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New England IPA: The Hop Aroma Champion of Beers

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ABSTRACT

To investigate their typical aroma profiles, commercial samples of New England India Pale Ale (NEIPA) were analyzed by headspace GC-MS and GC-MS/MS methods. As result of this study, the NEIPAs showed record levels of nonpolar terpenes like myrcene (up to 26 mg/L), alpha-humulene (up to 2.3 mg/L), or beta-caryophyllene (up to 1.6 mg/L). Polar monoterpene alcohols like linalool (up to 2.2 mg/L) or geraniol (up to 0.6 mg/L) and thiols like 4-mercapto-4-methylpentan-2-one (up to 150 ng/L) were also present in relatively high concentrations, which can only be achieved in an extremely dry-hopped (West Coast) IPA. Such high concentrations clearly exceeded the corresponding odor threshold values of these hop aroma components. Reduction in aroma compounds after haze removal by centrifugation proved to be dependent on polarity. Average losses of nonpolar terpenes were in the range of 85% (for myrcene) to 79% (both for alpha-humulene and beta-caryophyllene), whereas more polar components like ketones and esters were reduced to a lower extent (41 to 25%). Monoterpene alcohols and thiols experienced little to no loss in concentration after centrifugation. During storage for 6 months at 5°C the concentrations of thiols and terpenes decreased by more than 50%, but monoterpene alcohols exhibited almost full stability within this period. Such a storage behavior of hop-derived aroma components is not unusual and comparable with other beer styles.

Keywords: New England IPA, NEIPA, myrcene, 4-mercapto-4-methylpentan-2-one, geraniol, beta-citronellol

Introduction

It is well known that dry-hopped beers are pronounced in hop aroma, and this is especially true with India Pale Ales (IPAs). Because its cloudy variant, called New England IPA (NEIPA), is typically brewed with multiple hop additions late in the whirlpool and during active fermentation, it can be assumed that exceptional hop aroma can also be found in this type of craft beer. NEIPA is also called “Juicy IPA” or “Hazy IPA.” This beer style is unfiltered and produced with up to 50% high-protein adjuncts such as oats and/or wheat. Little to no hops are added to the kettle during the boil, but instead a large portion of the total hop dosage is added to the whirlpool. Such a hopping regime retains much of the hop oil compounds and minimizes isomerization of alpha-acids. At least part of dry-hopping is typically performed during active fermentation. This allows for yeast biotransformation processes like the release of glycosidically bound monoterpene alcohols or biosynthesis of citronellol from geraniol (17). Total rates of hop dosage are up to 2 kg/hL (~5 lbs/bbl) or sometimes even more.

Altogether such a recipe results in a hazy beer showing significant differences in flavor and composition as compared with the perhaps more familiar “West Coast” IPAs (WC IPAs). Usually, NEIPAs are not perceived as being very bitter but can impart fruity and juicy flavors. WC IPAs have higher concentrations of iso-alpha-acids but lower concentrations of polar humulinones and of less polar hop compounds (like alpha-acids, beta-acids, and xanthohumol) than NEIPAs. This was already reported on by Maye and Smith (9). The “hidden secret” learned from this publication is that the haze in NEIPAs can act as a carrier and increase the concentration of nonpolar compounds. In that study, beer samples were only analyzed by high-performance liquid chromatography (HPLC). Now in this paper the results of gas chromatographic (GC) analysis on various NEIPA samples are presented to give a deeper insight in the hop aroma composition of this beer style.

Many different methods for analysis of hop aroma components can be used (12), but the European Brewery Convention (EBC) Analysis Committee recently recommended a method based on headspace-trap GC in combination with mass spectrometry (MS) detection (4). This method is useful for analyzing most of the typical hop oil components including terpenes (e.g., myrcene), sesquiterpenes (e.g., alpha-humulene, beta-caryophyllene), monoterpene alcohols (e.g., linalool, geraniol, beta-citronellol), esters (e.g., 2- and 3-methylbutyl isobutyrate), or ketones (e.g., 2-undecanone), as published previously (15). However, for the hop oil fraction of thiols (e.g., 4-mercapto-4-methylpentan-2-one [4-MMP], also known as 4-methyl-4-sulfanylpentan-2-one), only present at trace levels in hops and beer, a more sophisticated method based on sample derivatization followed by GC in combination with tandem MS (GC-MS/MS) is necessary and was applied for this study on NEIPA as well (3).

