Bitterness Suppression of the Kampo Medicine 'Orengedokuto' Using Flavoured Jellies

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The purpose of this study was to evaluate the effect of various flavoured jellies on the palatability and bitterness of the Kampo medicine 'Orengedokuto', using human gustatory sensation testing. In the gustatory sensation test, eight items were evaluated according to the semantic differential (SD) method and four taste intensities (sweetness, astringency, sourness, and bitterness) were determined. Factor analysis of the results identified two main factors 'Disagreeable taste' and 'Agreeable taste and odour' as predominantly determining the palatability of Orengedokuto. To investigate the influence of jelly odour, evoked images were obtained for five fundamental tastes after smelling jellies. The inhibitory effect of the jelly odour on the bitterness of Orengedokuto was found to be small. When the influence of the various jellies on the disagreeable taste of Orengedokuto was investigated, the bitterness intensities of Orengedokuto mixed with chocolate or strawberry–chocolate jelly were found to be significantly lower than the bitterness of the control. The bitterness intensity of Orengedokuto was found to be significantly and negatively correlated with sweetness intensity using simple linear regression analysis. These results suggest that the sweetness intensity of various jellies inhibits the bitterness intensity of Orengedokuto.

Key words bitterness; jelly; Orengedokuto; sweetness; palatability

Orengedokuto is a Kampo medicine (Japanese herbal medicine) that has long been used for the treatment of facial suffusion (a burning sensation), headache, excitation, anxiety, and irritability.[1] In clinical practice, Orengedokuto is currently used for the treatment of palpitations, excitement, hot flushes,[2] facial suffusion, and neurosis.

Orengedokuto has been available on the Japanese market for a long time. Due to several of its active constituents, it has a complex unpleasant taste (predominantly bitter or astringent), which frequently causes noncompliance. Orengedokuto therefore needs to be taste-masked in order to improve compliance.

Recently, a number of jelly products have been developed that aim to improve compliance by aiding swallowing.[3—6] When drugs are incorporated into the jelly products, swallowing is made easier due to its moderate adhesion and liquidity properties, while drug adsorption by the tongue is prevented, and bitterness perception thereby decreased. Fukui has reported on the physicochemical characteristics of jellies, such as viscosity, strength, loss of water content, and effects on swallowing,[7] while Tsuji has reported bitterness suppression of Macrolide dry syrups by jellies.[8] There are, however, few reports in which the effect of jellies on the palatability of Kampo medicines have been evaluated.

In this study, we evaluated palatability improvement of Orengedokuto by the addition of jellies, using human gustatory sensation testing. Palatability scores were evaluated by the semantic differential (SD) method. A factor analysis (rotated with the varimax method) was performed on the data, and 'Disagreeable taste' and 'Agreeable taste and odour' were identified as the two main factors determining palatability. Secondly, we investigated the effects of the odour of the jellies on the palatability of Orengedokuto. Finally, we investigated the effects of the sweetness and sourness intensities of the jellies on suppression of the bitterness and astringency of Orengedokuto.

Experimental

Materials Orengedokuto extract granules were purchased from Tsumura Co., Ltd., Tokyo, Japan. The following jellies were used in this study: Okusurinometane (Chocolate, Strawberry, Peach, and Grape), Kanpoyaku fukuyou (Strawberry–chocolate and Coffee), and Engehojyo (Lemon). All jellies were purchased from Ryukakusan Co., Ltd., Tokyo, Japan.

Sample Preparation For human gustatory sensation test, 20 ml of the jelly (or water as a control) was uniformly mixed with 1 g of Orengedokuto for 30 s using a spoon.

Gustatory Sensation Tests Samples of Orengedokuto mixed with water (control) or jellies (chocolate, strawberry, peach, grape, strawberry–chocolate, coffee, or lemon) were used for gustatory sensation testing in six well-trained volunteers. Each volunteer provided informed consent for the procedures, which were approved by ethical committees of the Mukogawa Women's University.

