

# Adsorption and Desorption of Beer and Coffee on a Lipid Membrane as Related to Sensory Bitterness

Hiroataka Kaneda<sup>1,3</sup>, Junji Watari<sup>1</sup>, Masachika Takashio<sup>1</sup> and Yoshio Okahata<sup>2</sup>

## ABSTRACT

**J. Inst. Brew.** 109(1), 27–33, 2003

An objective evaluation method for the bitter characteristics of beverages was developed with a lipid-coated quartz-crystal microbalance connected with a flow injection system. Observation of the adsorption and desorption of beer and coffee on the lipid membrane in the buffer system specifically noted the hydrophobic interactions between the beer and coffee components and the lipid membrane, which could simulate the bitter reception reactions on the tongue. The adsorption or duration of beer or coffee on the lipid membrane in the acetate buffer successfully agreed with the bitter intensity or duration in the sensory evaluation, respectively.

**Key words:** Beer, coffee, lipid membrane sensor, bitterness.

## INTRODUCTION

Bitterness is one of the most important organoleptic characteristic of several beverages such as beer and coffee, and not only the intensity but also the duration affect the bitter quality in a sensory evaluation. Lewis et al.<sup>15</sup> have emphasized that the time-intensity rating of bitterness provides a category scaling and additional information, including the rates of increase and decrease of bitterness, persistence of maximum intensity, changes caused by swallowing, and duration of after-taste. Hughes et al.<sup>9–11</sup> have studied the sensory characteristics of isohumulones using the time-intensity method. However, sensory analysis, as traditionally applied, suffers from an objective, unbiased, and reproducible evaluation, thus necessitating statistical handling of the data.

Previously, we used a lipid-coated quartz-crystal microbalance (QCM)<sup>12</sup> to relate adsorption of beer/beer components on the lipid membrane with sensory perception of bitterness for the same beer/beer components. The adsorption of beers on the lipid membrane in the acetate buffer system showed a significant correlation with their bitter

intensity and duration as judged by a sensory evaluation. However, there is a potential problem in that the correlation between the adsorption value and bitter intensity in the sensory evaluation might mathematically also lead to a correlation between the adsorption value and bitter duration, because the relationship between bitter intensity and duration show good correlation.

The aim of this study was to improve the lipid membrane sensor for reasonable determination of not only bitter intensity but also bitter duration of beverages using a lipid-coated QCM connected with a flow injection system, imaging the behavior of bitter components on the tongue while drinking beverages, and to study the bitter characteristics of beer and coffee.

## MATERIALS AND METHOD

### Beers and coffees

Japanese all-malt beer and European stout beer were commercially purchased. The isohumulone contents by HPLC analyses were 24 mg/L and 41 mg/L, respectively. Beer lacking isohumulones was brewed in our 400 L pilot plant. Wort was produced with malt and adjuncts but without hop addition by a decoction method, fermented at 8°C for about 10 days, and then at 0°C for about 1.5 months.

Canned coffees were provided from Production Development laboratory, Sapporo Beer's Beverage Co., Ltd. They were brewed in the same procedure with commercial canned coffees. Coffees 60-D and 30-D were brewed with deeply roasted beans, and coffees 60-L and 30-L were brewed with lightly roasted beans. *Coffea arabica* from Guatemala was used as the beans samples. Coffees 60-D and 60-L were brewed with a higher ratio of roasted beans (60 g/L, brix: 1.3), and coffees 30-D and 30-L were brewed with a lower ratio of roasted beans (30 g/L, brix: 0.7).

### Reagents

Preparations of isohumulones and tetrahydroisohumulones were obtained from English Hop Products, Ltd. The content of isohumulones or tetrahydroisohumulones by HPLC analyses was 21.6% or 7.1%, respectively.

### Measurement system

The Fragrance Sensor SF-105 was purchased from Sogo Pharmaceutical Co., Ltd. (Tokyo). A quartz-crystal microbalance (QCM, 9 MHz, AT cut) was connected to an oscillator designed to drive the quartz at its resonance fre-

<sup>1</sup> Frontier Laboratories of Value Creation, Sapporo Breweries, Ltd., 10, Okatohme, Yaizu-City, Shizuoka 425-0013 Japan.

<sup>2</sup> Department of Biomolecular Engineering, Tokyo Institute of Technology, 4259 Nagatsuda, Midori-ku, Yokohama 226-8501 Japan.