For the substance class of aroma-active thiols derived from hops, 4-MMP can serve as lead component. In its free form, this thiol proved to be present in hops in a wide range of concentrations but was clearly dependent on growing area and variety (11). Moreover, its transfer to beer via dry-hopping can result in concentrations greatly exceeding its odor threshold value of approximately 1 ng/L (ppt) (7). Hops also contain 4-MMP (and other thiols) in bound forms (to amino acids, like glutathione or cys-

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teine), and it can be released from these sources by yeast activity (14). Another prominent thiol, 3-mercaptohexan-1-ol (3-MH), can be transferred to beer not only via hops but also from sources present in malt (6). During fermentation, this thiol is known to be converted to 3-mercaptohexyl acetate (3-MHA). Although this biosynthesis results in lowering the threshold value (3-MHA, 5 ng/L; 3-MH, 55 ng/L), it is higher compared with 4-MMP but still in the ppt range. On the other hand, all terpenes, monoterpene alcohols, esters, and ketones mentioned above show odor threshold concentrations in beer in the ppb (μg/L) range, much higher thresholds than the thiol compounds (ppt).

Materials and Methods

The commercial beers investigated in this study were supplied by several U.S. breweries. All NEIPAs were hazy. They were either analyzed directly without any sample preparation or after centrifugation and removal of the precipitate. Centrifugation was done for 15 min at 3,000 rpm. All samples (without and after centrifugation) were analyzed in duplicate, but only the mean values are reported on.

Analysis of most hop aroma components was according to Analytica-EBC Method 9.49 (4). A detailed description of the method and the equipment used for this analysis is also given in a paper of Schmidt and Biendl (15). It combines headspace-trap (Perkin Elmer, TurboMatrix HS-40) with GC-MS (Thermo Fisher Scientific, GC Thermo Focus coupled to a DSQ II quadrupole MS).

The analysis of thiols was done in the laboratory of VLB (Berlin) according to the method as published recently by Dennenloehr et al. (3). It consists of on-fiber derivatization of hop-derived thiols in beer in combination with automated headspace solid phase micro extraction (HS-SPME; Gerstel, MPS 2XL for automated HS-SPME sampling) and GC-MS/MS (Agilent Technologies, 7890B gas chromatograph interfaced to a 7000C Triple Quadrupole mass spectrometer).

Analysis of the most important bitter acids and xanthohumol in untreated beer (without centrifugation) was performed by HPLC-UV (Shimadzu, Prominence CBM-20A and SPS-20AV UV detector) according to the International Method "Bitter Compounds in Dry Hopped Beer by HPLC" just recently published by EBC (5) and ASBC (1). For xanthohumol measurement the detection wavelength was modified to 370 nm, instead of the 270 nm that is recommended for the bitter acids.

Results and Discussion

The compositions of hop bitter compounds of six commercial U.S. NEIPAs are listed in Table 1. The ranges and averages for iso-alpha acids, humulinones, alpha-acids, beta-acids and xanthohumol are very similar to the data as presented previously (9), although the investigated samples were different, and the beers were not always from the same breweries. Therefore, the selection of NEIPAs used in this study can again be considered as typical.

Table 1. Comparison of hop bitter acids and xanthohumol in NEIPAs analyzed in this study as compared to previous study (9)

	NEIPA investigated in this study (six commercial samples: NEIPA I–VI)		NEIPA investigated in previous study (12 commercial samples: NEIPA A–L)	
Compounds	Range (ppm)	Average (ppm)	Range (ppm)	Average (ppm)
Iso-alpha-acids	5–28	18	5–32	20
Humulinones	10–38	23	12–38	26
Alpha-acids	21–42	33	17–72	31
Beta-acids	2–7	4	1–14	5
Xanthohumol	1.0–1.8	1.4	0.9–3.5	2

GC combined with MS was performed on these six NEIPAs for measuring the concentrations of the monoterpene myrcene, the sesquiterpenes beta-caryophyllene and alpha-humulene, the monoterpene alcohols linalool, geraniol, and beta-citronellol, the esters 2-methylbutyl isobutyrate (2-MBIB) and 3-methylbutyl isobutyrate (3-MBIB), the ketone 2-undecanone, and the thiols 4-MMP, 3-MH, and 3-MHA.

