer, and coating technologies, PET containers have yet to penetrate the single-serve beer market beyond regional or limited venue distribution. While oxygen ingress was historically the primary concern in adopting plastic packaging, today several solutions are available to address its permeation. What continues to challenge bottle developers and brewers is how to retain carbon dioxide within the bottle in order to meet shelf-life requirements. This presentation will explore methods, materials, and equipment at the forefront of barrier technologies and demonstrate how the predictive M-RULE container performance model and analytical testing methods can be utilized to determine the most suitable barrier solution for a specific product. The M-RULE model has been used to predict package permeation under a variety of environmental and filling conditions to demonstrate their impact on shelf life. Additionally, virtual prototyping software and FEA solutions allow the impact of volume expansion and container creep on bottle design and orientation to be modeled. These model results will be related to the actual analytical test methods and tools employed to determine the impact of O<sub>2</sub> and CO<sub>2</sub> on container shelf life. The testing methods studied include both destructive and non-destructive O<sub>2</sub> and CO<sub>2</sub> permeation measurement techniques along with oxygen scavenging capacity tests. As the analytical tools are developed and refined, the ultimate proof of PET container performance will continue to be taste panel testing. Although numerous technical solutions exist for PET beer bottles and hopes remain high for barrier alternatives, no single technology has been shown to satisfy the broad needs of brewers and consumers. The barrier PET bottle stakeholders (PET resin manufacturers, barrier and additive producers, equipment manufacturers, and converters) are eager for the opportunity for the widespread commercial adoption of beer in plastic, although brewers and consumers remain cautious.

*Lorinda (Lori) Yoder has more than 15 years of experience in the plastic packaging industry, including preform and container design, orientation stretch-blow molding processing, container performance testing, materials, and barrier technologies development. She joined Plastic Technologies, Inc. (PTI) in 1997, where her current title is director, materials applications. Prior to joining PTI, Lori was a technical specialist responsible for antibiotic fermentation processes at Eli Lilly and Co. She received her B.S. degree in chemical engineering from The University of Toledo, graduating cum laude in 1993, and went on to receive her M.S. degree in chemical engineering from The Ohio State University in 1995. Additionally, Lori has a patent pending for a "Method of Blowing a Bottle from Bioresins."*

## O-39

### Beverage and package quality—Two inseparable key parameters in the modern quality control of bottled beverages

JOHANN ANGRES (1)

(1) Steinfurth, Inc., Marietta, GA

The last couple of years have been characterized by dramatically changing challenges in the area of QC of beverages. In addition to new beverage types and tastes, the rapidly transforming world of beverage packages offers a wide range of benefits for the bottler and consumer, but it is becoming at the same time a more and more critical impact factor for consumer-tasted beverage quality. Growing price pressure and cost competition under the packaging supplier is boosting the demand for standardized test methods and quality checks for packaging material during development and evaluation of new packages and bottling of beverages. Standardized test methods and procedures as designed and recommended by the ISBT and ASBC set today's global guidelines

for modern quality control of beverages. Along with the standardized quality guidelines for beverages and beverage packages, the requirements for modern tools and instruments to perform quality control tasks has changed. Measuring beverage parameters in combination with the packaging evaluation, knowledge about packaging performance, and evaluation of packaging dimensions under real working conditions are just few important steps in the evaluation of beverage quality and the key to customer satisfaction. The presentation discusses the importance of combined quality testing on beverages and beverage packages and looks at this important topic from the view of new instrument development and a highly sophisticated packaging laboratory.

*Johann Angres received a bachelor of technology diploma from Senior Technical College in Bochum, Germany. He began employment with Steinfurth in October 1992 as a development engineer. Since January 1996, he has functioned as technical director, with a focus on instrument production and technical customer support. He is serving Steinfurth on the international level as managing director, with a focus on new business development and became president of Steinfurth, Inc. in Georgia in 2005.*

## O-40

### A novel approach to brew alcohol-free beer

Zongcui Yue (1), GUANGTIAN ZHOU (1), Mengmeng Huang (1), Zhumao Jiang (2)

(1) Shandong Institute of Light Industry, Jinan, China; (2) Yantai University, Yantai, China

In the present report, a novel approach to producing alcohol-free beer is described, and the process mainly includes the following steps: special mashing, limited fermentation, and vacuum distillation to prepare "concentrated extract." Breweries can produce alcohol-free beer using the extract. Preparing the alcohol-free concentrate is the first step, and the step includes the following process. The wort of 11.0°P was prepared by the infusion mashing method with barley malts (70%) and wheat malts (30%). Top-fermenting yeast no. 303 was pitched, and the fermentation temperature was controlled between 17 and 20°C. As soon as the extract of fermented medium was decreased by about 25%, the temperature of fermented medium was quickly reduced to 0°C. After keeping 2 days at this temperature, the "young beer" was degassed, and the yeast was separated and then transferred to the "continuous vacuum evaporator." The young beer was concentrated under vacuum with a degree of 0.06 MPa in order to reduce the loss of flavor substances. After the extract was concentrated as high as 60°P, the concentrate could be used conveniently as raw material to produce alcohol-free beer for breweries. Second, the concentrate was used as raw material for preparing alcohol-free beer. The alcohol-free concentrate was diluted to 4.0°P using desoxygenated water and stored in a storage tank at 1°C. CO<sub>2</sub> was added to a level of 6.0 g/L. Then 8~10% of normal matured beer with an alcohol level of about 5.0% (v/v) was added. After keeping for 2~3 days under an appropriate pressure at a low temperature, the beer had a nice zest. Next, the alcohol-free beer was filtered and bottled or canned. Analysis of the alcohol-free beer showed the following results: alcohol, 0.5% (v/v); color, 7.0 EBC; TBA value, 1.05; ratio of alcohols to esters, 1.08. Compared with alcohol-free beer produced with traditional techniques, our novel approach, including the heat method, reverse osmosis, and limited fermentation, provides a useful technique for producing alcohol-free beer with the characteristics of normal beer and satisfactory stability.

*Guangtian Zhou is a professor in bioengineering and director of the China-Germany Brewing Technical Service Center in the Shandong Institute of Light Industry. Guangtian received his B.S. degree in bioengineering from Shandong Institute of Light Industry in Jinan, China. He worked in the Jinan Beer Group from 1982 to 1987 as a brewer. From 1987 until 1988, he studied at Doemens Brewing Akademie in Munich, Germany. He then worked in the Jinan Beer Group as a chief engineer. Since 1994, he has been working in the Shandong Institute of Light Industry in Jinan, China, as a professor. He is now the director of the*

China-Germany Brewing Technical Service Center. Guangtian is also an editor of China Brewing, a famous journal in China, and a council member of the Microorganism Association of Shandong, China.

#### O-41

##### Ingredients and energy from brewer's spent grain

ANNIKA WILHELMSON (1), Piritta Niemi (1), Juhani Sibakov (1), Pekka Lehtinen (1), Laura Flander (1), Raija-Liisa Heiniö (1), Kaarina Viljanen (1), Veli-Pekka Heiskanen (1), Niklas Von Weymarn (1), Johanna Buchert (1)

(1) VTT Technical Research Centre of Finland, Espoo, Finland

Brewer's spent grain (BSG) is the insoluble cereal residue that is separated from the mash before fermentation. It is estimated that the annual production of BSG is around 30–35 million metric tons worldwide. So far, BSG has been mostly used as animal feed. However, in areas where feed consumption is limited, it may become waste. BSG is an interesting raw material for use as a food ingredient. It has a high content of dietary fiber, proteins, and potential phenolic antioxidants. In this study, ultra-fine milling, air classification, and enzymatic hydrolysis were applied with the aim of producing food ingredients from BSG. BSG was also used as an ingredient in bread. BSG gave dark color to the bread, with slightly increased bitterness and a somewhat stick-like mouthfeel. Various dry-milling techniques were tested for BSG. Ultra-fine milling resulted in a very fine powder that enabled the addition of BSG even to drinks. The mouthfeel of ultrafine BSG was good in beverages that were consumed cold. When beverages were served at room temperature, the taste was slightly bitter, concurrently reducing the fresh perception of the beverages. The removal of bitter-tasting compounds would improve the sensory properties of BSG. Enzymes can be used for fractionation and selective extraction of high-value compounds from BSG. An enzyme-aided process for fractionation of BSG to peptides and xylo-oligosaccharides was applied with the aim of producing novel ingredients. The xylo-oligosaccharide fraction contained ferulic acid-substituted oligosaccharides. When freeze-dried, this fraction had a white color and an acceptable taste, and using a liposome model, it was shown to have antioxidative properties. This fraction could possibly be used as a natural, fiber-rich ingredient. Another attractive application for BSG or the fraction remaining after removal of high-value ingredients is energy production. Combustion of BSG offers a means of disposing of the material, as well as a means of producing energy in the form of heat or electricity. For both purposes either a small- or medium-scale local boiler or alternatively co-firing in a larger boiler is an option. The energy balance has to be calculated case by case. Dewatering and combustion technology is widely available, and some breweries are already applying this route. The worldwide effort to increase the proportion of renewable energy sources will probably increase the use of BSG as an energy source. The question is which valuable compounds could be extracted from BSG before it is used for energy production.

*Annika Wilhelmson, D.S. (Tech.), is the customer manager for the malting and brewing sector at the VTT Technical Research Centre of Finland. Before her current position, she worked as a senior research scientist at VTT. She graduated from Helsinki University of Technology in 1992. From 1994 to 1995, she worked at the ICBP at Heriot-Watt University as a visiting research student. Her research topics have included genetic engineering of plants, barley and malt quality, and the biochemistry of malting and mashing. She is currently chair of the Malting Barley Genetics and Physiology Subgroup of the EBC Brewing Science Group.*

#### O-42

##### Recycling and refining of alcohol from waste beer

ZHUMAO JIANG (1), Mengmeng Huang (2), Xiaolei Dong (2), Guangtian Zhou (2)

(1) Yantai University, Yantai, China; (2) Shandong Institute of Light Industry, Jinan, China

The yeast separated from the waste yeast slush is utilized to produce protein feed in most breweries of China, while the rest of the waste beer is abandoned as waste. The discharge of waste beer not only is a waste of resources, but also pollutes the environment. A technique is described in the present study for recycling and refining alcohol from waste beer. The content of the alcohol in waste beer is about 5.0% (v/v). A continuous rectification column with 28 plates was used to receive crude alcohol. The waste beer was fed on the twenty-second plate at a flow rate of 11.1 kg/min under 0.12 MPa of pressure. The temperature of the top plate of the column was 85°C. Then, the crude alcohol was refined by a 50-L batch distillation column, since the crude alcohol produced an astringent bitterness. Meanwhile, the technical parameters of the batch distillation column were optimized by Orthogonal Design Assistant V3.0. The technical parameters were as follows: initial alcohol concentration, 60%; reflux ratio, 3; foreshot, 3%; and feints, 14%. The refined alcohol was treated by 0.4% (m/m) activated carbon for 24 hr and then distilled by the atmospheric rectifier. The analysis of the end-product meets the Chinese standard of edible alcohol. Our present study provides a simple and economical approach to producing alcohol for breweries.