<sup>3</sup> Corresponding author. E-mail:

Hiroataka.Kaneda@sapporobeer.co.jp

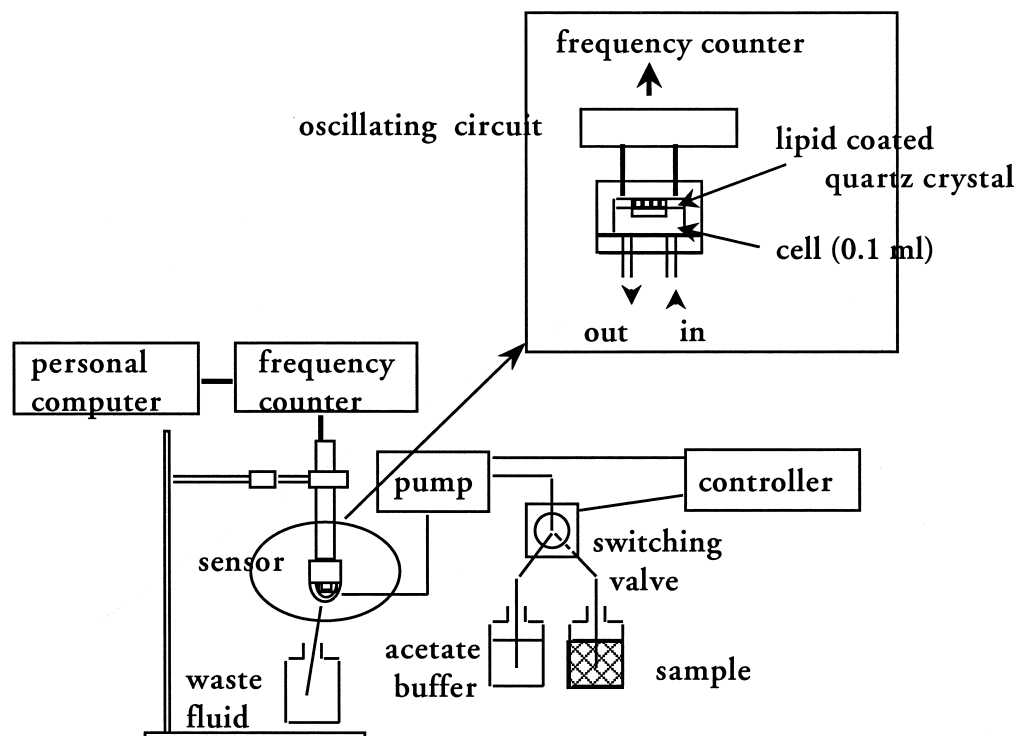


Fig. 1. Measurement system for adsorption and desorption on a lipid membrane by a lipid-coated quartz-crystal microbalance with a flow injection system.

quency in an aqueous solution according to the method employed by Okahata et al.<sup>16,17</sup>. The QCM is driven at 5 V dc, and the frequency of the oscillating quartz is measured by a frequency counter attached to a personal computer system (Think Pad, IBM). When beer or coffee components are adsorbed on the lipid membrane, the frequency of the oscillating quartz is decreased based on the adsorbed weight. Dimyristoylphosphatidylethanolamine was coated on the QCM.

The sensor was connected to a flow injection system (Fig. 1). The system was constructed with a pump (Perista bio-minipump, ATTO Corporation, Tokyo), a switching valve (Gradi Mixer AC-5905, ATTO Corporation, Tokyo), and a controller (Gradicon III AC-5900, ATTO Corporation, Tokyo). The measurement cell was handmade of silicon and the volume was 0.1 mL.

#### Measurement of adsorption of beer or coffee on lipid membrane

The degassed beer or coffee was diluted to 10 or 20% with 75 mM acetate buffer (pH 4.3), and the solution was fully degassed using supersonic equipment. The adsorption and desorption of beer or coffee was measured at room temperature (20°C). The flow time table was as follows: 0–5.0 min, 75 mM acetate buffer (pH 4.3); 5.1–10.0 min, 10 or 20% beer or coffee solution; 10.1–20.0 min, 75 mM acetate buffer (pH 4.3). The flow rate was 2.3–2.8 mL/min. Frequency changes in the QCM were monitored over time for the adsorption of the beer components onto the lipid matrix on the QCM and for their release.

In this study, we defined the adsorption value as the absolute value of the frequency at 4 min after switching to the beer or coffee solution flow from the acetate buffer

flow. The duration value was defined as the absolute value of the frequency at 1 min after switching to the acetate buffer flow from the beer or coffee solution flow.

A few beer or coffee components survived on the lipid membrane even after full rinsing with water and acetate buffer. It was confirmed that the adsorption and duration values were not affected by the residues over 100 measurements.

#### Analysis of isohumulones

The content of isohumulones was measured by HPLC according to the method of Ono et al.<sup>18</sup>

#### Sensory evaluation

It is known that carbon dioxide positively affects the sensory evaluation of bitterness<sup>8</sup>. Therefore, degassed beers were used for the sensory evaluation. The degassed beers and coffees were stored at room temperature (ca. 20°C) before the sensory evaluations.

Sensory evaluations were carried out at room temperature (ca. 20°C) using a slightly modified magnitude estimation of the descriptive analysis<sup>1</sup>. Seven tasters participated in the sensory evaluation. They had been fully trained in our laboratory for over 5 years according to the International Method<sup>1</sup> and had been selected as a panel possessing excellent tasting ability.

Three replicate measurements were performed on different days. Each sample volume was 50 mL. The samples were provided randomly to the tasters. The tasters kept the solution in their mouths for several seconds and then swallowed it. The participants fully rinsed their mouths with crackers before each trial. The interval between each trial was over 2 min.

