Changes of Hop-Derived Aroma Compounds in India Pale Ale during Brewing and Storage

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***Abstract*—Various beers contain many aroma compounds derived from malts, hops, yeast fermentation and other materials. Among these aroma compounds, terpenoids are mainly derived from hops. The aging of India Pale Ale (IPA) was accelerated to study the changes in aroma compounds derived from hops during storage. We focus on changes and contribution of terpenes (β-myrcene, caryophyllene, humulene, β-pinene) and monoterpene alcohols (linalool, geraniol) to hopped beer flavor, the beer quality during storage was also investigated using sensory methods. The results showed that the content of aroma compounds decreased during main fermentation, and increased after dry hopping, and then decreased, only the base beer and 1-month beer had greater sensory quality. Analysing both the correlation of aroma compounds and the correlation between the aroma compounds and beer aroma, it was found that there was a correlation between most aroma compounds. The results showed that the content changes of the aroma compounds from hops during the brewing and storage process provided a basis for future related research.**

***Index Terms*—beers; aroma compounds; storage; correlation**

I. Introduction

Aroma compounds in hops are important because they make a major contribution to the quality of the final product [1]. The aroma of beer has been studied extensively for several decades, and compounds have been identified that are responsible for flavor [2]-[5]. Hops are added during the brewing process for bittering purposes but also to impart hop-derived aroma to the final beer product [6], [7]. There are two practices of hopping, and they each give a different type of hop-derived aroma in the beer. A "dry" hop aroma is imparted by the addition of whole hops or hop pellets during the fermentation process and is described as resinous with the aim to preserve more aroma [8]. Hops can also be added to the boiling kettle (generally 2-3 times), which is called "conventional kettle hopping". When hops are dosed at the onset of boiling (5 min after boiling), both bitterness and aroma, which are characterized by spicy/herbal notes [9]-[11], are imparted to the final beer product. The composition of hop essential oil is complex, with 485 compounds currently identified [12], [13]. Fritsch and Schieberle proposed that β-myrcene and linalool are the most potent odorants in the hop cones (2005). However, it was noted that most of the hydrophobic terpene hydrocarbons from hops, including β-myrcene and α-humulene, would not remain as part of the finished beer after the brewing process [13]. Terpene alcohols are more hydrophilic than terpene hydrocarbons and are easier to retain in the wort [14]. Linalool has been found in many kinds of beers, and it is considered to be the most important aroma compounds derived from hops [15], [16]. Cascade, a type of hop from America, contains both linalool and geraniol in the hops and finished beer [14], [16]. King and Dickinson (2000) have reported the changes of geraniol and nerol during fermentation by lager yeast. Geraniol was found to be could be transformed into β-citronellol and linalool and nerol could be converted into linalool and α-terpineol. The most abundant terpene hydrocarbon in hop essential oil was myrcene and humulene and caryophyllene were almost completely removed during fermentation of hopped wort by adsorption of the hydrophobic yeast cells and migration to the foam layer [13], [16]-[19].

In this study, we researched the changes in the relative amount (%) of aroma compounds during brewing and storage in IPA. The changes in the aroma compounds during storage were further investigated by GC-MS. Finally, the correlation between the aroma compounds and aroma substances and beer quality was studied in detail.

II. Materials and Methods

A. Hop Pellets

Citra hops were provided by Dr. Patrick Ting (*MillerCoors, Milwaukee, WI, U.S.A*). They were grown in the Yakima Valley, Washington, USA. These are a type

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of cascarilla hops derived from the hybridization between Mf (Hallertau Mittelfrueh) and US Tettnag. Citra hops were released in 2008 by Hop Breeding Company (jointly operated by John Haas and Select Botanicals Group, LLC.). The pellets (100 g) were stored in a freezer (-18 ℃) to avoid the oxidative degradation of the hop oil compounds (Yakima Chief-Hopunion LLC., 2015).