The sample size was 2 ml, and all samples were kept in the mouth for 10 s. After testing, subjects gargled well before tasting the next sample. Various palatability scores were evaluated using the semantic differential (SD) method as follows[9]: the subjects were asked to score the samples on the basis of eight items, expressed as symmetrical terms representing both extremities, as follows: 1) Bad/Good odour (orthonasal), 2) Weak/Strong bitterness, 3) Weak/Strong astringency, 4) Weak/Strong sweetness, 5) Bad/Good odour (retronasal), 6) Weak/Strong bitterness (aftertaste), 7) Weak/Strong astringency (aftertaste), 8) Weak/Strong sweetness (aftertaste). Each item was scored using the following rating scale: 1, extremely; 2, slightly; 3, neither; 4, slightly; 5, extremely.

In the evaluation of four tastes (sweetness, sourness, bitterness and astringency), the gustatory sensation test was performed according to the method of Katsuragi *et al.*[10] using sucrose concentrations of 29.2, 87.7, 187.7, 409.4 and 994.2 mM as a standard for sweetness, tartaric acid concentrations of 0.17, 0.60, 1.73, 4.66 and 11.99 mM as a standard for sourness, quinine sulfate concentrations of 0.003, 0.012, 0.031, 0.078 and 0.201 mM as a standard for bitterness, and tannic acid concentrations of 0.003, 0.007, 0.020, 0.055 and 0.150% as a standard for astringency. Scores of 1, 2, 3, 4, 5 were allocated to increasing concentrations of all the standard solutions.

Evaluation of Intensity of Five Fundamental Tastes Evoked by the Odour of Jellies The subjects evaluated the smell of 2 ml of each jelly on a 5-point rating scale, from no sensation (1) to very strong sensation (5) for five fundamental tastes (sweetness, sourness, bitterness, astringency and saltiness).

Data Analysis S-PLUS 2000J (Mathematical Systems, Inc., Tokyo, Japan) was used for factor analysis, regression analysis, and principal component analysis. The Dunnett test was used for multiple comparisons. The 5% level of probability was considered significant.

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Results and Discussion

The Palatability of Orengedokuto with Jelly Evaluated by the SD Method Figure 1 shows the palatability scores for Orengedokuto with flavoured jellies compared with control, evaluated by the SD method. In item 1) Bad/Good odour (orthonasal), the scores of Orengedokuto with strawberry, grape, chocolate or strawberry–chocolate jelly especially were higher than control. In item 2) Weak/Strong bitterness, the scores were lower than control especially with chocolate and grape jellies. In item 3) Weak/Strong astringency, the scores were lower than control, especially with strawberry–chocolate jelly. In item 4) Weak/Strong sweetness, the scores were lower than control, especially with strawberry–chocolate, chocolate, strawberry and grape jellies.

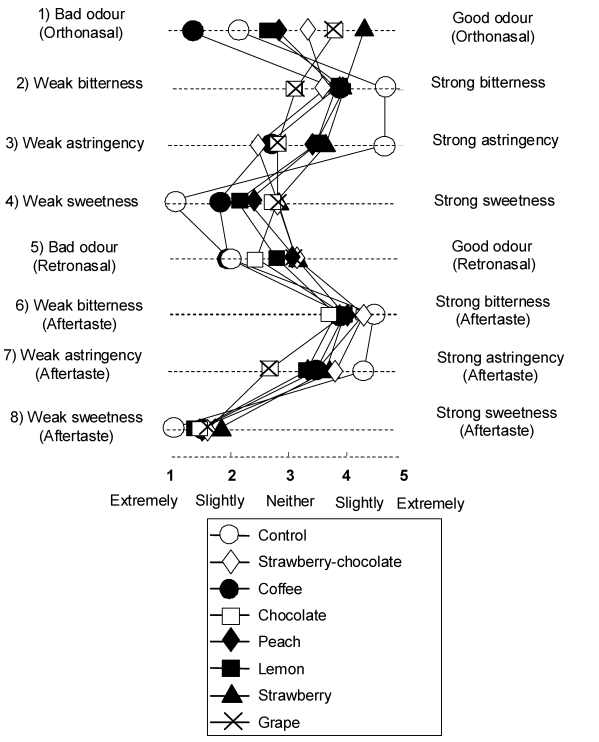

Fig. 1. Palatability Scores of Orengedokuto with Various Jellies Using the SD Method

The data represent the mean of six values.