The bitter intensity and bitter duration of each sample were assigned a number on a scale from 0 (no sense) to 5 (extremely strong). The bitter intensity was evaluated during the placement of the sample in mouth. The bitter duration was evaluated several seconds after swallowing. The value of the bitter intensity or bitter duration of each sample was estimated based on an average of 3 repeated tests.

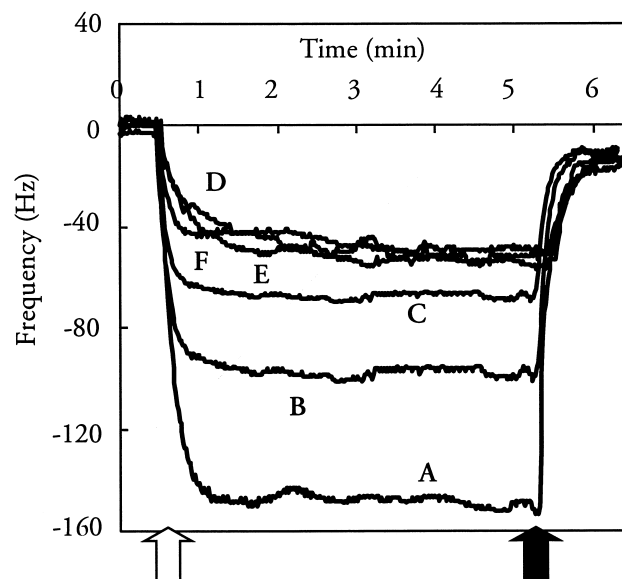
### Statistical analysis

The significance of each value was statistically confirmed with Microsoft Excel 2000 and SPSS 9.0J for Windows. The Friedman's test, which is a statistical analysis for non-parametric comparisons of over 3 samples, was used in this study.

## RESULTS AND DISCUSSION

### Adsorption and desorption of beer on the lipid membrane

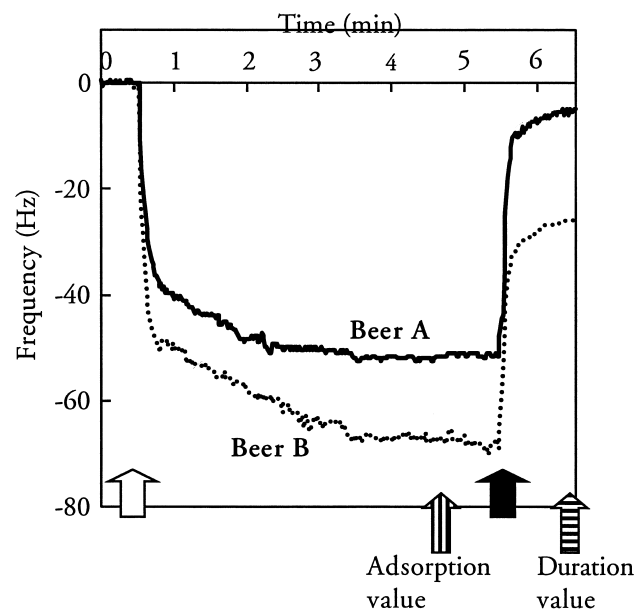
The curve A in Figure 2 shows the frequency change of the lipid-coated QCM in response to the addition of beer solution into a distilled water flow at the time indicated by the open arrow. The frequency of the QCM significantly decreased during several seconds following the beer injection and then gradually decreased. The addition of distilled water, at a time indicated by the closed arrow, increased the frequency. The frequency change increased with increasing beer volume and with increasing temperature



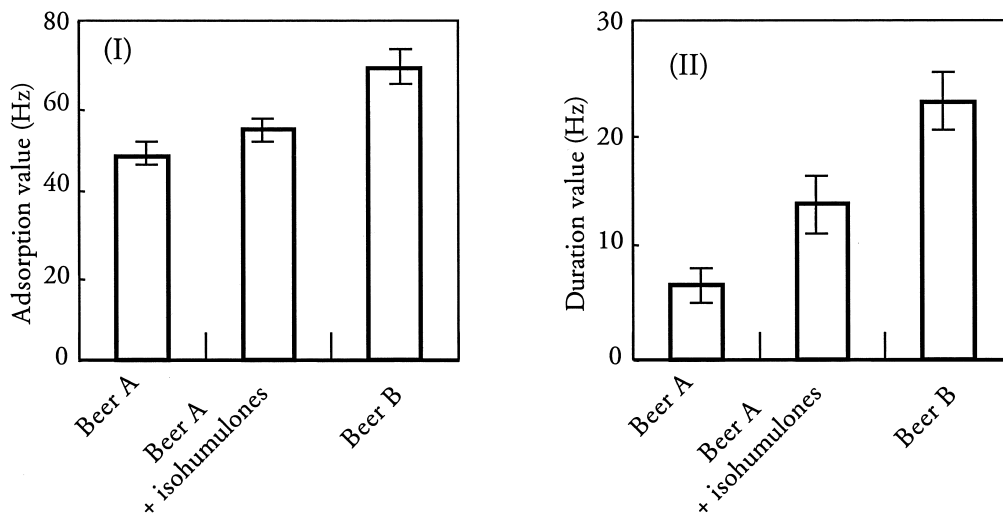
**Fig. 2.** Frequency changes of lipid-coated quartz-crystal microbalance (QCM) responding to the addition of Japanese all-malt beer and effect of buffer concentration on frequency changes of lipid-coated QCM by the addition of the beer. The beer solution (10%) dissolved in distilled water or acetate buffer was injected at the open arrow, and the solution was changed to distilled water or acetate buffer at the closed arrow. A, distilled water; B, 5 mM acetate buffer (pH 4.3); C, 10 mM acetate buffer (pH 4.3); D, 50 mM acetate buffer (pH 4.3); E, 75 mM acetate buffer (pH 4.3); F, 100 mM acetate buffer (pH 4.3).