B. Pilot-scale Brewing

The wort was prepared using commercially available 100% malts, the proportion of malt and water was 1:4. The amount of malt was 40 kg, and the volume of water was 160 L. The malt was added at 65 ℃ and keep warm for 1 h, then heated to 72 ℃ for 10 min, and finally the temperature was increased to 78 ℃. Then the mash was transferred to the filter tank, keep for 30 min before filtration. Grains washing was conducted when the spent was coming out of the water, the number of times of washing was 3-5 times, wash water for no more than 80 L. The original gravity was adjusted to 11 °P, and the 240 L of clarified wort was boiled for 90 min. The Citra hops were added during wort boiling, the hopping rate and addition times were: 25 % at 5 min after boiling; 50% at 30 min after boiling; and a further 25% at 5 min before the end of boiling; total hop dosage was 360 g (1.5 g/L). After cooling, the fermentation was started by adding 1.8 × 107 cells/ml of ale yeast (German yeast NO 303) to the cooled wort. IPAs were fermented at 22 ℃ for one week, then dry hopped with hops (1.5 g/L), cooled to 8 ℃ for 3 days, and lagered for one week. Filtration and bottling were conducted using the pilot-scale equipment under anti-oxidative conditions. Each storage drop was blended with water and adjusted to an alcohol content of 4% by weight (finished beer). Samples of hopped wort, fermenter drop, dry hopping, storage drop and finished beer were taken and analyzed by GC-MS for hop volatiles profile at New Hengji Biotechnology Co.(Liaoyang Road, 200 meters south of Guantang Bridge, Liaoyang City, Shandong Province).

C. Forced Aging Experiment

The forced aging method was used to accelerate aging of the beer. Thirty bottles of 750 ml were prepared, each containing 700-720 ml of beer. The beer samples were treated with hot and cold cycling at 0 ℃ and 60 ℃. Each cycle (stored at 0 ℃ for 24 h and 60 ℃ for 24 h, total of 48 h) was deemed to correspond to beer kept for 1 month at room temperature (1 cycle=1 month). The number of cycles equated to the shelf life in months. After one cycle, 3 bottles were removed for SPME-GC-MS analyses and sensory evaluation. This aging was repeated for the beer representing storage for 2, 3, 4, 6, 9 and 12 months, and SPME-GC-MS analyses and sensory evaluations were conducted at each of these storage times.

D. Collection of Aroma Compounds

SPME was accomplished using a Gerstel MPS multifunctional injector. A total of 2 g NaCl was added into a 20 ml headspace bottle, with 5 ml of the air removed when beer samples were added. A total of 50 µl of the internal standard substance was added and the sample bottle placed on the automatic sampler plate. The automatic sampler moved the sample bottle into the incubator, inserted the extraction head, and completed the adsorption under defined conditions, the fiber head was inserted into the GC injection port for 2 min at 250 ℃.

E. Quantification of Aroma Compounds

GC-MS analyses were carried out using a 6890N gas chromatograph (Agilent Technologies Palo Alto, CA). The carrier gas was helium with a column-head pressure of 15 psi and a flow rate of 1.8 mL/min. Gas chromatography conditions: the capillary column used was DB-WAXETR (60 m x 0.32 mm x 0.25 m); the carrier gas was high purity helium with a constant flow mode and a flow rate of 1.5 ml/min under non-shunt injection. The column temperature was 37 ℃ (10 min) and the temperature was increased to 120 ℃ at 8 ℃/min. 1 ℃/min to 155 ℃, and 5 ℃/min to 195 ℃.

The mass spectrometric conditions include an electron bombardment ion source (EI), electron energy of 70 eV, an ion source temperature of 230 ℃, and a temperature of the stage four rod of 150 ℃. The full scan mode (SCAN) and scanning range of 40~500 (m/z) were used in the qualitative analysis. The selected ion monitoring model (SIM) was used for quantification, and 1 quantitative ion was selected for each compound, (2-3 qualitative ions were used).

F. Sensory Evaluation of the Beer Samples

Sensory evaluation is an important way to judge beer quality and it is of vital importance to involve a group of professionals to establish a sound evaluation system. In this study, the beers were stored at 0 ℃ for 24 h for equilibration prior to the sensory evaluation. Two hours before sensory evaluation, the beers were removed from the refrigerator. Each sensory evaluation was performed by 12 trained panelists (6 males and 6 females), and the appearance and taste of the samples were scored. The average was determined after removing the highest and lowest values. The standard of the evaluation followed the sensory attributes of "look (foam, color and clarity), smell (aroma), and taste (flavour)".

G. Correlation Analysis and Statistic Analysis

The pearson correlation of the aroma compounds was analyzed by using the SPSS statistical software. The factor coefficient was between 0 and 1. The higher the coefficient, the stronger the correlation. When $r=0$, there was no correlation, when $0<r\leq0.39$, there was a weak correlation, when $0.4<r\leq0.59$, there was a moderate correlation, and when $0.6<r<1$, there was a strong correlation.

All analyses were performed in duplicate. The associations among the individual traits were determined with a simple linear correlation. A correlation analysis was performed using SPSS statistical software (version 16.0 for Windows, SPSS Inc., Chicago).