*Jiang Zhumao, associate professor of food fermentation engineering, is the director of the Food fermentation Engineering Survey Section at Yantai University Science and Engineering College of Chemistry and Biology. Zhumao received his bachelor degree in food engineering from Wuxi Institute of Light Industry (now Jiangnan University) in 1982. He then worked in Huaiyin Industrial College (now Huaiyin Institute of Technology) to teach and research food microbiology and brewing technology. Since 1995, he has been working in Yantai University Science and Engineering College of Chemistry and Biology, taught and researched in turn fermentation technology and bioengineering equipment in bioengineering major, fermented food technology, and food additives in food science and engineering.*

#### P-43

##### A rapid and sensitive genetic identification method for detecting beer-spoilage bacteria and wild yeast

YONG WU (1), Manami Saha (1), Beena Lee (1), Lily Nan (1), Handy Yowanto (1)

(1) Beckman Coulter, Inc., Brea, CA

Although beer is generally considered a beverage with low microbiological activity, many species of microorganisms have been reported to spoil beer. To improve the hygienic status of breweries and perform quality control of beer products, it is important to rapidly detect the presence of microorganisms and assess their spoilage potential. Traditional culturing techniques, aerobic and anaerobic, are the most common methods currently used. However, these methods generally take a few days to detect aerobic bacteria and up to a few weeks to detect anaerobic bacteria. Another drawback of culturing methods is that they cannot examine the capacity of microorganisms to cause beer spoilage. Therefore, a new approach that can rapidly and reliably detect microorganisms and determine their beer-spoilage potential has long been desired by the brewing industry. Here we present a multiplexed capillary electrophoresis method that can simultaneously identify six major genera of bacteria associated with beer-spoilage and assess their potential to spoil beer by detecting five hop-resistant genes. Equipped with an eight-channel capillary array, the GenomeLab GeXP genetic analysis system is able to identify beer-spoilage bacteria and determine their beer-spoilage potential from sample collection to data report within 24 hr. This multiplexed approach also detects more than 10 wild yeast

species that are commonly found during the brewing process and are associated with reduced beer quality. The capacity to detect all major beer-spoilage bacterial genera and wild yeast, along with the information to determine their beer-spoilage potential in a timely manner, will greatly help brewers to improve and accelerate the quality control process and significantly reduce the storage time, which results in lowering inventory costs.

*Yong Wu obtained his Ph.D. degree in molecular and cellular biology at the University of Florida in Gainesville in 2000. He joined the GenomeLab Group at Beckman Coulter in 2004 and is currently the staff scientist for developing applications on the multiplexed biomarker detection platform, the GenomeLab GeXP genetic analysis system. As an active ASBC member, Yong has been working on developing a rapid and sensitive QC method for brewers that can simultaneously detect beer-spoilage bacteria, hop-resistant markers, and wild yeast in a cost-effective manner.*

#### **P-44**

##### **A role for the COMPASS complex as determinant of brewing yeast fermentation performance?**

BRIAN R. GIBSON (1), Jari J. Rautio (2), Virve Vidgren (1), John Londesborough (1)

(1) VTT, Espoo, Finland; (2) PlexPress, Helsinki, Finland

COMPASS is a complex of proteins known to regulate the activity of a number of genes critical to yeast fermentation performance. These genes, including *FLO* and *MAL* genes, are located in the subtelomeric regions of chromosomes, where COMPASS exerts its influence via histone methylation. COMPASS has previously been implicated in control of *FLO* and *MAL* expression (Dietvorst and Brandt, *Yeast* 25:891, 2008). The aim of this investigation was to determine the relationship between COMPASS activity and the fermentation performance of a lager brewing yeast and, furthermore, to determine whether transcriptional analysis of genes encoding individual COMPASS protein subunits can be used as predictive biomarkers of fermentation performance. Tall tube fermentations (2 L) at either 15 or 25°C were carried out with a lager brewing yeast strain, and transcriptional profiling of COMPASS subunit genes (*SET1*, *SXD1*, *SDC1*, *SWC2*, *BRE2*, *SHG1*, *SPP1*) and subtelomeric genes responsible for maltose assimilation and flocculation was conducted using TRAC (transcriptional analysis with aid of affinity capture). Increasing gravity of the wort resulted in increased yeast biomass production, while the maximum rate of decrease in apparent extract (AE; 0.28 h<sup>-1</sup>) remained unchanged. Wort was, however, fermented more completely at 15°C (AE = 2.4) than at 25°C (AE = 4.2). Activation of the maltose transport and catabolism genes *MAL31* and *MAL32* occurred approximately 12 hours after pitching, indicating that their activity may be related to factors other than monosaccharide depletion. A decrease in the transcriptional activity of these genes in both fermentation systems coincided with increased transcription of the COMPASS gene *SET1*, suggesting both a role for this gene in transcriptional silencing of subtelomeric genes and its potential as a biomarker of yeast fermentation performance. The transcriptional activity of *SET1* was most pronounced at very high gravity and corresponded to a similarly pronounced deactivation of the *MAL* genes relative to the 15°C fermentation. Results indicate that COMPASS activity influences assimilation of maltose and, consequently, the residual sugar content at the end of fermentation. Improved fermentation performance may be possible with yeast strains in which the influence of COMPASS is attenuated. It is suggested that *SET1* may be used as a determinant of COMPASS activity.

*Brian Gibson was awarded a Ph.D. degree from University College Dublin, Ireland, in 2004, where he had specialized in fungal stress responses. On completion of his studies he joined Katherine Smart's research group, initially at Oxford Brookes University and later at Nottingham University, U.K., where his research covered a range of*

*subjects, including brewing yeast stress responses, yeast transcriptomics during industrial fermentation, genetic stability of brewing yeast, and molecular identification of brewery contaminants. Since 2009 he has been employed as a senior research scientist and project manager at VTT, Finland, with responsibility for yeast physiology and fermentation research.*

#### **P-45**

##### **Analysis of volatile components in beer using automated solid-phase microextraction (SPME) and high-speed GC×GC-TOFMS**

MARK LIBARDONI (1), Cory Fix (1)

(1) LECO Corporation

The analysis of trace level components in beer is important for the determination of "off" flavor such as furfural, furfural-ethyl-ether, and various disulfides, in addition to the uniformity of the product. In this work, automated solid-phase micro-extraction (SPME) is coupled with high-speed two-dimensional gas chromatography (high-speed GC×GC), utilizing time of flight mass spectrometry (TOFMS) as the mass selective detector for the trace analysis of light- and heat-abused beer. The results for different SPME fibers, as well as sample extraction techniques, will be presented, along with the methodology for high-speed GC×GC analysis. Statistically, the results of complex sample analysis by high-speed GC are overlapping peaks with minimal resolution. However, the increased chromatographic resolution, increased detection limits, and favorable peak capacity makes high-speed GC×GC a powerful analytical tool for the trace-level detection of off-flavor components in complex samples such as beer. When high-speed GC×GC separations are employed, the generated peak widths are significantly narrower compared with traditional GC, thus requiring a high-speed data acquisition system. In addition to the results of the beer samples analyzed, advanced data processing with statistical analysis will be shown.

*Mark Libardoni received his B.S. degree in chemistry from California State University. He attended graduate school at the University of Michigan in Ann Arbor, where he received a Ph.D. degree in chemistry while studying under the direction of Richard Sacks. Mark began employment with LECO Corporation in 2005 and currently holds the position of technical director, application laboratories and advanced research. Mark's research interests include trace level detection of volatile and semivolatile components, detection of biomarkers for precancerous states, as well as novel applications for high-speed and multidimensional gas chromatography.*

#### **P-46**

##### **Application of ultra performance liquid chromatography for the determination of amino acids in wort and beer**

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(1) Canadian Grain Commission, Winnipeg, MB, Canada

Amino acids are essential for good fermentation performance and also influence the flavor profile of the finished beer. Ensuring an adequate supply of amino acids for yeast nutrition has become even more critical given the increased popularity of high-gravity and high-adjunct brewing practices. The importance of amino acids in wort has led to many studies investigating the development of amino acids during malting, their extraction during mashing, and assimilation by yeast during fermentation. However, there is considerable inconsistency with respect to the results reported for amino acid levels in malt-derived worts and beers. The analysis of individual amino acids is most often conducted by high-performance liquid chromatography (HPLC). These methods exhibit long analysis times that result in low sample throughput. Other separation techniques, such as gas chromatography (GC) techniques, involve complex sample preparation and cleanup procedures. The Waters ACQUITY UPLC system employs columns packed with 1.7- $\mu$ m particles and higher flow rates than conventional HPLC, resulting in reduced

analysis times with superior resolution and sensitivity. The technique is ideal for the analysis of free amino acids in wort and beer. The pre-column derivatization employs an ultra-violet chromophore that is specific for amino acids, and samples require no additional preparation or cleanup other than filtration and dilution. Full profiles of 20 amino acids can be separated in under 8 min, representing a 10-fold enhancement over traditional HPLC. Good repeatability has been achieved with this technique when applied to the analysis of amino acids in wort and beer, with coefficients of variation for individual amino acids ranging from 2.5 to 11.6%. Samples of AC Metcalfe, CDC Copeland, and Legacy malt from the 2009 Canadian harvest were analyzed for free amino acids in Congress wort. No significant difference in the relative level of amino acids among varieties was observed. However, correlations of levels of individual amino acids with FAN ranged from 0.23 to 0.88, demonstrating that considerable variation in amino acid levels can exist that cannot be identified by the determination of FAN alone. Levels of individual amino acids have implications for fermentation and beer flavor.