(data not shown). It is known that the QCM is a very sensitive mass-measuring device at the nanogram level, because the resonance frequency changes sharply upon deposition of a given weight on the electrode<sup>16,17</sup>. The calibration of the QCM has shown that a frequency decrease of 1 Hz corresponds to a mass increase of  $1.05 \pm 0.01$  ng on the electrode of the QCM. The time course of the frequency decrease indicates penetration or diffusion of substances into the lipid matrix. Previously we have shown that the lipid membrane QCM can monitor the adsorption and desorption of organic and inorganic beer components on the lipid membrane based on electrostatic and/or hydrophobic interactions<sup>13</sup>. The adsorption curve can be simulated as the interactions of the beer components with the tongue and throat surface when drinking beer. The desorption curve can be simulated as the duration of the components on the tongue and throat after drinking beer.

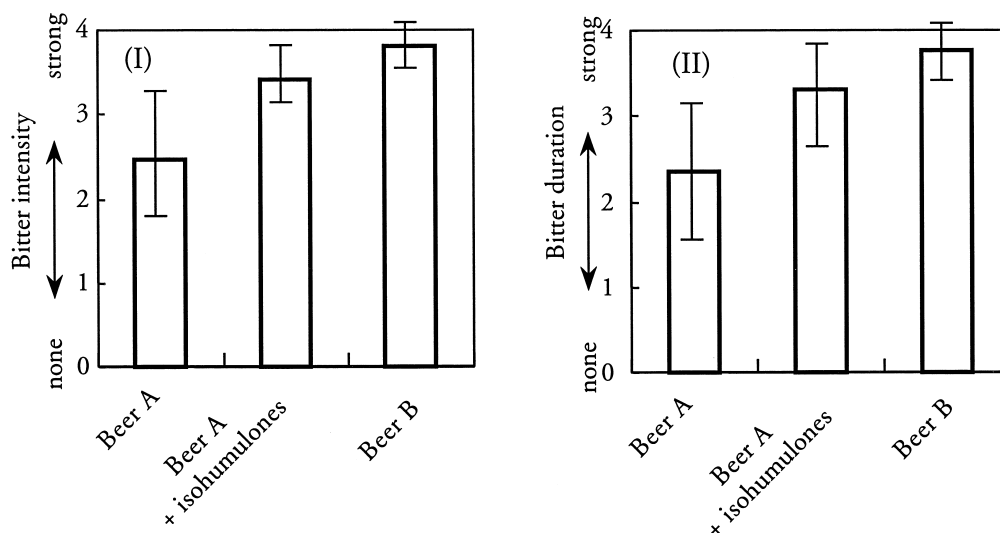
The curves B–F show the adsorption and desorption patterns of the beer components onto the lipid membrane in the acetate buffer (pH 4.3). The frequency change decreased with increasing acetate concentration but maintained a constant level over 50 mM. The same result was observed from 10 to 20% of the beers and coffees used in this study, although the constant level was increased with increasing beer or coffee content. We have previously shown that ionic interactions or electrostatic interactions in the adsorption of beer on the lipid membrane were eliminated in the acetate buffer<sup>12</sup>. It was thought that the ionic interaction between the beer components and lipid membrane was completely eliminated in the over 50 mM acetate buffer and that only a hydrophobic interaction was conspicuous in it. The effects of several organic acids and amino acids, possessing sour and umami tastes, on the adsorption were also masked in the buffer system, although



**Fig. 3.** Frequency changes of lipid-coated quartz-crystal microbalance (QCM) responding to the addition of Japanese all-malt beer (beer A) and European stout beer (beer B). The beer solution (10%) dissolved in 75 mM acetate buffer (pH 4.3) was injected at the open arrow, and the solution was changed to 75 mM acetate buffer (pH 4.3) at the closed arrow.



**Fig. 4.** The adsorption (I) and duration values (II) of Japanese all-malt beer (beer A), the beer A with addition of 17 mg/L isohumulones and European stout beer (beer B). The adsorption and duration values of 10% beer were measured 9 times, and the averages and SD are shown.