III. Results

A. Changes in Aroma Compounds During Boiling

Ten compounds were assessed and were regarded as the major flavor compounds in hops: terpenoids (β-myrcene, caryophyllene, humulene, β-pinene linalool, and geraniol), esters (isoamyl butyrate, isoamyl acetate, and ethyl laurate) and a ketone (2-undecanone). The content of the aroma compounds in hops was measured before boiling. These measurements were determined as the relative amount (%) in 1 g hop. From the results shown in Table I, the 10 compounds all showed a decrease after boiling. It is worth mentioning that the content of β-myrcene was the highest both before and after boiling, at 67.15% after boiling and only decreased by only 8.15%. The loss of humulene is also very low after boiling, and only decreased by 1.01%.

The other 8 substances are very low after boiling, and most of them evaporated in the high-temperature environment, with isoamyl acetate almost at a trace level after boiling. The percentage of variation in the relative amounts before boiling is also provided in Table I, and showed that isoamyl acetate and ethyl laurate had the largest proportion of relative change.

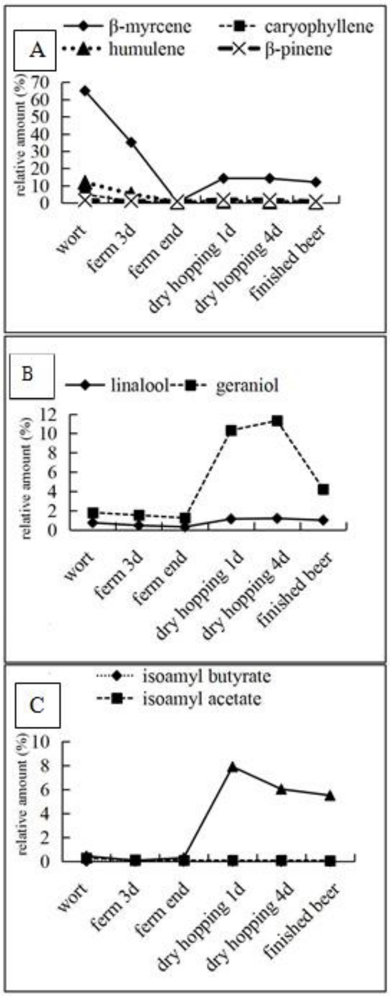
TABLE I. COMPARISON OF THE CONTENTS OF AROMA COMPOUNDS (RELATIVE AMOUNT %) DURING THE BOILING PROCESS

Volatile compounds	Before boiling(%)	After boiling(%)	Variation(%)
β-myrcene	75.2976	67.1516	-8.146
caryophyllene	10.3952	5.5786	-4.8166
humulene	13.7954	12.7871	-1.0083
β-pinene	1.8242	1.1971	-0.6271
linalool	1.1673	0.7418	-0.4255
geraniol	0.4523	0.1098	-0.3425
isoamyl butyrate	0.1724	tr	-0.1724
isoamy	3.5426	0.3	-1.3281
acetate ethyl			
laurate	2.4	0.1	-2.3
2-undecanone	2.0953	1.2211	-0.8742

[a]Legend: before boiling, calculated from the measurement of Citra hops; after boiling, calculated from the measurement of the wort with added hops (boiled at 100 ℃ for 90 min); and tr, trace.

B. Changes in Aroma Compounds During Brewing

Fig. 1 shows the change in terpenoids, esters and ketone during brewing. The content of all of the aroma compounds decreased to different degrees before dry hopping. The linalool had no obvious fluctuation during the fermentation (from wort to the end of fermentation), and the composition percentage was always below 2% throughout the brewing process. Before fermentation, linalool decreased by 58%, and further decreased from 0.74% to 0.31% after fermentation, with the content in finished beer approximately 1.35 times that of the wort (Fig. 1B). Compared with linalool, the variation in the content of geraniol was larger, declining by 30% after fermentation, and after dry hopping, the content of it increased sharply from 1.24% to 10.32% (Fig. 1B). The content of geraniol in the finished beer was appoximately 4.23%, which was more than twice that in the wort. Next, the changes of 4 terpenes (β-myrcene, caryophyllene, β-pinene, and humulene) were analysed with results shown in Fig 1A. They all decreased during fermentation, and β-myrcene had a sharp decrease, from 65.01% to 0.57%, for an overall decrease of 90%. After dry hopping, β-myrcene had increased to 14.30% from 0.57%, however, the other 3 terpenes had no obvious change and their relative amounts were very low and close to 0. The changes of esters (isoamyl acetate and ethyl laurate) are shown in Fig. 1C. There was no obvious fluctuation in their content, being always less than 1% throughout the brewing process. The relative amounts (%) of isoamyl butyrate was also very low during the fermentation period, until after dry hopping, when its content increased. Fig. 1D shows the changes of 2-undecanone, a rapid decrease was observed during the fermentation process, where it decreased from 3.15% to 0.54%, followed by a slight increase after dry hopping; with the content in the finished beer was 0.24%.