*Aaron MacLeod is the analytical malting technician in the Applied Barley Research Unit of the Grain Research Laboratory in Winnipeg, MB, Canada. The unit conducts research on factors affecting malting barley quality and measurement methods. Aaron received his B.S. degree in chemistry from the University of Western Ontario in 2004. He is currently a member of several ASBC Technical Subcommittees and actively involved in the collaborative study of numerous methods. Aaron also serves as secretary of the Canadian Prairie Section of AACC International.*

#### **P-47**

##### **Are magnetic fields a technical opportunity to influence the structure of CO<sub>2</sub> nanobubbles responsible for primary gushing in beer?**

SYLVIE M. DECKERS (1), Kurt Gebruers (1), Yannick Lorgouilloux (2), Geert Baggerman (3), Johan A. Martens (2), Jan A. Delcour (1), Chris W. Michiels (1), Guy Derdelinckx (1), Firmin Velghe (4)

(1) Leuven Food Science and Nutrition Research Centre, Leuven, Belgium; (2) Centre for Surface Chemistry and Catalysis, Leuven, Belgium; (3) ProMeta, Interfaculty Centre for Proteomics and Metabolomics, Leuven, Belgium; (4) M4E (Magnets for Emulsions), Heverlee, Belgium

Gushing is the spontaneous wild and uncontrolled overfoaming phenomenon that occurs in beer and other over-carbonated beverages. This problem represents important commercial damages for companies, since this phenomenon can only be disclosed after the opening of bottled or canned beverages. Only a few preventive and no curative techniques are currently available, but some detection methods, such as the patented ELISA test, exist or are in progress. We demonstrated recently that the key factor for gushing is carbon dioxide, mainly the bubble sizes during the carbonation phase. There is a critical diameter for bubbles in a liquid above which they will grow until they explode at the surface and under which they will shrink in size until they have been completely dissolved except in the presence of a contaminant (in the latter case, the bubble becomes unable to solubilize because of the contaminant that adsorbs on the surface of the bubble). In beer, it is well recognized that hydrophobins are responsible for gushing, and they can be defined as the contaminants. Hydrophobins are amphiphilic molecules produced by filamentous fungi and are present when the weather conditions for barley fields are wet. Hydrophobins can adsorb at hydrophilic-hydrophobic interfaces, and it is proposed in our model that they coat the CO<sub>2</sub> nano-bubbles and, thus, prevent their complete dissolution. As a consequence, they are a source of primary gushing when decapping a bottle or opening a can. The objective of this research is to show if a magnetic field can modify the self-assembly of hydrophobins and prevent them from coating CO<sub>2</sub> nano-bubbles. For this, we have used different extracts of gushing and non-gushing malt. The extraction is realized with a 170 mM Tris/HCl buffer (pH 9) containing 1% SDS (sodium dodecyl sulfate). The SDS is then

removed by precipitation with 2M KCl. For experiments at the pilot scale, several liquids are studied: first, water as the simplest model and then beer without gushing. Gushing malt extracts are added to the liquids to simulate the gushing beers, and the buffer is added to the control samples. To study the effect of a magnetic field during the carbonation process, CO<sub>2</sub> is injected by a Venturi in the liquid, coming from a container, and just after the Venturi, the carbonated liquid is submitted to a magnetic field. The objective is to assess the interaction of hydrophobins with CO<sub>2</sub> in water and beer after treatment with a magnetic field and to determine whether the hydrophobins can still stabilize the CO<sub>2</sub> bubbles to provoke gushing in the magnetically treated carbonated liquid. This research is being conducted under the Hydrophobin Leerstoel, which is funded by Duvel-Moortgat N.V., Brasserie d Orval S.A., Bières de Chimay S.A., and Cargill Malt.

*Sylvie Deckers received a M.S. degree in chemical bioengineering from ULg-Gembloux Agro-Bio Tech, with honors, in 2008. Her master's thesis was on the "Possible Influence of Surfactants and Proteins on the Efficiency of Microbiological Surface Sampling." Since February 2009, she is active as a Ph.D. student in the research consortium of KULeuven-LFoRCe and M<sup>2</sup>S. Her topic is on the comprehension of the primary gushing phenomenon and mainly about the interaction between CO<sub>2</sub> nano-bubbles and amphiphilic contaminants such as hydrophobins.*

#### **P-48**

##### **Assessing the impact of extraction condition and grist particle size on the phenolic acids composition and antioxidant activity of malt**

YIN LI (1), Paul Schwarz (1)

(1) North Dakota State University

Malt with phenolic compounds associated with high antioxidant activities is of great interest to the malting and brewing industry. In this study, the combined impact of grain particle size, extract solvent, and grist/extract solvent ratio on antioxidant activities, total phenolic content, and phenolic acid compositions in malt was measured and analyzed. In terms of extract solvent, 80% acetone extract showed significant higher values in DPPH• and ABTS•<sup>+</sup> radical scavenging activities, while 80% methanol extract favored reducing power, superoxide anion radical scavenging activity, and iron chelating activity. Stepwise linear regression showed that extract solvent was the most important factor for DPPH• radical scavenging activity. As for ABTS•<sup>+</sup> radical cation scavenging activity, extract solvent and grist/extract solvent ratio have equivalent contributions in the model. Grind level and grist/extract solvent ratio can explain 82% variation in total phenolic acids. The ratio of grist to extract solvent at 1:20 showed the highest averaged values for antioxidant activities, total phenolic content, and individual phenolic acid compositions. Of four extract solvents, the highest averaged ferulic acid content was found in water extract, indicating the major phenolic acid in malt water extract was ferulic acid. With the exception of chlorogenic acid and *p*-coumaric acid, all the other phenolic acids showed the highest values in fine grind.

*Yin Li is a research assistant professor in the Department of Plant Sciences at North Dakota State University. He received his Ph.D. degree in the area of malting and brewing in 2006 and started his post-doctoral research work with Professor Paul Schwarz. He has published more than 30 papers in international peer-review journals. Recently, he is interested in lipoxygenase, antioxidant activity of barley and malt, arabinoxylans, extract, and organic acids.*

#### **P-49**

##### **Bioethanol from brewer's spent grains: Novel pre-treatment and hydrolysis approaches**

JASON BENNETT (1), Graeme M. Walker (1), David Bremner (1)  
(1) University of Abertay, Dundee, U.K.

Given the current debate raging within both political and scientific circles on what measures to adopt as effective combatants to global climate

change, it is clear that one major component within our arsenal is a sustainable and carbon-neutral alternative to currently utilized liquid transportation fuels. Bioethanol (fuel alcohol produced via fermentation) has become a major contributor in this regard. Optimized production processes together with sustainable feedstocks are major foci of current bioethanol research. Our particular focus is on bioethanol derived from waste materials that exhibit limited potential for use in other applications. Conversion of lignocellulosic biomass represents one of the most attractive, but challenging, opportunities for meeting the demand for sustainable bioethanol production systems. Abundant sources of lignocellulose come in many forms, including waste streams from the brewing/distilling, forestry, and paper industries. We have investigated novel pre-treatment and enzyme technologies to extract fermentable carbohydrates from brewer's spent grains (SG). Such material represents a rich source of fermentable carbohydrate, and we have shown that these can be readily extracted from SG using physicochemical techniques such as thermal/acidic pre-treatments, followed by further enzymatic hydrolysis (White et al, FEMS Yeast, 2008). As an alternative to energy-intensive methods, we are evaluating the applicability of sonochemical technology in SG pre-treatments. For example, we have applied ultrasound to SG with a variety of experimental ultrasonic parameters, including output frequencies of 20 kHz to 1.1 MHz and output powers of 50–150 W. Ultrasonic reaction temperatures and types of reaction vessel have also been optimized. As well as evaluating ultrasound as a pre-treatment method for brewer's SG, we are also investigating the hydrolytic efficiency of ultrasound combined with oxidative chemical reactions such as ozonolysis. The main aim of this project is to develop novel low-energy alternatives to current pre-treatment strategies, and we have compared energy "costs" associated with ultrasonic irradiation of SG with those of more traditional pre-treatment methods. We are also evaluating a new generation of hydrolytic cellulases and xylanases in terms of fermentable sugar release from SG pre-treated with acid. This presentation will discuss these findings, as well as examine future challenges in converting brewer's SG to biofuel.

*Jason Bennett received a B.S. degree in biotechnology from the University of Abertay Dundee, Scotland, in 2008. He commenced his Ph.D. studies at the same institution in 2009, with a research interest focusing on novel lignocellulosic pretreatment mechanisms for fuel ethanol production. He is a student member of both the ASBC and IBD.*

**P-50**  
**Characterization of dry Nottingham ale yeast under different fermentation conditions**

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Dry beer yeast is gaining more and more acceptance in craft brewing, as well as in industrial brewing, because of its high quality and consistency. Despite the increased interest, there is very little information available on the behavior of dry beer yeast strains under different fermentation conditions. In this presentation Nottingham ale yeast was used in three different beer wort compositions at three different temperatures and with different pitching rates. Each fermentation was monitored for a variety of characteristics, including sugar utilization, pH, and cells in suspension, as well as the production of flavor compounds, higher alcohols, and esters. The results highlight the versatility of this particular strain, which can be used for a wide variety of beers and allows brewers to express their creativity.

*Tobias Fischborn was appointed research scientist for Lallemand Inc. in 1998. He is now responsible for brewing research and development at Lallemand and is also responsible for quality control and quality assurance of all brewing yeasts. He graduated from the Technical University Munich/Weihenstephan in 1993, where he obtained an engineering degree in brewing and beverage technology. He continued*

*studying for a Ph.D. degree in brewing, which he finished in 1997. Prior to his studies at Weihenstephan, he worked as a brewer at Brewery Ph. & C. Andres in Kirm, Germany.*

**P-51**  
**Determination of bisphenol-A (BPA) in polycarbonate plastic bottles by SIDA**

LEIF A. GARBE (1)  
(1) VLB Berlin, Berlin, Germany

Bisphenol A (BPA) is one of the monomers found in polycarbonate plastic (PC) bottles. PC is used for production of CDs, DVDs, Blu-ray discs, drinking bottles, plastic wrapping, and packaging in general. BPA is controversial due to its health effects. It is a "xenoestrogen" that inhibits sexual differentiation and brain development of birds and mice and is suspected to have adverse health effects and to be mutagenic. Therefore, BPA is an "endocrine disruptor." A tetra-deuterated isotope standard of BPA was chemically synthesized and used in stable isotope dilution analysis (SIDA) of BPA in PC drinking bottles. The bottles were filled with water, the standard was added, and the water was extracted with organic solvent. Evaporation of solvent and GC-MS analysis in SIM mode enabled a very sensitive and reliable BPA analysis. The synthesis of tetra-deuterated BPA and stable isotope dilution analysis (SIDA) of BPA were successful. The SIDA of BPA in PC showed low but significant amounts of BPA. We found concentrations of 0.4 µg/kg BPA in drinking bottles designed for infants. The age and freshness of the drinking bottles did not significantly influence the amount of BPA in the product (water). Nowadays, PC bottles are vanishing from the food market. However, they are still sometimes used. This study shall show the power, sensitivity, and reliability of SIDA for BPA as well as other plastic monomers.