All the analyzed single compounds are well-known from literature and can be considered as important components for several substance classes unfolding different aroma profiles in beer, mainly after dry-hopping. Whereas mono- and sesquiterpenes give herbal or spicy notes, the odor coming from monoterpene alcohols is usually described as floral or citrussy. Esters, ketones, and thiols are more responsible for fruity notes.

The odor threshold concentrations of all the aroma components (Fig. 1) analyzed in our study have been published and can be found in several sources (Table 2). Most of them are so-called key aroma compounds, which means their detected concentrations in beer can be above the corresponding threshold values. But even if such levels are not fully reached, aroma contributions can be expected via additive effects between different hop-derived volatiles (18).

Compared with the other substance classes, thiols have some special features. They are not present in all hop varieties (11), can be released from precursors during fermentation (14), and are extremely odor-active even in the ppt range (7). Another im-

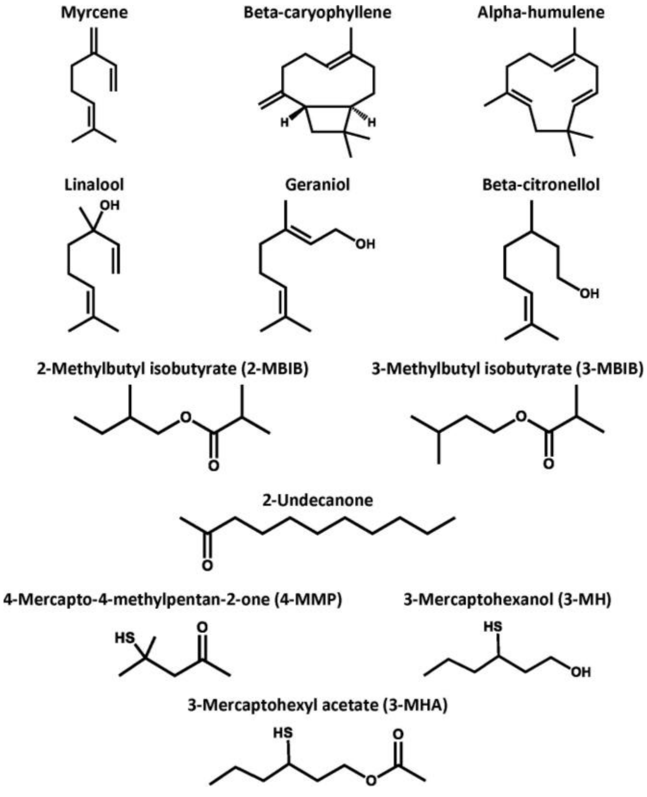

Figure 1. Chemical diagrams of aroma compounds.

portant aspect is the biosynthesis of beta-citronellol from other monoterpene alcohols (mainly geraniol) by yeast biotransformation processes. Therefore, high concentrations of beta-citronellol in beer can be attributed to hop varieties rich in geraniol (17).

The six NEIPAs analyzed in this study are based on a wide range of different brewing recipes. By far most of the hops were aroma/flavor varieties from the U.S. Pacific Northwest. Total hop additions for the several beers were between 700 and 1,700 g/hL, with the main part always added late in the brewing process (i.e., to the whirlpool, during active fermentation and afterwards).

Tables 3, 4, 5, and 6 list the single and average concentrations of the analyzed compounds in beer samples before and after centrifugation. The percent yield is calculated by dividing the concentration of hop oil measured in the supernatant after centrifugation by the concentration of hop oil measured before centrifugation.

Table 2. Odor threshold concentrations (literature data)

Compound	Odor threshold in ppb	References
Mono- and sesquiterpenes		
Myrcene	9–1,000	2
Beta-caryophyllene	160–420	2
Alpha-humulene	747	2
Monoterpene alcohols		
Linalool	2–80	2
Geraniol	4–300 / 6 / 4	2 / 17 / 7
Beta-citronellol	9–40 / 8	2 / 17
Esters and ketones		
2-Methylbutyl isobutyrate (2-MBIB)	78	16
3-Methylbutyl isobutyrate (3-MBIB)	100	2
2-Undecanone	7	8
Thiols		
4-Mercapto-4-methylpentan-2-one (4-MMP)	0.0015	7
3-Mercaptohexanol (3-MH)	0.055	7
3-Mercaptohexyl acetate (3-MHA)	0.005	6

The recovery rate after centrifugation is dependent on polarity. Because polar monoterpene alcohols and thiols are very soluble in beer, they are hardly removed by centrifugation. On the other hand, the most nonpolar mono- and sesquiterpenes are only recovered at a rate of 15 to 20%, and the yield of middle polar esters and ketones is in between, at 75 and 59%, respectively. But even after centrifugation, the detected concentrations of most compounds are still above the corresponding odor threshold levels as presented in Table 2.