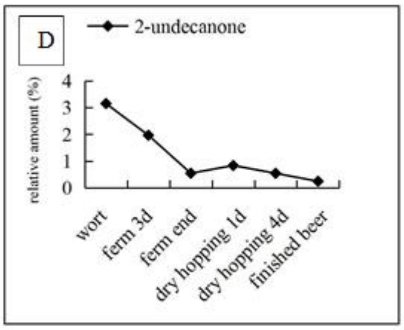
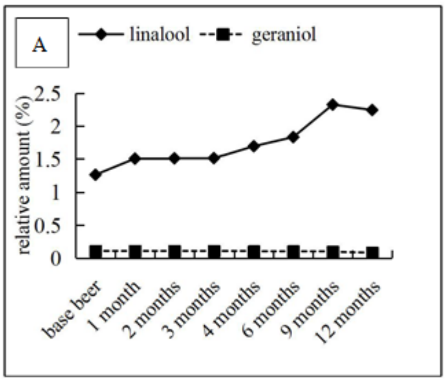


Figure 1. Comparison of aroma compounds (%) during the brewing process of hopped beer. The sampling points are respectively wort, ferm 3d, ferm end, dry hopping 1d, dry hopping 4d); (A) linalool and geraniol; (B) terpenes: β-myrcene, caryophyllene, β-pinene, and humulene; (C) esters: isoamyl butyrate, isoamyl acetate and ethyl laurate (D) 2-undecanone

C. Changes in Aroma Substances During Storage

Fig. 2 shows the changes in the content of the aroma compounds during storage. For the 2 terpene alcohols (Fig. 2A), the content of linalool was much higher than that of geraniol, and the change was quite different from that during brewing. Linalool presented a rising trend, the content after 12 months increased from 1.26% to 2.34%. The content of geraniol was always below 1%, and there was a slight fluctuation, with a decrease from 0.11% to 0.08%. The content of terpenes declined (Fig. 2B), of which β-myrcene decreased the most quickly and to the greatest degree. In addition, β-myrcene decreased faster during the first 3 months, from 6.88% to 2.01%, followed by a further decrease from 2.01% to 0.10% 12 months later. Overall, β-myrcene and decreased by 98.5% compared to the amount in the base beer. The change of β-pinene ranked the second greatest decrease, from 0.43% to 0.08% after 12 months with an overall decrease of 81% compared with the base beer. The other 2 terpenes decreased by approximately 50% after 12 months. The 3 esters were analyzed and the ethyl laurate decreased drastically during storage from 5.51% to 0.19%, for an overall decrease of 96.6% compared with the base beer. However, isoamyl acetate showed an increase from 0.07% to 1.95 %, and the content after 12 months was approximately 28.81 times greater than that in the base beer. Isoamyl butyrate was at trace levels (Fig. 2C). The 2-undecanone content after storage was approximately 40% that in the base beer (Fig. 2D).