*Leif-Alexander Garbe is a professor and the head of the TU Berlin Chair for Bio-analysis/Molecular Analysis and, since 2002, also the head of the VLB Research Institute for Spezialanalytik. Leif graduated from TU Berlin (TUB), Germany, with a diploma in chemistry in 1996. He then worked at the Research and Teaching Institute for Brewing in Berlin (VLB). From 1997 to 2002 he worked on his Ph.D. thesis and received his degree in April 2002 from the Institute of Biotechnology, TUB. His work included supervision of students in biotechnology and brewing. In 2002 he established a new research group at TUB that focuses on trace analysis, biotransformations, and biocatalysis. In July 2008 he became the head of the TUB Department for Bio-analysis/Molecular Analysis.*

**P-52**  
**Determination of volatile monophenols in beer using solid-phase microextraction combined with GC-MS**

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(1) Centre for Malting and Brewing Science, Leuven, Belgium

Monophenols are a diverse group of volatiles with one common feature: the presence of an aryl ring with a hydroxyl group attached to it. In beer and wine, the most studied monophenols are 4-vinylguaiacol, 4-ethylguaiacol, 4-ethylphenol, and 4-vinylphenol. They are known to be essential for the overall flavor perception in many beers and wines, and when present in high concentrations, they can cause off-flavors. In addition to these compounds there are several other flavor-active monophenols to contemplate. Vanillin is described as one of the key aroma compounds of wines, whiskies, malts, fruit juices, and dairy products. Thymol and eugenol are important constituents of plant essential oils. Salicylaldehyde is a characteristic aroma component of buckwheat, and guaiacol, acetovanillone, methyl vanillate, syringaldehyde, and acetosyringone all contribute to wine flavor. All of these monophenols originate from plant material, and possible precursors are reported to be lignin, hydroxycinnamic acids, or glycosides. Since malt and hops are the main raw materials of beer, and thermal and fermentation processes during beer production allow monophenols

to be released from their precursors, beer most likely contains these monophenols. Therefore, it is interesting to determine the monophenol content of beer in order to determine their influence on beer flavor. However, detecting monophenols in beer is difficult because of their low concentrations, their low volatility, and the high reactivity attributed to the polar hydroxyl group. The abundance of higher alcohols and esters in the beer matrix renders this task even more difficult, and a targeted approach is needed. For this reason, an adequate method was developed combining in situ acetylation of the monophenols, followed by headspace solid-phase microextraction (SPME) and gas chromatography-mass spectrometry (GC-MS). In the first step, the acetylation reaction was optimized, varying the amount of acetylation agent (acetic anhydride) and  $\text{KHCO}_3$ , as well as the pH, of the medium. Then, a series of parameters relevant for headspace SPME, such as salt addition, fiber composition, extraction temperature, and extraction time, were optimized. Afterward, calibration and validation of the method was performed. The repeatability was good (<10%) for 7 of the 14 monophenols studied and acceptable (<15%) for the other 7 phenols. The limits of detection were well below the sensory thresholds of the individual compounds. Finally, the method was applied successfully for the determination of monophenols in 16 commercial beer samples, including various Belgian beer styles.

*Femke Sterckx was born in 1984 in Herentals, Belgium. In 2007, she graduated with an M.S. degree in applied biological sciences and engineering from K.U. Leuven, Belgium. She carried out her master's thesis on the influence of Saccharomyces cerevisiae and Brettanomyces custersii on glycosidically bound flavor compounds in hops and sour cherries at the Centre for Malting and Brewing Science at K.U. Leuven. After graduation she started a Ph.D. program at the Centre for Malting and Brewing Science. Her work is focused on the identification of flavor-active monophenols in beer and their influence on beer flavor. For this research, IWT Vlaanderen grants her financial support.*

#### **P-53**

##### **Development of an auto-cuvette machine to measure semiautomatically premature yeast flocculation activity in malt**

SETSUZO TADA (1), Nariaki Wasano (1), Asumi Nakahoshi (2), Naomu Nishiwaki (1)

(1) Kirin Brewery Company, Ltd., Yokohama-shi, Japan; (2) Kirin Brewery Company, Ltd., Asakura-shi, Japan

Premature yeast flocculation (PYF) is the phenomenon whereby yeast flocculates prior to the depletion of nutrients in the wort. Kirin has already developed a rapid, sensitive, and reproducible method—the cuvette method—to measure the premature yeast flocculation activity level in malt without the need for mashing or fermentation. While the cuvette method is a powerful tool for research use, it is difficult to measure multiple samples simultaneously, so its practical use is limited, mainly because the cuvette method is conducted manually and the sedimentation of yeast cells is measured by optical absorbance. In order to solve these problems, we developed a semi-automatic auto-cuvette machine in which agitation of the reaction mixture is conducted automatically, and the sedimentation of yeast cells is monitored by digital video camera and the rate of sedimentation is calculated by imaging analysis.

*SetSUzo Tada received his M.S. degree in fermentation technology from Osaka University and began employment with Kirin Brewery, Co. Ltd. in 1981. He studied yeast genetics at Massachusetts Institute of Technology as a visiting scientist (1984–1986) and fungal genetics at the National Research Institute of Brewing (1987–1990). He received his Ph.D. degree from Tokyo University in agricultural chemistry in 1991. At Kirin Brewery, Tada served as project leader for the genetic modification of brewer's yeast (1990–1997) and manager of the Quality Assurance Department at the Kirin Sendai plant (1997–2000). Since 2000, he has been engaged at the Quality Assurance Center for Alcoholic Beverages.*

#### **P-54**

##### **Differential RNA expression of Bmy1 during late seed development in wild and cultivated barley and the association with $\beta$ -amylase activity**

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(1) USDA-ARS Cereal Crops Research Unit, Madison, WI; (2) USDA-ARS Vegetable Crops Research Unit, Madison, WI; (3) University of Wisconsin, Madison, WI

Four genotypes carrying different  $\beta$ -amylase 1 (Bmy1) intron III alleles (Bmy1.a, Bmy1.b, Bmy1.c, Bmy1.d) were analyzed for differences in Bmy1 DNA sequence, Bmy1 RNA expression,  $\beta$ -amylase activity and protein, and total protein content during late seed development. Wild barleys Ashqelon (Bmy1.c) and PI 296897 (Bmy1.d) had 2.5- to 3-fold higher Bmy1 RNA expression than cvs. Legacy (Bmy1.a) and Harrington (Bmy1.b). Large insertion/deletions in the Bmy1 promoter and third intron do not appear to affect the amount of Bmy1 mRNA. Approximately 503 bp upstream of the Bmy1 gene are highly conserved among cultivated and wild barleys and may contain transcription-factor binding sites necessary for Bmy1 expression. Bmy1 expression patterns and putative transcription-factor binding sites in the promoter indicate Bmy1 may be under the control of seed storage protein transcription factors.  $\beta$ -Amylase activities per milligram of protein were not significantly different at maturity between all genotypes, whereas Ashqelon and PI 296897 had significantly higher activities per gram fresh weight than Legacy and Harrington, because wild barleys had more total protein. These data indicate wild barley genotypes produce more total proteins during seed development, causing an increase in Bmy1 expression and Bmy1 protein, thus higher  $\beta$ -amylase activity per milligram fresh weight.

*Cynthia Henson earned a Ph.D. degree at the University of Wisconsin-Madison and in 1986 became a research plant physiologist with the USDA-ARS-Cereal Crops Research Unit and an agronomy professor with the University of Wisconsin. She currently serves as research leader of the Cereal Crops Research Unit, Madison, WI.*

#### **P-55**

##### **Effect of lauter turbidity on the brewing process and beer quality**

TAKUYA HATANAKA (1), Thomas Becker (2), Martin Krottenthaler (2)

(1) Asahi Breweries, Ltd., Tokyo, Japan; (2) Lehrstuhl für Brau- und Getränke-technologie, Freising, Germany

To investigate the effect of lauter turbidity on beer quality, pilot-plant (60-L scale) trials were carried out varying grist modification levels (well and poorly modified malts) and lauter turbidities (low and high turbidities). Whether grist modification level was well modified or not, turbid lauter wort caused an increase in extract recovery, caused a loss of bitter substances during wort boiling, and accelerated primary fermentation. But, there was not great difference in beer quality. Next, we researched the relationship between lauter turbidity and its influence on the brewing process. The results showed that loss of bitter substances increased with an increase in lauter turbidity (however, the degree of the loss was not high) and that extract recovery also increased with an increase in lauter turbidity (however, there was a limit to increases in extract recovery). On the other hand, fermentation was accelerated when lauter turbidity exceeded a certain level. From these results, it was concluded that lauter turbidity should be controlled within the moderate range if extract recovery is not sufficient or fermentation problems occur in a brewery.

*Takuya Hatanaka received an M.S. degree in engineering from the University of Osaka in 1998 and began working for Asahi Breweries, Ltd. Since 2008, he has been studying brewing science at Munich Technical University (Weihenstephan) as a guest student.*

**P-56****EPR-detected free-radical formation following photoactivation of a commercial hop oil product**

DAVE BARR (1)

(1) Bruker BioSpin Corp., Billerica, MA

Hops used in the brewing process contain many phenolic- and hydroquinone-type components such as alpha-acids (which include humulones, cohumulones, and adhumulones). Hops also contain beta-acids, iso-alpha-acids, and essential oils such as humulene. Some forms of these compounds are photo-reactive. For example, exposure of beer to light often leads to the formation of a specific radical that combines with sulfur compounds (e.g., cysteine) to form a mercaptan compound that is involved in the “skunking” of beer. The product studied here was described as an aqueous alkaline solution of potassium salts from tetrahydroiso-alpha-acids (9%). It is used as a stabilizer for beer and to provide bitterness and foaming properties in cases where a normal dry-hopping process is not used. Upon exposing the hop oil sample to a concentrated and focused light source, the EPR spectra from light-induced free radicals were recorded in the presence and absence of the spin-trap reagent DMPO (5,5-dimethyl-1-pyrroline-*N*-oxide). Upon opening the shutter to the lamp accessory, a direct EPR spectrum for a free radical was recorded from the hop oil sample. When the shutter was closed the signal disappeared. The free radical formed revealed an EPR spectrum that had at least seven lines and may be that of a semiquinone anion radical. Addition of the spin-trap reagent DMPO to the illuminated hop oil sample resulted in the trapping of three major radicals. One of the radicals (indicated by hyperfine coupling constant analysis) was consistent with DMPO-trapped superoxide anion radical. Absolute confirmation of the superoxide anion radical was provided by the addition of SOD (superoxide dismutase), which completely abolished the spectrum assigned to the DMPO superoxide radical adduct. Phosphorescence (green glow) was also observed when the sample was exposed to the light source. In addition the sample seemed to undergo some sort of polymerization, as aggregates formed in the area of the sample cell where the light was directed. The possible mechanisms for light-induced free-radical production and phosphorescence are discussed.