**Fig. 5.** Bitter intensity (I) and duration (II) of Japanese all-malt beer (beer A), the beer A with addition of 17 mg/L isohumulones and European stout beer (beer B) in the sensory evaluation. The sensory evaluation by 7 panelists was carried out 3 times, and the averages and SD are shown.

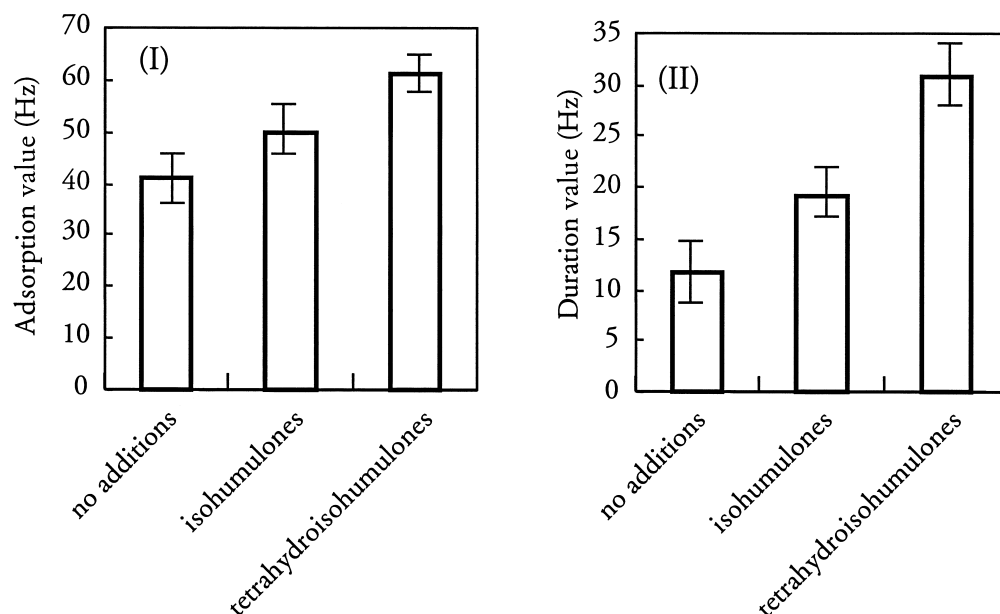
data are not shown. In this study, 75 mM acetate buffer (pH 4.3) was used in the measurement of the adsorption and duration of beers and coffees on the lipid membrane.

#### Measurement of bitterness of beers and coffees

Figure 3 shows the adsorption and desorption patterns of Japanese all-malt beer (beer A) and European stout beer (beer B) on the lipid membrane. Beer B showed higher adsorption and duration on the lipid membrane than beer A. When isohumulones were added to the beer A to the same content as in beer B, the adsorption and duration values increased but did not reach those of beer B (Fig. 4). The sensory evaluation agreed with those results (Fig. 5 and Table I). The addition of isohumulones increased the bitter intensity and duration of beer A but did not reach those of beer B. It seems that bitter components other than

isohumulones can contribute not only to bitter intensity but also to the bitter duration of beer B.

The addition of 30 mg/L of isohumulones or tetrahydroisohumulones to the beer brewed without the addition of hop significantly increased the adsorption and duration values of the beer on the lipid membrane (Fig. 6). Tetrahydroisohumulones increased the adsorption and duration values more compared to the isohumulones. Hughes and Bolshaw<sup>9</sup> have shown with the time-intensity methodology that tetrahydroisohumulones are bitterer than isohumulones and have a more prolonged aftertaste. Guzinski<sup>7</sup> has reported that the bitterness of tetrahydroisohumulones in water is 1.9 times stronger than that of isohumulones. Those reports support our results. Tetrahydroisohumulones are the isohumulone analogues whose double bonds are saturated with hydrogen and have a higher hydrophobicity



**Fig. 6.** Effect of isohumulones or tetrahydroisohumulones on the adsorption (I) and duration values (II) of beer. Thirty milligrams per liter of isohumulones or tetrahydroisohumulones were added to the beer brewed without the addition of hops. The adsorption and duration values of 10% beer were measured 9 times, and the averages and SD are shown.

Table I. Friedman's test for the evaluation values of bitter intensity and duration in Figure 5 (panel number = 21, sample number = 3).

Bitter intensity	Beer A	Beer A + isohumulones	Beer B
	Beer A	—	13.02*
Beer A + isohumulones	—	—	14.49*
Beer B	—	—	—

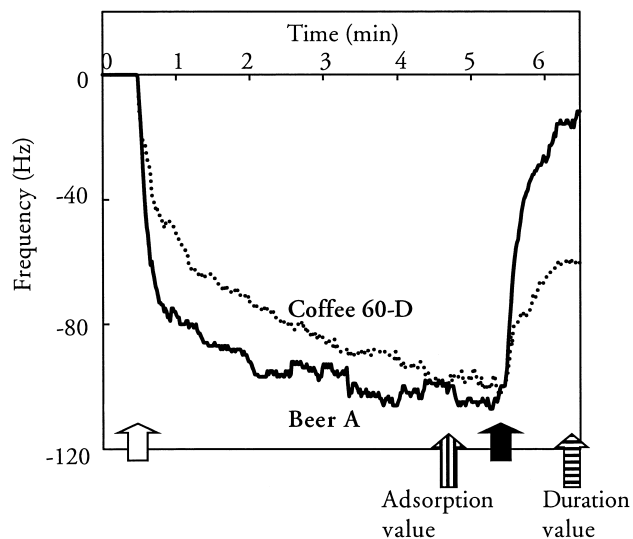
  

Bitter duration	Beer A	Beer A + isohumulones	Beer B
	Beer A	—	16.38*
Beer A + isohumulones	—	—	14.07*
Beer B	—	—	—

\* $p < 0.05$ . \*\* $p < 0.01$ .