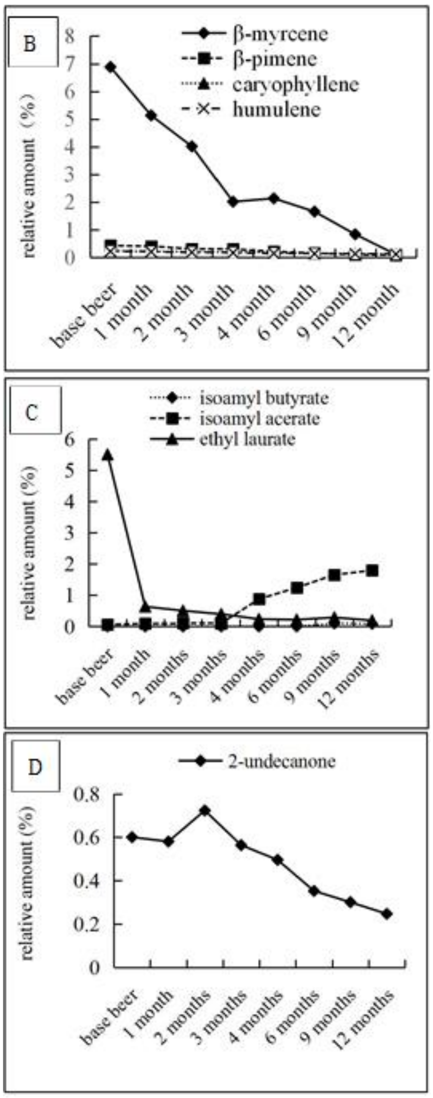


Figure 2. Comparison of aroma compounds (%) during the storage process of hopped beer (wort, ferm 3d, ferm end, dry hopping 1d, and dry hopping 4d); (A) linalool and geraniol; (B) terpenes: β-myrcene, caryophyllene, β-pinene, and humulene; (C) esters: isoamyl butyrate, isoamyl acetate and ethyl laurate (D) 2-undecanone

D. Sensory Evaluation

A panel comprising 12 trained individuals (6 males, 6 females) was asked to evaluate and grade each beer sample. The intensity of beer quality was estimated on a 10-point scale (9-10 = high quality, 8-9 = general quality, and <8 =low quality). According to the results (Table II), it was not difficult to see that the base beer and 1 month beer had the higher scores, all of which were higher than 9. Therefore, according to the sensory analysis, these two groups of beer were considered higher quality. Beers that were stored for 3, 4 and 6 months belonged to the general quality beer, with all scores higher than 8. The 9 and 12 months beers were classified as low-quality beer. The deterioration in quality of the beer was also visually observed (Fig. 3A, B). Comparing Fig. 3A with 3B, it was apparent that the score gap of the beers that were stored for 0-4 months was large, the first 3 months of storage showed the fastest decline in quality. It is therefore better to consume the beer within 3 months of the beer production. In addition, color, foam and transparency changed fastest, as also shown in both Fig. 3A and 3B.

E. Correlation Analysis of Aroma Compounds.

The results of the correlation analysis of the 10 aroma compounds during storage are depicted in Table III. Linalool showed a significant negative correlation with the other 6 aroma compounds (geraniol, β-myrcene, β-pinene, caryophyllene, humulene and 2-undecanone) and showed a significant positive correlation with isoamyl acetate. In addition, geraniol, β-myrcene, caryophyllene, humulene, and isoamyl butyrate showed a positive correlation with the other aroma compounds except isoamyl acetate. β-pinene correlated positively with caryophyllene, humulene, isoamyl butyrate and ethyl laurate. There was no significant correlation recorded between 2-undecanone and ethyl laurate. The results indicated that isoamyl butyrate was positively correlated with other aroma compounds, and isoamyl acetate showed a negative correlation.

TABLE II. THE FINAL SCORE OF EACH GROUP OF BEERS AFTER SENSORY EVALUATION

Quality [a] / Storage time [b] (month)	Appearance			Smell	Taste		
	Color	Foam	Clarity	Aroma	Bitterness	Killing force	Refreshing feeling
0 [c]	9.9	9.8	9.8	9.9	9.8	9.8	9.7
1	9.5	9.6	9.4	9.5	9.5	9.6	9.6
2	9.3	9.2	9.0	8.9	9.2	9.3	9.2
3	9.0	8.9	8.9	8.7	9.0	9.1	9.0
4	8.8	8.6	8.4	8.6	8.8	8.8	8.4
6	8.5	8.3	8.0	8.5	8.5	8.6	8.0
9	8.0	7.9	7.8	8.0	8.4	8.3	7.8
12	7.0	6.8	6.7	7.2	8.0	7.9	7.5

[a]The beer quality determined by 12 panelists. The evaluation basis includes three aspects: appearance (color, foam, and clarity), smell (aroma), and taste (bitterness, killing force, and refreshing feeling). Classification standard (high quality= all scores were higher than 9; general quality = all scores were higher than 8; low quality= any score was less than 8). [b] Storage time was the storage time of the beer in each group, with a month as the unit of measure. [c] 0 indicated stored for 0 months, i.e. base beer.