*Dave Barr is a senior applications scientist working in the EPR division of Bruker BioSpin Corporation. Among the many EPR applications, free-radical oxidation in food and beverage products has been a major focus for Dave for the past 12 years. He was recently chair of the ASBC Technical Subcommittee that approved the method for analysis titled “Measurement of Oxidative Resistance in Beer by Electron Paramagnetic Resonance.”*

**P-57****Ethanol tolerance of lactic acid bacteria**

BARRY ZIOLA (1), Vanessa P. Pittet (1)

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Microbial tolerance of ethanol is relevant in various industrial settings, one of which is the brewing industry. This is because despite high ethanol levels being reached during fermentation, bacterial contamination can still be an issue. Most bacteria can only survive in low levels of ethanol (approx. 5%). However, it has been found that lactic acid bacteria have the ability to survive and grow in high ethanol concentrations. To further understand microbial contamination in a brewery setting, where the most common problematic isolates are lactic acid bacteria (e.g., lactobacilli and pediococci), the ethanol tolerance of beer-spoiling and non-spoiling lactic acid bacteria was analyzed. Two levels of tolerance were investigated—one being the minimum inhibitory concentration (MIC), which was defined as the level of ethanol in which isolates could no longer grow. The second tolerance level was the minimum bactericidal concentration (MBC), which was defined as the level of ethanol in which organisms could not survive after a week

of exposure. Interestingly, no difference in ethanol MIC or MBC was found between beer-spoiling and non-spoiling bacteria. Furthermore, some bacteria had a high ethanol MIC and MBC (i.e., as high as 15 and 25%, respectively). This information has implications for breweries; specifically, lactic acid bacteria that have a high ethanol MIC and MBC can be carried through successive fermentations, as the organisms are simply static while ethanol levels are high but can continue growing once the ethanol levels drop back down to lower levels as the contaminated yeast is pitched into the next fermenter. Now that a better understanding of levels of ethanol tolerance in various lactic acid bacteria is available, a genomic-based approach will be used to determine the mechanisms used by these organisms to survive and grow in the presence of these high levels of ethanol.

*Barry Ziola received a B.S. (honors) degree in botany from McGill University, Montreal, Canada, in 1970. After completing a Ph.D. degree in biochemistry at the University of Alberta, Edmonton, Canada, in 1975, he undertook a three-year post-doctoral stint at the University of Turku, Turku, Finland. Barry has been at the University of Saskatchewan, Saskatoon, Canada, since 1978, and was promoted to professor in 1986. His interest and continuing research in brewing spoilage bacteria dates to the mid-1980s.*

**P-58****Experiences with a special circulation system in a rectangular nonpressurized fermenter**

URS WELLHOENER (1)

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With every yeast pitch, it is essential to provide homogenized, even distribution of yeast in the wort. This promotes a quick and smooth launch of the fermentation. When the main fermentation is over with the depletion of the wort extract, the yeast begins to settle. Settlement is not desired until the diacetyl reduction is complete. Large fermenters (CCVs and rectangular fermenters) require homogenized suspension until the end of diacetyl reduction. To promote suspension through fermentation in large vessels, a recirculation system from Alfa Laval has been tested at our brewery. This system uses the existing CIP piping of a fermenter. From the outlet, a pipe is connected upstream to the circulation pump, from which the batch is pumped via the sprayballs back into the fermenter. The existing sprayballs were replaced by a special sprayball from Alfa Laval, which provides homogeneity and reduces the mechanical impact to yeast. While this system was implemented successfully in CCVs in Denmark, there have been no tests in rectangular non-pressurized fermenters. Trials will be performed in two identical, rectangular 1,800-bbl fermenters (circulation vs. non-circulation), using the same wort composition and the same pitch yeast conditions. Wort analysis includes OG, color, BU, Ca, Zn, and free amino nitrogen (FAN). The pitching conditions are verified via pitching rate, viability, and solids. The fermenters will be compared via typical fermentation parameters (extract reduction, alcohol formation, pH, cell counts, diacetyl). Additionally the fermentation by-products (acetaldehyde, higher alcohols, esters) will be included in this evaluation. A sensory evaluation of the final beer will complete these checks. The first trial uses a standard circulation regime recommended by Alfa Laval. Based on these results the following trials will be further optimized regarding typical fermentation parameters and circulation speed. If the trials are successful, it is expected, that the fermentation time can be reduced, resulting in same or improved beer flavor at the end of fermentation using recirculation. Such improvement will allow for increased beer production without the addition of more fermentation vessels.

*Urs Wellhoener is the corporate manager for yeast and fermentation for the Boston Beer Company and joined the company in 2007. His focuses are yeast management and microbiology. He is a technical graduate as brewer and maltster (1991–1993) and received a Dipl.-Eng. degree*

from the Faculty of Brewing and Food Technology of the Technische Universität München-Weihenstephan (TUM) in 1999. After graduation in 1999 he was a project manager on a yeast project at Veltins Brewery, Meschede-Grevenstein (1999–2000). Between 2000 and 2007 Wellhoener was a scientific assistant and doctoral student at the Chair of Brewing Technology II at the Weihenstephan Center of Food and Life Sciences, TUM. Wellhoener received his Ph.D. degree for his studies on yeast physiology during fermentation and propagation. During this time he also worked for Muellerbraeu, Pfaffenhofen/GER, as QC manager.

#### P-59

##### Factors influencing free-radical development in malt, as measured by EPR

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(1) Prairie Malt/Cargill Malt, Biggar, SK Canada; (2) Cargill Malt, Minneapolis, MN

Studies were undertaken to identify factors that correlate with the EPR signal in boiled Congress wort. Variety, malt quality parameters (including LOX), geographic origin, and processing conditions were evaluated as possible sources of elevated free radicals. Within varieties, analytical factors that explain most of the variation in EPR signal in pale malts and the variation in highly colored malts were examined and identified through both micromalted and production samples. Additional processing factors such as high temperatures, mineral content of steep water, and anaerobic conditions were investigated as possible events that resulted in elevated EPR signals. Limited pilot brewing trials suggested that the magnitude of EPR signals in wort were proportional to the EPR signals seen in beer.

*Marcia Browsers is currently an inventory and barley quality manager for Prairie Malt Limited. She moved to Cargill's Prairie Malt Limited in Biggar, SK, Canada, eight years ago as quality assurance manager, before continuing on as senior analytical chemist on the Cargill Malt Technical Team and then moving over to the Operations Department three years ago. Marcia holds a B.S. degree from Michigan State University in Horticulture, M.S. degree from the University of California, Davis, in vegetable crops (genetics), and Ph.D. degree in agronomy (plant breeding and genetics) from Colorado State University. Prior to Cargill, she was a research scientist and manager for ConAgra Malt Americas at the Vancouver, WA, and Calgary, AB, Canada, plants. Her first job in malting was in the Malting R&D Department of Coors Brewing Company screening new barley varieties.*

#### P-60

##### High gelatinization temperatures of barley starch—Effects on malt and beer quality

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(1) Technische Universität München, Freising, Germany

The main task of the mashing process is to convert water-insoluble starch into water-soluble, fermentable extract. This degradation process has an optimum that depends first on the activity of amylases and second on the degree of starch gelatinization. Gelatinization means a destructuring of starch molecules and a significant swelling of starch granules. After gelatinization amylases are able to attack the starch granule from the inside. Commonly an infusion mash with mash-in temperature at 62°C (144°F) is used. In years with normal gelatinization temperatures (gelatinization behavior is year dependent) of barley malt (60–62°C) starch granules swell considerably at 62°C and could be attacked by amylases. In years with increased gelatinization temperatures starch does not swell at a mashing temperature of 62°C. This means that enzymes cannot enter and act inside the starch granules, but they are incrementally inactivated at a rest at this temperature. Especially beta-amylase, which is essential for the degradation of starch to maltose, is increasingly inactivated at temperatures above 62°C. If starch is not gelatinized, beta-amylase cannot work optimally during the saccharification rest,

since there is not enough substrate available. The resulting lack of fermentable carbohydrates is expressed in a low final fermentation. At worst, insufficient amylolysis causes turbidity by alpha-glucans and lower extract yields. Response surface methodology was used to investigate the influence of the three malting parameters, vegetation time, degree of steeping, and temperature, on the gelatinization temperature of barley malt. Each predictor variable was tested at three levels. Vegetation times were 5, 6, and 7 days, degrees of steeping were 42, 45, and 48%, and vegetation temperatures were 12, 15, and 18°C. To show the impact of kilning on gelatinization properties of barley malt, different temperatures in the range of 40 to 95°C were used. To detect factors that affect gelatinization of starch during mashing and how that gelatinization process influences the beer quality, the following variables were tested: fineness of malt grist, variation of mashing pH, brewing water composition, and variation of mashing regimes, taking the gelatinization temperature into particular consideration. The analyses used were based on methods outlined by MEBAK or EBC. The intent of this paper is to show the factors that influence the gelatinization process. For brewers and maltsters it is necessary to know the negative effects of increased gelatinization temperatures and how to react to avoid these effects by adapting brewing and malting technology. This work was completed at the Lehrstuhl für Brau- und Getränketechnologie, Technische Universität München-Weihenstephan.

*Florian Schuell received a Dipl.-Ing. degree in brewing and beverage technology from Technische Universität München in Weihenstephan, Germany, in 2006. He began employment with Lehrstuhl für Brau- und Getränketechnologie in 2006 as a Ph.D. student. Since 2008 he has been the deputy lab supervisor of the malt laboratory of this chair.*

#### P-61

##### Influence of the malting process on the content of bioactive compounds in malt

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Nowadays our nutrition is mainly based on only three cereals—wheat, rice and corn, which contribute over 75% of the world's starch production. At the same time diseases caused by wrong or unbalanced diets are becoming a severe problem in Western countries. In this regard the enrichment of bioactive compounds in the malting process with the objective of providing their beneficial health effects to the consumer is a very important field. The malting process is influenced by the quality of the raw material and several process parameters (e.g., moisture, temperature, and time), and the enrichment of bioactive compounds depends on the same variables. A statistical design of experiments (DOE) approach (response surface methodology) was used to evaluate the interactions of the influencing factors and to optimize the enrichment process. One group of the functional components we investigated was the arabinoxylans. In order to investigate this group of substances it was necessary to establish a method for the determination of water-soluble and -insoluble arabinoxylans. The method we used consists of acidic hydrolysis of the arabinoxylans, followed by HPAEC/PAD detection. Using this method we were able to enrich water-soluble arabinoxylans in wheat malt. Another interesting group of components are the flavanols. We established a method for the characterization of flavonols like rutin, vitexin, and quercetin by HPLC separation. This method has helped us to determine optimum malting parameters for the enrichment of these functional components in buckwheat malt. Additionally, changes in the vitamin B<sub>1</sub>, B<sub>2</sub>, and B<sub>12</sub> content of cereals were monitored during the whole germination and malting process of different cereals, as well as the changes in water-soluble arabinoxylan and fructan.