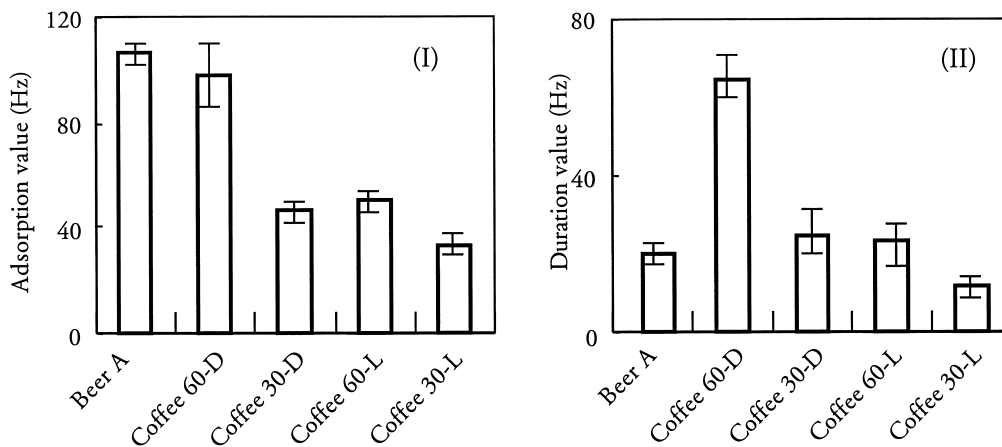
than isohumulones, leading to higher adsorption and duration of beer on the lipid membrane. The results presented so far indicate that the adsorption and duration of beer on the lipid membrane can objectively measure the bitter intensity and duration. However, the question of whether the adsorption or duration value can independently show the bitter intensity or duration has not been solved yet, because the bitter intensity of beer used in this study showed a good correlation with the bitter duration in the sensory evaluation.

Figure 7 shows the adsorption and desorption curve of beer and coffee on the lipid membrane. Beer A more rapidly decreased the frequency of the QCM than coffee 60-D just after the addition, but their frequency decreases reached almost the same level at 4 min after the addition. After switching to the acetate buffer flow from the beer or coffee solution flow, the frequency increase of coffee 60-D was significantly less than that of beer A, indicating the longer duration of coffee 60-D on the lipid membrane than that of beer A.

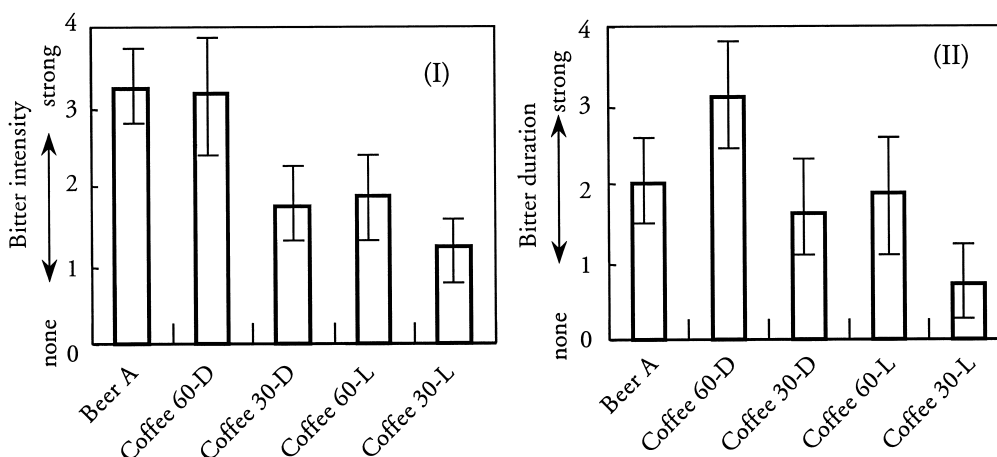


**Fig. 7.** Frequency changes of the lipid-coated quartz-crystal microbalance (QCM) responding to the addition of Japanese all-malt beer (beer A) or coffee 60-D. The beer or coffee solution (20%) dissolved in 75 mM acetate buffer (pH 4.3) was injected at the open arrow, and the solution was changed to 75 mM acetate buffer (pH 4.3) at the closed arrow.