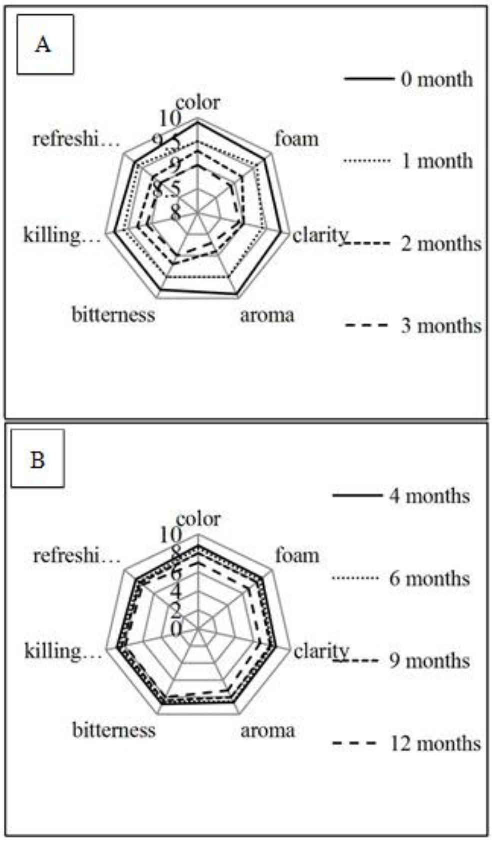


Figure 3. Comparison of the quality features of beer stored for different times (A) The sensory scoring of beers for base beer, 1, 2 and 3 months; (B) The sensory scoring of beers for 4, 6, 9 and 12 months.

F. Correlation Analysis of Aroma Compounds

The results of the correlation analysis of the 10 aroma compounds during storage are depicted in TABLE III. Linalool showed a significant negative correlation with the other 6 aroma compounds (geraniol, β-myrcene, β-pinene, caryophyllene, humulene and 2-undecanone) and showed a significant positive correlation with isoamyl acetate. In addition, geraniol, β-myrcene, caryophyllene, humulene, and isoamyl butyrate showed a positive correlation with the other aroma compounds except isoamyl acetate. β-pinene correlated positively with caryophyllene, humulene, isoamyl butyrate and ethyl laurate. There was no significant correlation recorded between 2-undecanone and ethyl laurate. The results indicated that isoamyl butyrate was positively correlated with other aroma compounds, and isoamyl acetate showed a negative correlation.

G. Correlation Analysis between Aroma Compounds and Beer Aroma

Fig. 4 shows the relationship between the 10 aroma compounds and beer aroma. Among them, linalool and isoamyl acetate showed a negative correlation with beer aroma. Geraniol, caryophyllene, humulene, β-pinene, β-myrcene, isoamyl butyrate and ethyl laurate were positively correlated with beer aroma. However, the R^2 value shows that their relationship is not significant and not linear. The aroma of beer is complex and influenced by many factors, and therefore, there is not a simple linear relationship between aroma compounds.

TABLE III. CORRELATION ANALYSIS BETWEEN THE AROMA COMPOUNDS DURING STORAGE

Variables[a]	Li	Ge	β-m	β-p	Ca	Hu	I-b	I-a	E-l	2-u
Li	1									
Ge	-0.812	1								
β-m	-0.876	0.791	1							
β-p	-0.931	0.811	0.935	1						
Ca	-0.87	0.812	0.957	0.983	1					
Hu	-0.918	0.786	0.961	0.979	0.977	1				
I-b	0.379	0.856	0.734	0.728	0.798	0.294	1			
I-a	0.938	-0.847	-0.821	-0.948	-0.934	-0.934	-0.71	1		
E-l	-0.555	0.404	0.76	0.608	0.667	0.686	0.798	-0.819	1	
2-u	-0.905	0.815	0.796	-0.888	0.897	0.801	0.801	-0.963	0.403	1

[a]Li: linalool; Ge: geraniol; β-m: β-myrcene; β-p: β-pinene; Ca: caryophyllene; Hu: humulene; I-b: isoamyl butyrate; I-a: isoamyl acetate; E-l: ethyl lautate; 2-: 2-undecanone**)**