*Moritz Krahl was born in 1980 in Schwetzingen, Germany. In 2000 he started his education in brewing and beverage science at the Technische Universität München in Germany. In 2004 he graduated with a B.S. degree and in 2005 with a Dipl. Ing. degree. Since 2005 he has been*

working as a Ph.D. student at the Institute for Brewing and Beverage Technology in Weihenstephan.

## P-62

### **Innovative yeast extract as nutrient and natural antioxidant during propagation**

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In the beer industry, it is a common practice to crop and pitch yeast from one cylindroconical tank to another, several times depending on economics. However, this practice applies stresses on the organisms and affects quite irreversibly their physiology, morphology, and gene expression, playing an important role in the behavior and performance of fermentation. Therefore, the difficulties in successive fermentations are mainly due to a progressive decrease in the viability and vitality, yeast flocculation, membrane permeability, and fermentation rate, affecting especially sugar and nitrogen uptake, alcohol production, and consequently the taste of the beer. In order to guarantee a better reproducibility of the end-product, an innovative regulator and activator, GSH1, has been elaborated. Combining nutritive properties to an elevated glutathione concentration of a selected yeast extract (from *Saccharomyces cerevisiae* PB GS 12 and PB GS 14), GSH1 permits the maintenance of yeast characteristics through several repitchings. Analyses have been carried out both on cold wort and during fermentations, allowing us to follow the physiological evolution of the yeast and its ambient for each successive generation: sugar spectrum; amino acids content (in particular glycine, phenylalanine, tyrosine, tryptophan, and alanine); vitamin content; biotin and thiamine for their catalytic role; and inositol, which is involved in membrane synthesis. Mineral content also was analyzed (trace elements Mg, Ca, Zn, Mn, K, Cu, and Fe as cofactors for numerous catalytic activities and reactions; sterol and fatty acids content such as ergosterol, which exerts a significant role in the stability of the plasma membrane and trehalose reserve against yeast stress). We also studied the release of mannoproteins during post-fermentation and beer rest, which may act negatively on turbidity and organoleptic aspects of the final beer. The readjustment and optimization of the original medium was orchestrated every two generations, bringing consequent improvement, as shown in the comparative analysis. GSH1 also was evaluated in top fermentation compared with traditional activators mainly composed with vitamins (long-chain polysaccharidic celluloses stabilizing diammonium phosphate, ammonium sulfate, and thiamine hydrochloride). A production scheme illustrates the major steps in selecting the yeast extract GSH1 and its glutathione fraction.

*Caroline Scholtes graduated from the Catholic University of Louvain-La-Neuve (Belgium), section brewing science. Caroline has been working for the past year as a consulting engineer for AEB Group. The international company specializes in commodities for the brewing industry, as well as the beverage industry, in the fields of stabilization, clarification, fermentation, and filtration and has developed well this branch of research and development in biotechnology, especially in fermentation process and yeast metabolism. Caroline is now in charge of western Europe for the Brewing Department and takes part in different fields of research and development for the company.*

## P-63

### **Iron-chelating properties and hydroxyl-scavenging activities of hop acids**

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Hops (*Humulus lupulus*) provide beer with bitterness, aroma, foam

stability, antimicrobial power, and flavor stability. Its antioxidant properties are a topic of considerable debate. The interaction of transition metals, such as iron and copper, with oxygen and oxygen-derived species can lead to highly reactive hydroxyl radicals that damage beer. The purpose of this work was to investigate the behavior of hop acids in Fenton-based oxidation systems and to examine whether their antioxidant action derives from their ability to chelate iron ions or to scavenge hydroxyl radicals. The antioxidative reaction mechanism of hop acids was investigated using ferrozine- and deoxyribose-based assays to examine their chelating power and ability to scavenge hydroxyl radicals, respectively. Purified  $\alpha$ -acids and iso- $\alpha$ -acids and  $\beta$ -acids were obtained from a commercial source. These hop materials were examined individually in an acetate buffer at wort and beer pH. The principle of the ferrozine assay relies on the formation of a stable complex between ferrous iron and ferrozine. Therefore, the iron-chelating power of the hop acids can be measured by assessing their ability to compete with ferrozine for ferrous iron, avoiding the formation of this colored complex with absorbance peaking at 562 nm. The formation of  $\bullet$ OH radicals from Fenton reagents was quantified using 2-deoxyribose oxidative degradation. The principle of the assay is the quantification of the 2-deoxyribose oxidative degradation product, malonaldehyde, by its condensation with thiobarbituric acid and measurement of the reaction product absorbance at 532 nm. Typical reactions were determined by measuring the formation of  $\bullet$ OH radicals from the Fe(II)-hop acid complexes when incubated with  $H_2O_2$ . The scavenging activities of the hop acids were examined by producing  $\bullet$ OH radicals from Fe(III)-EDTA, ascorbate, and  $O_2$  followed by measurement of the concentration-dependent decline of 2-deoxyribose oxidative degradation when hop acids were added to the system. Hop  $\alpha$ -acids and iso- $\alpha$ -acids showed both strong chelating power and hydroxyl radical scavenging properties. However,  $\alpha$ -acids were shown to be the stronger chelators. Because of their low solubility in aqueous solutions, a true comparison of the action of the  $\beta$ -acids relative to the  $\alpha$ -acids and iso- $\alpha$ -acids could not be made. Even though the  $\alpha$ -acids and iso- $\alpha$ -acids did chelate iron, these iron chelates did not inhibit  $\bullet$ OH radical formation. In previous work in our lab and elsewhere, hop acids have been shown to confer significant antioxidant properties in beer. It is hypothesized that the antioxidant characteristic can be traced back to the  $\bullet$ OH quenching properties of the hop acids.

*After qualifying his final secondary-school examinations (2000), Philip Wietstock started his biotechnology studies as a graduate engineer at the Technische Universität Berlin. Before starting to work as a student research assistant, with his main focus on ESR spectroscopy at the VLB/ Technische Universität Berlin (2007-2009), he performed a student research project in the field of yeast propagation at the Karlsbergbrauerei GmbH (2006). Philip successfully graduated as a diploma engineer in biotechnology, with specialization in brewing science, in November 2009 and recently started a Ph.D. program in the field of antioxidative properties of hops. Currently he is performing an exchange year at the Department of Food Science and Technology, Oregon State University, Corvallis.*

## P-64

### **Levels of $\beta$ -glucan and pentosan and their degradation products in beer**

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While  $\beta$ -glucans and pentosans are of concern to brewers for their adverse impact on liquid-solid separation processes and beer stability, there is nonetheless interest in them for their potential classification as soluble fiber. Furthermore, their degradation products, if they survive into the final beer, may be putative prebiotics. We have analyzed a variety of

beers, using enzymic, HPLC, and TLC methods, for their carbohydrate contents and distributions, and the data will be reported.

*Makoto Kanauchi graduated from the Tokyo University of Agriculture in 1996. He received a Ph.D. degree from the Tokyo University of Agriculture in 1999. After post-doctoral work at the University of California, Davis, he was employed at the Institute of Food Science in Fuji Oil Corporation in Moriya, Ibaraki, Japan. Since 2005, he has been with the Department of Food Management, Miyagi University.*

#### **P-65**

##### **Pro- and antioxidative effects of the Maillard reaction products in malt on oxidative beer stability**

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(1) Technische Universität Berlin, Germany

The effect of special malts on oxidative beer stability has been reported by different authors, although the effects are controversial. The changes in color of finished beer generally originate from the Maillard reaction products found in malts, which are generated at high temperatures during kilning and/or roasting. The role of these components on the flavor stability of beer has been a matter of discussion during the recent past. In a former publication, it was shown that the concentration of stable organic radicals in malt is directly related to the content of Maillard reaction products and the color of the malt, resulting in a lower antioxidative potential of the wort and final beer. This negative effect of specialty malts is based on the more rapid consumption of SO<sub>2</sub> during the brewing process and in the finished beer. Following this investigation, beers were produced in small-scale trials (1 L) consisting of 10% specialty malt and 90% pilsner malt. The endogenous anti-oxidative potential (EAP) and radical generation of wort and beer was measured by EPR spectroscopy (EAP determination, T-400 value). In direct comparison with the EPR methods, traditional methods were used according to MEBAK and Chapon to determine the reduction power. The method of Chapon provides information about the content of reductive substances that are able to reduce Fe<sup>+3</sup>, which is one of the Fenton reaction products. The reducing power method of MEBAK indicates the quantity of fast-reacting reducing substances. The results show a direct correlation between the content of Maillard reaction products in malt (color) and lower oxidative stability (EAP) and higher radical generation in the wort and final beer. In contrast, the same specialty malts lead to a higher reducing power when measured by the Chapon or MEBAK methods. These contradictory results seem to be one reason why the influence of specialty malts on beer stability has been a matter of discussion in the literature. The explanation of this indirect correlation arises from the strong reduction properties of specific Maillard reaction products that can rapidly reduce oxidized metal ions like Fe<sup>+3</sup> to Fe<sup>+2</sup> and consequently intensify the Fenton and Haber-Weiss reaction system. Based on the acceleration of the Fenton and Haber-Weiss reaction system a stronger radical generation of very reactive radicals (e.g., OH•) can be observed in the wort and beer matrix. This leads to faster consumption of specific antioxidative substances such as SO<sub>2</sub>. The results present the pro- and antioxidative effects of the Maillard reaction products in beer caused by the addition of specialty malts.

*After qualifying as a certified technician in preservation engineering (1991–1993), Thomas Kunz completed his basic studies in chemistry at Isny Fachhochschule (1994–1995) and his basic studies in food chemistry at Wuppertal University (1995–1998) before starting to study food technology at Trier Fachhochschule (1998–2002). After graduating, he worked as a chartered engineer (Dipl. Ing. degree) in the field of ESR spectroscopy at the Institute of Biophysics at Saarland University (2002–2004). Since January 2005, he has been employed as a Ph.D. student at the Research Institute of Brewing Sciences at the Technical University Berlin. His main research focus lies in analyzing radical reaction mechanisms in beer and other beverages using ESR spectroscopy.*

#### **P-66**

##### **Reduction of hazardous solvent usage in the hops laboratory**

JOYCE E. CARR (1), Timothy J. Kostelecky (1)  
(1) John I. Haas, Inc., Yakima, WA

The brewing and hop industries can help with environmental concerns by making an effort to reduce organic solvent consumption and hazardous waste disposal resulting from hop analysis, while maintaining analytical accuracy and precision. In this study we have made strides in achieving this with two steps: 1) reducing the hop sample size in half, with a concomitant reduction in extraction solvent; and 2) using an automated diluter to prepare all samples, which allows the final sample volume to be reduced from 50 to 10 mL. The overall solvent consumption per hop sample utilizing the modified methodology is 60 mL, which is a savings of 90 mL per sample over the 150 mL used in conventional ASBC methods. In using the modified methodology, we estimate that our solvent purchases and hazardous waste disposal costs can be reduced by about 60%. Initial data from spectrophotometric and high-performance liquid chromatography (HPLC) analyses show a statistically acceptable correlation of the sample means between the modified and conventional ASBC methods in analyzing hop cones, hop pellets, and pure hop resin extract. Further studies are needed to ensure the robustness of the new methodology in terms of the precision and reproducibility needed to be considered as an acceptable industry standard method.