The adsorption values of beer A and coffee 60-D were almost the same level and were higher than those of coffees 30-D, 60-L, and 30-L (Fig. 8(I)). Those of coffees 30-D and 60-L were almost the same level and were higher than that of coffee 30-L. Their bitter intensities in the sensory evaluation successfully coincided with those results (Fig. 9(I), Table II). The bitter intensities of beer A and coffee 60-D were at almost the same level and were stronger than those of coffees 30-D, 60-L, and 30-L.



**Fig. 8.** The adsorption (I) and duration values (II) of Japanese all-malt beer (beer A) and coffees. The adsorption and duration values of 20% beer and coffees were measured 9 times, and the averages and SD are shown.



**Fig. 9.** The bitter intensity (I) and duration (II) of Japanese all-malt beer (beer A) and coffees in the sensory evaluation. The sensory evaluation by 7 panelists was carried out 3 times, and the averages and SD are shown.

Those of coffees 30-D and 60-L were at almost the same level and were stronger than that of coffee 30-L.

The duration value of coffee 60-D was higher than those of beer A and coffees 60-L and 30-L (Fig. 8(II)). Those of beer A and coffees 30-D and 60-L were at almost

Table II. Friedman's test for the evaluation values of bitter intensity and duration in Figure 9 (panel number = 21, sample number = 5).

Bitter intensity	Beer A	Coffee 60-D	Coffee 30-D	Coffee 60-L	Coffee 30-L
Beer A	—	6.93	36.54**	34.02**	57.54**
Coffee 60-D	—	—	43.47**	40.95**	64.47**
Coffee 30-D	—	—	—	2.52	21.00*
Coffee 60-L	—	—	—	—	23.52*
Coffee 30-L	—	—	—	—	—
Bitter duration	Beer A	Coffee 60-D	Coffee 30-D	Coffee 60-L	Coffee 30-L
Beer A	—	22.47*	17.85	8.40	43.47**
Coffee 60-D	—	—	40.32**	30.87**	65.94**
Coffee 30-D	—	—	—	9.45	25.62*
Coffee 60-L	—	—	—	—	35.07**
Coffee 30-L	—	—	—	—	—

\* $p < 0.05$ . \*\* $p < 0.01$ .

the same level and were higher than that of coffee 30-L. Their bitter durations in the sensory evaluation also agreed with those results (Fig. 9(II), Table II). The bitter duration of coffee 60-D was stronger than beer A and coffees 30-D, 60-L, and 30-L. Those of beer A and coffees 30-D and 60-L were at almost the same level. Therefore, it indicated that the adsorption and duration of coffee and beer on the lipid membrane could independently show the bitter intensity and duration in the sensory evaluation, respectively.

Caffeine is the characterized bitter compound in coffee and accounts for 10–30% of the total bitterness<sup>20</sup>. Other green coffee constituents, such as trigonelline, chlorogenic acid, and quinic acid, are not important for the bitterness of roasted coffee<sup>2</sup>. It has been suggested that the bitter compounds in roasted coffee are formed during the roasting process<sup>2</sup>. Recently, diketopiperazines formed during the roasting process have attracted attention as one of the bitter components in coffee and beer. Ginz and Engelhardt<sup>6</sup> have identified five proline-based diketopiperazines in roasted coffee. Gautshi and Schmid<sup>4</sup> have characterized seven proline-based diketopiperazines in several types of beers and showed that Irish stout beer contains higher con-

centrations of them than ale and lager beers. However, they have described that their contribution to either the aroma or taste of beer is questionable. It may be difficult to clarify the role of small amounts of components on taste by only a sensory evaluation.

Kumazawa et al.<sup>14</sup> have confirmed that membrane potential changes in planar lipid bilayers result from responses to bitter substances adsorbed on the hydrophobic region of the membrane. Recently, it has been predominantly supported by gustatory researchers that the transduction of bitter taste occurs via a receptor-second messenger mechanism in which the interaction of a bitter ligand with its receptor results in the generation of an intracellular second messenger, thus leading to neurotransmitter release<sup>5</sup>. In either case, the hydrophobic interaction of bitter substances with taste cells in the tongue must play an important role in the perception of bitter taste, because most of the bitter substances are hydrophobic or have hydrophobic active sites<sup>19</sup>.

Based on the results presented so far, it was concluded that the adsorption and desorption behavior of beverages on the lipid membrane in the buffer system could specifically express the hydrophobic interactions between the organic components and the lipid membrane, which simulate the bitter reception reactions on the tongue, so that they could reasonably measure the sensory bitter characteristics, such as bitter intensity and duration. Cepicka et al.<sup>3</sup> showed that the development of beer bitterness was different based on the types and recommended determining the bitterness 30 seconds after swallowing the beer sample. Work is in progress to confirm whether the adsorption curve on the lipid membrane can show the developing process of bitterness. Our system will be able to monitor the developing and disappearing processes of bitterness when drinking beverages.

#### ACKNOWLEDGEMENTS

The authors thank Kashiwada, S., Ishii, S., Ishida, F., Umemoto, S., and Ohno, M., Brewing Research Laboratories, Sapporo Breweries, Ltd., for providing the beer brewed without hop addition and Inoue, T. and Sekine, S., Production Development Laboratory, Sapporo Beer's Beverage Co., Ltd., for providing the coffees.