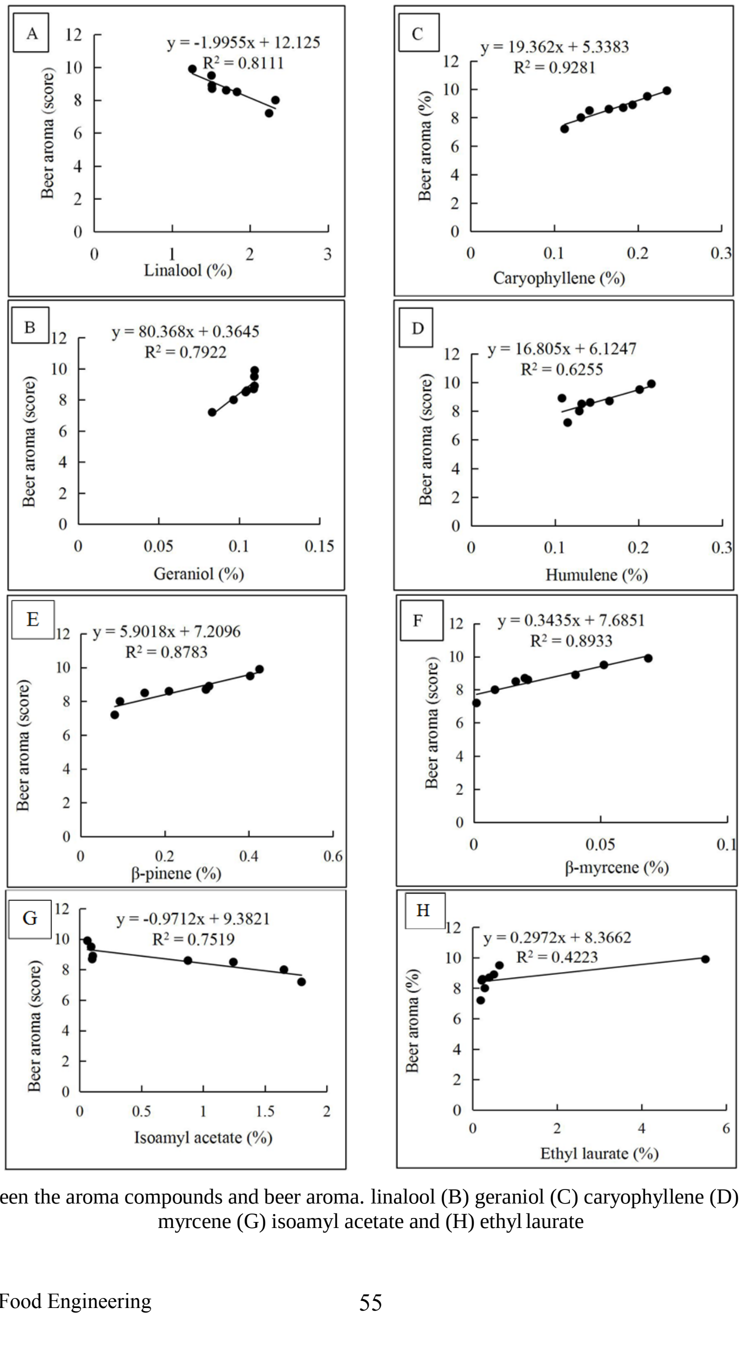


Figure 4. Correlation between the aroma compounds and beer aroma. linalool (B) geraniol (C) caryophyllene (D) humulene (E) β-pinen(F) β-myrcene (G) isoamyl acetate and (H) ethyl laurate

IV. Discussion

The aim of this study was to determine how the aroma compounds of IPA change during the brewing and storage and to further explore the relationship between the aroma compounds and that between aroma compounds and beer aroma. Although we selected only 10 aroma compounds to investigate, they represented the changes in major flavor compounds in hops.

This work provided evidence that the content of aroma compounds are reduced during wort boiling and fermentation. It is worth mentioning that the terpenes β-myrcene, caryophyllene and humulene, declined by more than 90% during the fermentation, whereas linalool and isoamyl acetate declined by approximately 70%. These results indicated that the boiling point of these aroma compounds was low and that they experienced a great deal of volatilization during the boiling and are likely transformed into other compounds during fermentation. This was supported by other studies [20], who examined changes in the aroma substances of 3 hops (HHT, 9702A, 9803A) before and after boiling (Table IV). They found that terpene hydrocarbons and isobutyric esters decreased drastically during the boiling, and terpene hydrocarbons especially remained below 1 μg/g of hops after boiling, which corresponded to less than 1% of their hop content before boiling. Several researchers have also reported that most of the hydrophobic terpene hydrocarbon would remain from hops in the finished beer during beer production and that isobutyric esters could be unstable during boiling and fermentation [11], [21] Although the aroma compounds we studied are not exactly the same compounds, their decreasing trends are the same. Because the boiling time and conditions are different, the proportion of decline is also different. King *et al* [22], [23] described the biotransformation of aroma compounds and the reasons for the reduction in the content of some aroma substances (Fig. 5). Further, we analyzed the changes in the content of aroma compounds during the storage; where the relative amounts of linalool and isoamyl acetate decreased.