This is almost always true for monoterpene alcohols, esters, and ketones, for the monoterpene myrcene and the thiol 4-MMP. In contrast, the concentrations of the two sesquiterpenes and the thiol 3-MH are usually clearly below their odor threshold levels, and the acetate of 3-MH could not be detected at all.

NEIPAs II–VI were also analyzed after storage at 5°C for 3 and 6 months, respectively. As presented in Figure 2, monoterpene alcohols showed the best storage stability. Stable levels or even increases of linalool or geraniol can be explained by the release of free alcohols from their glycosidic precursors even during storage (17). Ketones and esters proved to be less stable, with decreases between 31 and 42% after 6 months. And both the concentrations for thiols and terpenes were reduced by more than half within this period.

Such a storage behavior of NEIPA is not atypical as compared with other beer styles. We found very similar stabilities of hop aroma components in dry-hopped beers, both in Pilsener and Pale Ale (15). Rettberg et al. investigated the storage stability of 2-MBIB in 11 ales and reported on an average reduction of 60% during a storage period of 24 months at 4°C (13). According to Reglitz et al., the 4-MMP concentrations in a dry-hopped Pilsener had dropped to 59% (filtered beer) and 67% (unfiltered beer) after 3 months of storage at 5 °C, and a further decline was observed when the beers were stored for an additional 3 months (10).

Table 3. Concentrations (ppb) of mono- and sesquiterpenes in six commercial NEIPAs without and after centrifugation

	Myrcene		Beta-caryophyllene		Alpha-humulene	
Sample	Without centrifugation	After centrifugation	Without centrifugation	After centrifugation	Without centrifugation	After centrifugation
NEIPA I	1,409	201	43	15	102	31
NEIPA II	7,107	1,584	188	50	206	52
NEIPA III	18,498	1,827	1,558	229	2,261	324
NEIPA IV	2,450	1,345	158	82	267	130
NEIPA V	26,390	2,019	301	77	572	143
NEIPA VI	7,192	2,598	153	49	101	43
Average NEIPA I–VI	10,508	1,596	400	84	584	121
Yield (*n* = 6)	15%		21%		21%	

Table 4. Concentrations (ppb) of monoterpene alcohols in six commercial NEIPAs without and after centrifugation

	Linalool		Geraniol		Beta-citronellol	
Sample	Without centrifugation	After centrifugation	Without centrifugation	After centrifugation	Without centrifugation	After centrifugation
NEIPA I	714	720	35	34	16	15
NEIPA II	615	592	470	437	87	87
NEIPA III	553	534	308	393	55	44
NEIPA IV	2,183	2,204	307	350	13	16
NEIPA V	851	813	338	301	31	28
NEIPA VI	1,203	1,191	592	526	21	21
Average NEIPA I–VI	1,020	1,009	342	340	37	35
Yield (*n* = 6)	99%		99%		95%	

Finally, a comparison between the results of this study and the average composition of commercial WC IPAs is presented in Table 7. Such data have been collected in the past few years also based on the same headspace-trap GC-MS method as used for NEIPA analysis in this study. On average, NEIPAs are showing much higher concentrations for all hop aroma lead components as compared with WC IPAs. This difference is most pronounced for the nonpolar mono- and sesquiterpenes, whose

Table 5. Concentrations (ppb) of esters and ketones in six commercial NEIPAs without and after centrifugation

	3-Methylbutyl isobutyrate		2-Methylbutyl isobutyrate		2-Undecanone	
Sample	Without centrifugation	After centrifugation	Without centrifugation	After centrifugation	Without centrifugation	After centrifugation
NEIPA I	81	67	239	193	42	23
NEIPA II	184	127	521	369	21	16
NEIPA III	109	81	243	177	35	21
NEIPA IV	63	50	189	145	29	16
NEIPA V	96	65	210	142	46	28
NEIPA VI	173	146	822	650	23	12
Average NEIPA I–VI	118	89	371	279	33	19
Yield (*n* = 6)	75%		75%		59%	

Table 6. Concentrations (ppt) of thiols in six commercial NEIPAs without and after centrifugation