*Joyce Carr received a B.A. degree in chemistry from California State University, Sacramento, in 1986. She has more than 12 years of experience in the hop industry and began employment with John I. Haas, Inc. in 2003 as a chemist in their Advanced Hop Products Division. Since November 2005 she has been the quality assurance manager for Haas. Joyce has served on the ASBC Board of Directors as secretary and is currently the editor of the ASBC Buzz.*

#### **P-67**

##### **Simplified mashing methods for initial prediction of malting quality**

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Methods to assess the potential malting quality of barley lines commonly use the Congress mashing (CM) protocol for wort production. The utility of the CM protocol for wort generation in malting quality assessments comes not from any direct reflection of commercial mashing conditions, but rather as an industry standard, commonly used protocol to generate worts that can be interpreted by maltsters and brewers in light of their organization-specific processes. The CM protocol involves multiple temperature steps and controlled-rate heating or cooling temperature ramps, requiring relatively complex temperature controls. In contrast, the isothermal (65°C) hot water extract (HWE), originally developed by the IoB, is much less widely used to evaluate malting quality. The simpler (isothermal) conditions make the HWE easier to execute than the CM and much more amenable to simple laboratory instrumentation compared to the more complex instrumentation required to implement a CM. In this presentation we compare the analytical results from worts produced by standard CM protocols with those from standard-scale (25 g) and reduced-scale (0.2 g) HWE protocols. Through this comparison we will evaluate the ability of the HWE to predict the results from a CM, and thus its utility as a simple pre-screen for malting quality.

*Mark Schmitt has been a research chemist with the USDA Agricultural Research Service at the Cereal Crops Research Unit in Madison, WI, since 2003. His research interests relate to the roles of barley proteinases in malting quality, as well as methods to miniaturize and simplify malt quality analyses.*

## P-68

### Studies on the utility of $\beta$ -amylase 1 intron III sequences as markers for $\beta$ -amylase activity and thermostability, diastatic power, and malt quality

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The third intron of barley (*Hordeum vulgare* L.)  $\beta$ -amylase 1 (Bmy1) is extremely polymorphic. The use of specific insertion/deletions (indels) in the third intron as markers for cultivar development has been recommended based on associations with  $\beta$ -amylase activity and thermostability. The third intron of Bmy1 was categorized into four alleles (Bmy1.a, Bmy1.b, Bmy1.c, and Bmy1.d) based on indels of 126, 38, 11, and 21 bp. However, the Bmy1.d allele has only been found in one accession and was not analyzed in this study.  $\beta$ -Amylase activity and thermostability were assayed in composite grain samples from 22 North American malting cultivars and 12 wild barley genotypes to determine if the third intron could be used as a predictor of activity and thermostability and, thus, be a reliable marker for marker-assisted selection. North American malting cultivars were found to carry only the Bmy1.a (14 cultivars) and Bmy1.b (8 cultivars) alleles, whereas the wild barleys were found to have the Bmy1.a (three genotypes), Bmy1.b (three genotypes), and Bmy1.c (six genotypes) alleles. Broad ranges of  $\beta$ -amylase activity and thermostability were observed in both wild and cultivated genotypes. Significantly different activities were observed in cultivars carrying either the Bmy1.a or Bmy1.b allele when calculated on a fresh weight (FW) basis and the Bmy1.a allele when calculated on a protein basis. Significantly different thermostabilities were observed in cultivars carrying the Bmy1.a allele. Significantly different activities were found in wild barley genotypes with any of these three alleles when calculated on a FW basis, yet only in those with the Bmy1.c allele when calculated on a protein basis. Significantly different thermostabilities in wild barley genotypes carrying either the Bmy1.b or Bmy1.c allele were observed. In another study, malting quality data were collected on malts from three barley (*H. vulgare*) breeding program trials (165 lines) containing both winter/facultative and spring growth habits. Abundant malting quality variation, including  $\beta$ -amylase activity, exists in the spring barley germplasm (114 lines) despite the parents all carrying the Bmy1.a allele. In the winter/facultative germplasm, broad ranges were also observed for malting quality traits,  $\beta$ -amylase activity, and thermostability, despite 47 of 51 lines carrying the Bmy1.a allele. In conclusion, broad ranges were observed for  $\beta$ -amylase activity, thermostability, and other malting quality traits that indicate the Bmy1 intron III alleles are not reliable predictors of malting quality traits and, thus, would not be a useful as a selection tool for breeding elite malting cultivars in the germplasm studied here.

*Cynthia Henson earned a Ph.D. degree at the University of Wisconsin-Madison and in 1986 became a research plant physiologist with the USDA-ARS-Cereal Crops Research Unit and an agronomy professor with the University of Wisconsin. She currently serves as research leader of the Cereal Crops Research Unit, Madison, WI.*

## P-69

### The effect of ethanol–sucrose interactions on specific gravity. Part 2: A new algorithm for estimating the specific gravity of aqueous solutions

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(1) The Gambrinus Company, San Antonio, TX

One method for the estimation of specific gravity is the Tabarie equation:  $SG_v = 1 + \sum (SG_{v_i} - 1)$ , which combines volume-adjusted binary solution-specific gravities ( $SG_{v_i}$ ) independently. It has been shown previously that the Tabarie equation deviates from measured values above about 5% (wt/wt) sucrose and 18% (wt/wt) ethanol due to solute

interactions. When ethanol is above 0.07 mol fraction and the total solute exceeds 30% (wt/wt) the deviation from observation becomes extreme. In this region the observed ternary solutions will appear to expand because the mixing collapse of hydrogen-bonded bulk water is overestimated by the Tabarie equation. A new algorithm is proposed that makes the assumption that changes in solution volume are related only to changes in water structure and the specific gravity of water, while holding the specific gravity of pure solutes constant. The model then takes the form of an ideal solution:  $SG = 1/(\sum X_i/SG_i)$ , where  $X_i$  is the mass fraction of species  $i$ . Ethanol–sucrose aqueous solutions up to 68.5% by weight total solute were measured using an oscillating U-tube densitometer. Ordinary least squares (OLS) regression analysis was used to fit an empirical model to the difference between the experimental specific gravity and the combined binary ideal solutions. Adjusted  $R^2 = 0.997$ ,  $SE = 2.9 \times 10^{-5}$ ,  $n = 145$ .

*Jim Hackbarth served in the U.S. Air Force as a nuclear weapons specialist and used the GI Bill to complete a master's degree in chemistry at the University of Illinois, Chicago, with research in X-ray crystallography. His first career path was as a research pharmacologist at Abbot Laboratories in North Chicago, IL, in the field of QSAR (quantitative structure activity relationships), followed by a move back home to Milwaukee, WI, to join the Jos. Schlitz Brewing Co. as a research chemist. Jim has been on corporate brewing staff member for Schlitz, Stroh's, and now The Gambrinus Company and is currently the manager of brewing development, physical chemist, in San Antonio, TX.*

## P-70

### The influence of metallic ion oxidation states and pH-value on haze formation in beer

THOMAS KUNZ (1), Patricia De Sousa Diniz (1), Frank-Jürgen Methner (1)

(1) Technical University of Berlin, Berlin, Germany

The aim of this study was to investigate the influence of the metallic ion content on the formation of complexes responsible for haze in beer and the correlation with pH value. It is generally known that haze in beer is directly related to the formation of complexes resulting from the interaction of polyphenols and protein fraction groups. However, recent publications support the theory that turbidity formation in stabilized beers could be correlated to the oxidative processes that take place after the consumption of the endogenous antioxidative potential (EAP) of a stabilized beer. The oxidative processes that occur in the Fenton and Haber-Weiss reaction system result in reaction products such as  $Fe^{+3}$ ,  $Cu^+$ , and  $OH^-$  radicals, which begin interacting and producing protein–polyphenol–metallic ion complexes that are also responsible for haze formation. Due to the complex formation among oxidized polyphenol–protein complexes and  $Fe^{+3}$ , as well as  $Cu^+$  ions, the development of chill haze can be observed. During the progress of beer aging, the oxidized polyphenol–protein–metallic ion complexes (including stable organic radicals) react with each other through attendant radical reactions, and the formation of covalent bonds occurs, converting chill haze to permanent haze. Specific storage experiments with addition of metallic ions in different oxidation states and different pH ranges were done. Results from these experiments clearly show that the turbidity formed when adding  $Fe^{+3}$  ions reaches values about two times higher than the standard beer without any addition.  $Cu^+$  ions also increase turbidity. When facing different pH ranges, higher haze formation was at around pH 3.5. At lower pH ranges, the bonding power of the  $Fe^{+3}$  ions was stronger, and the haze was more stable. However, the EAP consumption was slower because there was a lower concentration of iron ions available for the radical generation by the Fenton and Haber-Weiss reaction system. The result was later chill haze formation. Experiments at higher pH areas resulted in earlier but lower haze formation due to weaker bonding power in the metallic complex with haze-active polyphenol–protein complexes and in significantly faster consumption of the EAP because of the

higher availability of metallic ions for oxidative processes and radical generation. Furthermore, experiments with and without oxygen addition allowed us to conclude that oxygen content is the most important factor for the formation of oxidized polyphenol–protein complexes and their further complexation with specific metallic ions. In both cases (with and without oxygen addition) the metallic ions were responsible for higher haze generation by metallic ion–protein–polyphenol complexes. This research can provide further knowledge about avoiding undesired haze formation in beer and other beverages through lower metallic ion content and specific pH ranges.

*After qualifying as a certified technician in preservation engineering (1991–1993), Thomas Kunz completed his basic studies in chemistry at Isny Fachhochschule (1994–1995) and his basic studies in food chemistry at Wuppertal University (1995–1998) before starting to study food technology at Trier Fachhochschule (1998–2002). After graduating, he worked as a chartered engineer (Dipl. Ing. degree) in the field of ESR spectroscopy at the Institute of Biophysics at Saarland University (2002–2004). Since January 2005, he has been employed as a Ph.D. student at the Research Institute of Brewing Sciences at the Technical University Berlin. His main research focus lies in analyzing radical reaction mechanisms in beer and other beverages using ESR spectroscopy.*

#### **P-71**

##### **New insights for product quality of packaged beverages**

FRANK VERKOELEN (1)

(1) Norit Haffmans, Venlo, The Netherlands

In the beverage industry, monitoring the filling process is a final and important step to assure beverage quality. Major product specifications, including alcohol, extract, turbidity, CO<sub>2</sub>, and dissolved oxygen, are measured before the product enters the filler. However, one important quality parameter that has a strong influence on the shelf life and flavor stability of the beverage can only be measured after the packaging process. This is known as “total package oxygen” (TPO). The novel method for measuring TPO has proven that its results enable breweries to improve beverage quality and, at the same time, improve filler efficiency and reduce product losses.