TABLE IV. Comparison of the Content of Volatile Compounds (%) During Boiling a (Proposed by Takoi ET AL)

Volatile compd	HHT		9702A		9803A	
	Before boiling	After boiling	Before boiling	After boiling	Before boiling	After boiling
α-humulene	50.9	0.1	5.1	tr	36.1	0.1
β-myrcene	127	0.1	153	0.8	134	0.2
Isobutyl isobutyrate	4.5	0.2	3.8	0.3	2.4	0.1
Isoamyl isobutyrate	1.1	Tr	4.1	0.1	1.3	Tr
2-methylbuty Isobutyrate	12.2	0.2	16.4	0.4	6.6	0.1
Linalool	37.4	6.9	126	42.3	65.6	17.4
α-terpineol	1.3	0.4	4	2.4	3.9	1.7
β-citoronellol	Tr	Tr	Tr	Tr	Tr	Tr
Nerol	0.1	Tr	0.4	0.2	0.5	0.2
Geraniol	0.8	0.2	5.2	2	17.2	4.7

[a] Legend: before boiling, calculated from the measurement of ground hop; after boiling, calculated from the measurement of hop water extract (autoclaved at 105 ℃ for 5 min); and tr, trace.

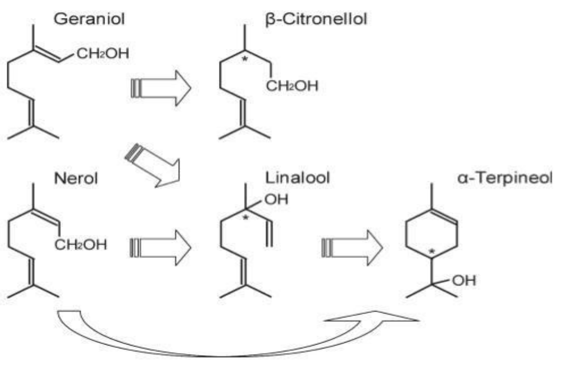


Figure 5. Metabolism cascade of monoterpene alcohols by lager and ale yeast (proposed by King *et al.*) An asterisk indicates a chiral center

In addition to the changes in the content of aroma compounds in hops during brewing and storage, the correlation between aroma compounds and the correlation between aroma compounds and beer quality were studied. Our results indicated that there was a certain correlation between aroma compounds (Table III). In fact, this was also consistent with the research of King and Dickinson (Fig. 5) [23], [24], who proposed this cascade from the results of model fermentation containing each monoterpene alcohol. Geraniol could be transformed into β-citronellol and adjunctively into linalool; nerol could be converted to linalool and R-terpineol; and part of linalool could be cyclized to R-terpineol. In particular, the biotransformation of geraniol to β-citronellol was observed to be rapid and occurred within the first 2-4 days of the model fermentation. A decrease in geraniol and a corresponding increase in β-citronellol occurred drastically during this period [23], [24]. Their research explained in detail the transformation of aroma compounds during the fermentation. In addition, this research not only explained the reasons for the decrease of some compounds and the increase of others but also provided a basis for the correlation between compounds, which provided theoretical support for our research. Praet et al (2012) also proposed the transformation of the hops [22]. They proposed that the flavor-active constituents for hoppy aroma (terpenes, sulfur compounds and oxygenated compounds) originate from hop essential oil and discussed the possible transformations of these compounds during the fermentation.

Finally, we explored the relationship between the aroma of hops and beer. In theory, they should be positively correlated, however our results showed that linalool and isoamyl acetate were negatively correlated with the aroma of beer. Our hypothesis is that as the contents of the other 8 aroma compounds decline, the increase of linalool and isoamyl acetate were insufficient to change the declining trend in quality of the beer aroma. This demonstrated that the contribution of hop aroma compounds to beer aroma was not negligible and certainly warrants further investigation.

V. Conclusions

Currently, the hoppy aroma of beer remains a highly controversial topic that still requires in depth investigation. Although a series of articles have appeared on the biotransformation of terpenes and terpenoids by yeast, few studies have been published that investigate the changes in the content of aroma compounds during brewing and storage of IPA. In this study, we proved that changes occurred in the relative content of several main aroma compounds from hops during the brewing and storage. We also compared the quality of beers stored for different periods to study the influence of storage time on beer quality. In addition, the correlation between aroma and beer quality was analyzed. These findings clarified that most primary aroma substances in beer decreased with longer storage times. In addition, there was a close relationship between the content of the aroma compounds and the quality of the beer. Taken together, these findings may represented a clue to identifying the true nature of the hoppy aroma of beer and further support the relevance of boiling aroma hops and dry hopping to develop the hop aroma in real brewing practice. The potential of this research is to identify a compound that can be combined with the fragrance compounds to slow the release of aromas and result in beer that have a long shelf-life.

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