	4-Mercapto-4-methylpentan-2-one		3-Mercaptohexanol		3-Mercaptohexyl acetate	
Sample	Without centrifugation	After centrifugation	Without centrifugation	After centrifugation	Without centrifugation	After centrifugation
NEIPA I	39	39	19	17	<5	<5
NEIPA II	107	93	37	34	<5	<5
NEIPA III	57	49	15	15	<5	<5
NEIPA IV	33	27	17	13	<5	<5
NEIPA V	149	163	30	35	<5	<5
NEIPA VI	136	118	35	56	<5	<5
Average NEIPA I–VI	87	82	26	28	<5	<5
Yield (*n* = 6)	94%		108%		–	

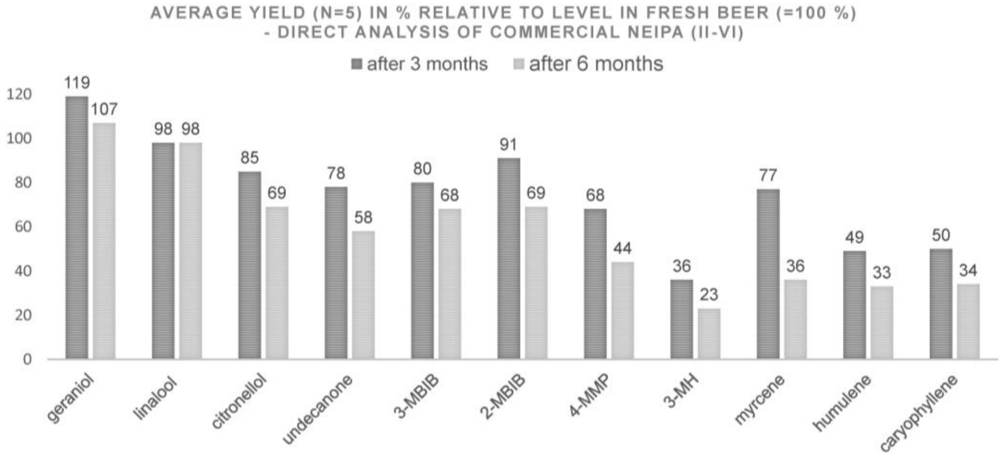

Figure 2. Storage stability of important hop aroma components in New England IPA after storage at 5°C.

Table 7. Average concentrations (ppb) of hop aroma components in six commercial NEIPAs compared with seven commercial West Coast IPAs (WC IPAs)

	Commercial NEIPAs I–VI (average: *n* = 6)		Commercial WC IPAs (average: *n* = 7)	Difference factor: concentrations NEIPA / IPA	
Sample	Without centrifugation	After centrifugation	Without centrifugation	Without / without centrifugation	After / without centrifugation
Myrcene	10,508	1,596	1,577	6.7	1.0
Caryophyllene	400	84	35	11	2.4
Humulene	584	121	59	10	2.1
Linalool	1,020	1,009	488	2.1	2.1
Geraniol	342	340	229	1.5	1.5
Citronellol	37	35	10	3.7	3.5
2-MBIB	371	279	209	1.8	1.3
3-MBIB	118	89	81	1.5	1.1
Undecanone	33	19	19	1.7	1.0

concentration is increased by the haze. But even after centrifugation the average levels of the NEIPA supernatants are almost always greater than those in untreated WC IPAs.

Concentrations for most monoterpene alcohols, thiols, esters, and ketones typical for NEIPA may only be reached with extremely dry-hopped WC IPAs. But beta-citronellol is the exception. Significantly higher concentrations of this compound in NEIPA compared with WC IPA indicates that large hop additions have taken place and that monoterpene alcohol biotransformation processes probably occurred during active fermentation.

Overall, the typical composition of hop aroma components in NEIPA is quite different compared with WC IPA, and their total concentration is hardly reached in any other type of beer.

In conclusion, no beer contains more hop aroma components than NEIPA, and this style can be considered the "hop aroma champion of beers."