*Frank Verkoelen studied mechanical engineering at HTS Venlo and finished in 1982. Frank began working with Haffmans BV in 1984 as a project engineer for CO<sub>2</sub> recovery. He switched to R&D in 1987 and became the R&D manager. In 2001 Frank became quality control product manager and then senior product manager responsible for sales of QC equipment and in-line equipment.*

#### **P-72 New Products and Services Poster**

##### **Real-time tasting systems**

RICHARD A. BOUGHTON (1), Bill Simpson (1), Ronald Nixdorf (1)  
(1) FlavorActiV

Breweries long ago recognized the importance of tasting throughout the brewing process. Consequently much attention has been paid to training tasters with best practice on-line systems and regularly assessing their competence. In addition the use of common terminology has reduced confusion and allowed a greater understanding between flavors detected and brewing process control. An increased understanding has been gained of the key flavors required in specific beer brands, as well as flavors that should not be present. The consequence of having professional tasters carrying out these daily tastings generates a mass of taste data often manually analyzed, slowly compiled, and then later distributed within the business. In most cases the time delay between tasting and being able to intelligently use the information for beer still in process prevents potential beneficial action being taken with beer in the brewery. This paper reports on the size of this opportunity and describes real-time taste management systems that are being installed to capture these benefits.

*Richard Boughton, current MD of FlavorActiV and originally a master brewer with Courage Breweries, has presented and published a number of papers covering yeast management, CIP systems, and container management. Since 1998 Richard has been MD of FlavorActiV, a world leader in beer tasting management with more than 1,200 brewery customers in more than 170 countries.*

#### **P-73 New Products and Services Poster**

##### **Real-time verification technology based on Persulphate**

PHILIP THONHAUSER (1)

(1) THONHAUSER USA, Inc., Harrison, NY

THONHAUSER has developed color-changing cleaning and verification chemicals based on patented Persulphate technology. This technology makes visible, and at the same time mineralizes, organic residues on hard-to-access surfaces. In addition to this, THONHAUSER is launching an iPhone application in 2010 that can “translate” the colors of the solution into numerical values. These technologies make possible an easy, fast, and accurate evaluation of the hygienic status of beer dispensing systems, bottles, kegs, lines, etc. When standing in front of a bar, a customer’s decision for that “second order” is strongly influenced by the refreshing and authentic taste of the beer—which can quite often be impaired by microbiological contamination inside the dispensing system. If customers complain about poor beer quality, one might wish “to have a look inside the line.” Conventional line cleaners, however, using generic chemical compounds (like acids, caustics, chlorine, etc.), pay no attention to monitoring or validating cleaning efficacy. Soaking and recirculation times are mostly set based on experience or estimates. But, when you think about it, isn’t it obvious that the correct cleaning time must be determined by the degree of pollution inside the line? The dirtier the line, the greater the cleaning task. That’s why THONHAUSER has dedicated its research in beverage hygiene science to draft beer dispensing hygiene and has developed two products that help keep up the quality of kegged beer after it has left the brewery: TM DESANA MAX is an alkaline indicator and cleaner based on the THONHAUSER Persulphate technologies. As soon as the TM DESANA MAX cleaning solution gets in contact with organic residues in beer lines, the original purple color changes to green (polluted) and further to yellow (heavily polluted). In addition, TM DESANA MAX effectively dissolves and removes all organics at the same time without chlorine or any surfactants! Every color species corresponds exactly to a certain amount of organics dissolved in the used cleaning solution. While TM DESANA MAX is a cleaning and indicator solution that changes color when organic residues are in the lines, TM puriSCOPE is the tool to translate the colors into data. It measures the hygienic conditions of all internal surfaces instantly and delivers numerical results in real time (in microgram organics per square centimeter surface or milligram per liter). It’s a set of tools for sampling purposes (sampling glasses, light panel, etc.) and an iPhone with the pre-installed verification software. The easy-to-use operation with the iPhone makes it a handy tool—no microbiological training is necessary to perform an accurate hygiene evaluation. These two technologies make precise and instant verification of the status of clean and, at the same time, an easy cleaning operation of the (whole) dispensing system possible for the first time in the United States.

*Philip Thonhauser has an education in business management and attended the Chemistry School in Vienna. He started his career at his father’s company, THONHAUSER Co. Ltd. in 1996 and became CEO of the company in 2001. The company specializes in cleaning and validation technologies for the food and beverage industries and has developed the worldwide patented THONHAUSER Persulphate technologies for in-line cleaning and validation. After Philip allocated considerable resources for R&D to develop a chlorine-free, color-changing cleaning and verification technology, THONHAUSER presented the new generation of beverage line treatment products in 2000. With the brand TM DESANA MAX indicator and cleaner, THONHAUSER won the trust of Heineken and*

*Coca Cola in Austria, Switzerland, and Germany from the start. Later the company developed technologies to convert the colors of the indicator substance into numbers and, thus, make accurate, measurable in-line verification possible. In 2001 THONHAUSER was invited to be a board member of the German DIN (German Industrial Norms) Association. Under the strong participation of THONHAUSER, a special DIN (6650) norm for the hygiene of draft installations has been developed and was published in 2006. In the DIN 6650 standard, the color indicator is mentioned as an official validation tool for the proof of clean in dispensing systems.*

**P-74 New Products and Services Poster**

**Rebiana: A new, natural, zero-calorie sweetener for beverages**

JOHN C. FRY (1)

(1) Cargill Health & Nutrition

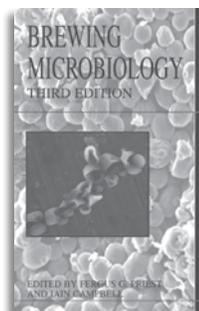
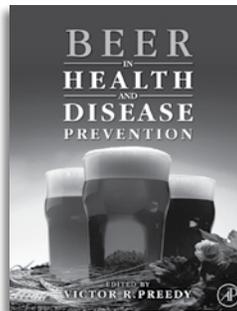
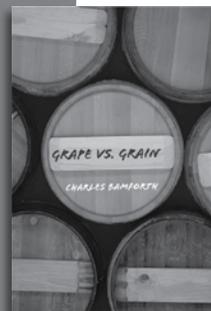
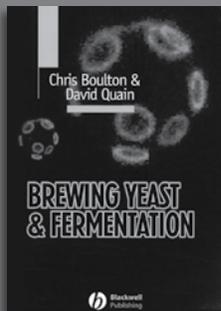
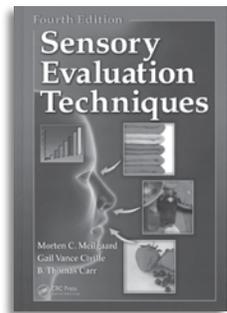
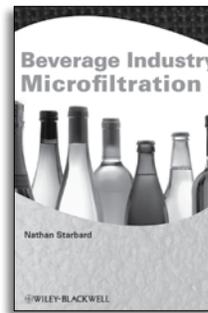
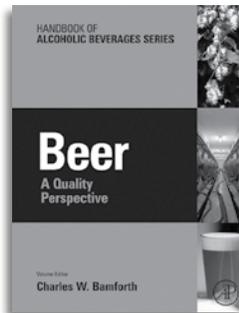
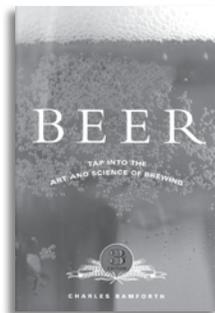
Rebiana is the common name for high-purity rebaudioside A, a natural, GRAS, zero-calorie sweetener, newly available to U.S. beverage manufacturers. Rebiana is the best-tasting member of a group of related, potentially sweet, steviol glycosides secreted in the leaves of the stevia plant (*Stevia rebaudiana*). The origins, extraction, and purification of rebiana will be described, contrasting this route with processes used for other

stevia leaf extracts and clarifying the nomenclature used by suppliers. The key properties of rebiana relevant to beverage manufacture will be outlined. The latter will include sweetness potency (concentration-response curve and Beidler equation), taste quality, solubility, and stability.

*John Fry is the principal consultant to Cargill Health & Nutrition and founding director of Connect Consulting, providing consulting services in food ingredient innovation and technical research management. He is an internationally acknowledged expert on high-potency sweeteners. John has been personally involved as a senior consultant in the technical marketing of aspartame, acesulfame-K, Twinsweet, sucralose, and stevia/rebiana worldwide. Previously John assisted in the start-up of the Holland Sweetener Company and was its director of scientific and technical services for 10 years. Prior to this John was a senior manager at Leatherhead Food Research. He obtained his B.S. and Ph.D. degrees in food science from Leeds University, U.K. He is also a chartered chemist, a Fellow of the Royal Society of Chemistry, and a Fellow of the U.K. Institute of Food Science and Technology. He speaks and trains widely on sweeteners, sweetness, and calorie control and has authored more than 70 publications, ranging from research studies and patents to trade press articles.*

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Thank you to all of the volunteers who make ASBC a valuable society. And a very special thank you to the Program Committee that made this meeting possible and to the officers, committee chairs, and section chairs who pull everything together.

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# ASBC Scholarship and Award Recipients

Every year ASBC recognizes those who show outstanding promise, have contributed valuable research to the industry, or have enhanced the brewing industry with their contributions. Congratulations and sincere thanks to this year's recipients.

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## 2010 Eric Kneen Memorial Award Recipients

The winners of the 2010 ASBC Eric Kneen Memorial Award are H. Kojima, H. Kaneda, J. Watari, Y. Nakamura, and T. Hayashi for their article, "Relationships Among Throat Sensation, Beer Flavor, and Swallowing Motion While Drinking Beer." The article was published in the *Journal of the American Society of Brewing Chemists* (Vol. 67, No. 4, pp. 217-221).

The ASBC Eric Kneen Memorial Award is presented to the author(s) of the best paper published in the *Journal of the American Society of Brewing Chemists* during the previous calendar year. The Awards Committee is composed of the *ASBC Journal's* editor-in-chief, the Editorial Board, and the Technical Committee. The team will receive \$1,000, and each person will receive an engraved plaque.



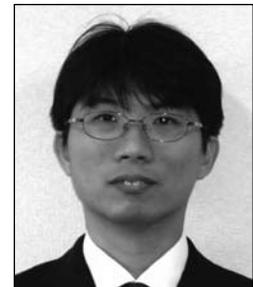
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