#### REFERENCES

1. American Society of Brewing Chemists, Sensory Analysis, In: Methods of Analysis of the American Society of Brewing Chemists, 8th ed., The Society: St. Paul, MN, 1992.
2. Belitz, H. D., Geschmacksaktive Substantzen in Kaffee. Proceedings of the International Scientific Colloquium on Coffee, Hamburg; Association Scientifique Internationale du Café: Bremen, 1976, pp. 243–252.
3. Cepicka, J., Strejcek, F. and Pokorny, J., Development of the sensory bitterness during the tasting of beer. *Monatsschr. Brauwiss.*, 1992, **45**, 329–331.
4. Gautshi, M. and Schmid, J. P., Chemical characterization of di-

- ketopiperazines in beer. *Journal of Agricultural and Food Chemistry*, 1997, **45**, 3183–3189.
5. Gilbertson, T. A., The physiology of vertebrate taste reception. *Current Opinions of Neurobiology*, 1993, **3**, 532–539.
6. Ginz, M. and Engelhardt, U. H., Identification of proline-based diketopiperazines in roasted coffee. *Journal of Agricultural and Food Chemistry*, 2000, **48**, 3528–3532.
7. Guzinski, J. A., Practical considerations of reduced hop extracts. European Brewery Convention, Monograph, E.B.C. Symposium on Hops, Zoeterwoude, 1994, **22**, 105–113.
8. Hashimoto, N., Beer taste. *New Food Industry (Japanese)*, 1997, **39**, 16–22.
9. Hughes, P.S. and Bolshaw, L.H., Time-dependent sensory responses to chemically-modified hop bitter acids. Proceedings of the European Brewing Convention Congress, Brussels, IRL Press: Oxford, 1995, pp. 151–158.
10. Hughes, P.S. and Menneer, I.D., The relationship between sensory data and the composition of beer. Proceedings of the European Brewing Convention Congress, Maastricht, IRL Press: Oxford, 1997, pp. 579–588.
11. Hughes, P.S. and Simpson, W.J., Sensory impact of hop-derived compounds. European Brewery Convention, Monograph, E.B.C. Symposium on Hops, Zoeterwoude, 1994, **22**, 128–140.
12. Kaneda, H., Shinotzuka, K., Kobayakawa, T., Saito, S. and Okahata, Y., Beer adsorption on a lipid membrane as related to sensory evaluation. *Journal of the American Society of Brewing Chemists*, 2001, **59**, 167–171.
13. Kaneda, H., Takashio, M., Shinotzuka, K. and Okahata, Y., Adsorption to or desorption of beer components from a lipid membrane related to sensory evaluation. *Journal of Bioscience and Bioengineering*, 2001, **92**, 221–226.
14. Kumazawa, T., Nomura, T. and Kurihara, K., Liposomes as model for taste cells: receptor sites for bitter substances including N=C=S substances and mechanism of membrane potential changes. *Biochemistry*, 1988, **27**, 1239–1244.
15. Lewis, M.J., Pangborn, R.M. and Yamashita, J.F., Bitterness of beer. A comparison of traditional scaling and time intensity methods. The Institute of Brewing, Australia and New Zealand Section, Proceedings of the 16th Convention, Sydney, 1980, pp. 165–171.
16. Okahata, Y., A lipid-coated quartz crystal microbalance as an olfaction sensor. In: Olfaction and Taste XI, K. Kurihara, N. Suzuki, and H. Ogawa Eds., Springer-Verlag: Tokyo, 1994, pp. 703–707.
17. Okahata, Y., En-na, G. and Ebato, H., Synthetic chemoreceptive membrane. Sensing bitter or odorous substances on a synthetic lipid multibilayer film by using quartz-crystal microbalances and electric responses. *Analytical Chemistry*, 1990, **62**, 1431–1438.
18. Ono, M., Kakudo, Y., Yamamoto, Y., Nagami, K. and Kumada, J., Quantitative analysis of hop bittering components and its application to hop evaluation. *Journal of the American Society of Brewing Chemists*, 1984, **42**, 167–172.
19. Shinoda, I., Fushimi, A., Koto, H., Okai, H. and Fukui, S., Bitter taste of synthetic C-terminal tetradecapeptide of bovin  $\beta$ -casein, H-Pro196-Val-Leu-Gly-Pro-Val-Arg-Gly-Phe-Pro-Ile-Ile-Val209OF, and its related peptides. *Agricultural and Biological Chemistry*, 1985, **49**, 2587–2596.
20. Voilley, A., Sauvageot, F. and Durand, D., Influence, sur l'amer-tume d'un café-boisson, de quelques paramètres d'extrac-tion. Proceedings of the 8th International Scientific Colloquium on Coffee, Abidjan, 1977, Association Scientifique Internationale du Café: Paris, 1979, pp. 251–259.

(Manuscript accepted for publication January 2003)