REFERENCES

1. ASBC Methods of Analysis, online. Beer Method 23G. Bitter compounds in dry-hopped beer by HPLC (IM). Approved 2020. American Society of Brewing Chemists: St. Paul, MN, U.S.A.
2. Biendl, M., Engelhard, B., Forster, A., Gahr, A., Lutz, A., Mitter, W., Schmidt, R., and Schoenberger, C. 2014. Chemical compounds in hops. Pages 47-93 in: Hops – Their Cultivation, Composition and Usage. Fachverlag Hans Carl GmbH: Nuremberg, Germany.
3. Dennenloehr, J., Thoerner, S., and Rettberg, N. 2020. Analysis of hop-derived thiols in beer using on-fiber derivatization in combination with HS-SPME and GC-MS/MS. J. Agric. Food Chem. 78:15036-15047.
4. European Brewery Convention. Analytica-EBC. Beer Method 9.49. Hop aroma components in beer by headspace-trap gas chromatography (Guideline Method) – 2018. EBC: Brussels, Belgium.
5. European Brewery Convention. Analytica-EBC. Beer Method 9.50. Bitter compounds in dry-hopped beer by HPLC (IM) – 2020. EBC: Brussels, Belgium.
6. Kishimoto, T., Morimoto, M., Kobayashi, M., Yako, N., and Wanikawa, A. 2008. Behaviors of 3-mercaptohexan-1-ol and 3-mercaptohexyl acetate during brewing processes. J. Am. Soc. Brew. Chem. 66:192-196.
7. Kishimoto, T., Wanikawa, A., Kono, K., and Shibata, K. 2006. Comparison of the odor-active compounds in unhopped beer and beers hopped with different hop varieties. J. Agric. Food Chem. 54:8855-8861.
8. Leffingwell, J. C., and Leffingwell, D. 1991. GRAS flavor chemicals – Detection thresholds. Perfumer&Flavorist 16:1-19.
9. Maye, J. P., and Smith, R. 2018. Hidden secrets of the New England IPA. Tech. Q. Master Brew. Assoc. Am. 55:88-92.
10. Reglitz, K., Lemke, N., Hanke, S., and Steinhaus, M. 2018. On the behavior of the important hop odorant 4-mercapto-4-methylpentan-2-one (4MMP) during dry hopping and during storage of dry hopped beer. BrewingScience 71:96-99.
11. Reglitz, K., and Steinhaus, M. 2017. Quantitation of 4-methyl-4-sulfanylpentan-2-one (4MSP) in hops by a stable isotope dilution assay in combination with GC×GC-TOFMS: Method development and application to study the influence of variety, provenance, harvest year, and processing on 4MSP concentrations. J. Agric. Food Chem. 65: 2364-2372.
12. Rettberg, N., Biendl, M., and Garbe, L.-A. 2018. Hop aroma and hoppy beer flavor: Chemical backgrounds and analytical tools – A review. J. Am. Soc. Brew. Chem. 76:1-20.
13. Rettberg, N., Schubert, C., Dennenloehr, J., Thörner, S., Knoke, L., and Maxminer, J. 2020. Instability of hop-derived 2-methylbutyl isobutyrate during aging of commercial pasteurized and unpasteurized ales. J. Am. Soc. Brew. Chem. 78:175-184.
14. Roland, A., Viel, C., Reillon, F., Delpech, S., Boivin, P., Schneider, R., and Dagan, L. 2016. First identification and quantification of glutathionylated and cysteinylated precursors of 3-mercaptohexan-1-ol and 4-methyl-4-mercaptopentan-2-one in hops (*Humulus lupulus*). Flavour Fragr. J. 31:455-463.
15. Schmidt, C., and Biendl, M. 2016. Headspace trap GC-MS analysis of hop aroma compounds in beer. BrewingScience 69:9-16.
16. Takoi, K., Degueil, M., Shinkaruk, S., Thibon, C., Kurihara, T., Toyoshima, K., Ito, K., Bennetau, B., Dubourdieu, D., and Tominaga, T. 2009. Specific flavor compounds derived from Nelson Sauvin hop and synergy of these compounds. BrewingScience 62:108-118.
17. Takoi, K., Itoga, Y., Koie, K., Takayanagi, J., Kaneko, T., Watanabe, T., Matsumoto, I., and Nomura, M. 2017. Systematic analysis of behaviour of hop-derived monoterpene alcohols during fermentation and new classification of geraniol-rich flavour hops. BrewingScience 70:177-186.
18. Takoi, K., Itoga, Y., Takayanagi, J., Matsumoto, I., and Nakayama, Y. 2016. Control of hop aroma impression of beer with blend-hopping using geraniol-rich hop and new hypothesis of synergy among hop-derived flavour compounds. BrewingScience 69:85-93.