A factor analysis (rotated using the varimax method) was performed on the data obtained by the SD method. As a result, two factors (factor I and factor II) with values greater than 1.0 were identified. The contributions of these factors I and II were 35.7% and 14.0%, respectively. The factor loading scales are shown in Table 1. Of the eight palatability items, items 2) Weak/Strong bitterness, 3) Weak/Strong astringency, 6) Weak/Strong bitterness (aftertaste), and 7) Weak/Strong astringency (aftertaste) showed high factor loadings of factor I. 'Disagreeable taste' was adopted as the composite factor for these items. Consequently, factor I was interpreted 'Disagreeable taste'. Items 1) Bad/Good odour (orthonasal), 4) Weak/Strong sweetness, 5) Bad/Good odour (retronasal), and 8) Weak/Strong sweetness (aftertaste) showed high factor loadings of factor II. 'Agreeable taste and odour' was adopted as the composite factor for these items. Consequently, factor II was interpreted 'Agreeable taste and odour'.

Principal Component Analysis of Palatability of Orengedokuto with Jellies Determined by Gustatory Sensation Tests As shown in Fig. 2, the palatability of Orengedokuto with any of the jellies was significantly improved compared with control when analysed on the basis of the two factors determined by the principal component analysis. Palatabilities of Orengedokuto with jellies or water (control) were divided into two groups (control and group with any

Table 1. Factor Loading I and II (Factor Loading I: Factor Loading for Factor I, Factor Loading II: Factor Loading for Factor II, Respectively) Resulting from Factor Analysis Using the SD Method for Palatability of Orengedokuto with Jelly ($n=6$)

	Scales	Factor loading	
		I	II
Disagreeable taste	2) Bitterness	0.65891	0.05098
	3) Astringency	0.87464	0.14757
	6) Bitterness (Aftertaste)	0.39337	−0.36930
	7) Astringency (Aftertaste)	0.64497	−0.06115
Agreeable taste and odour	1) Odor (Orthonasal)	−0.28590	0.43640
	4) Sweetness	−0.21402	0.72060
	5) Odour (Retronasal)	−0.41984	0.28967
	8) Sweetness (Aftertaste)	0.34840	0.85432

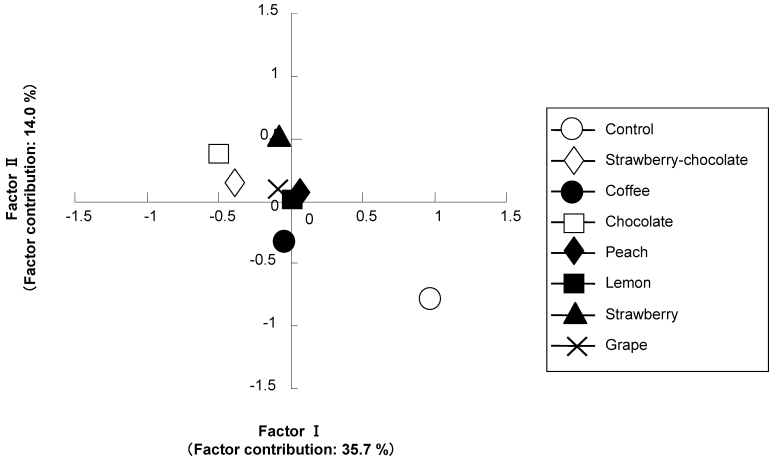

Fig. 2. Principal Component Analysis Result of the Palatability of Orengedokuto with Jellies

Each value is mean±S.E.M. of six subjects.

jellies). The addition of the jellies decreased factor I (disagreeable taste) considerably while increasing factor II (agreeable taste and odour).

Bitterness and Astringency of Orengedokuto Mixed with Jellies as Shown in Gustatory Sensation Tests To further investigate the influence of jellies on the disagreeable taste of Orengedokuto, the bitterness and astringency intensities of Orengedokuto with jellies or control were determined by gustatory sensation tests. As shown in Fig. 3A, mixing with jelly was useful for bitterness suppression of Orengedokuto. The bitterness intensities of Orengedokuto with chocolate or strawberry–chocolate jelly were significantly higher than that of control ($p<0.01$). As shown in Fig. 3B, mixing with jelly was also useful for suppression of astringency. The astringency intensity of Orengedokuto with any jelly was significantly lower than the astringency intensity of control ($p<0.01$).

Influence of Jelly Odour on Disagreeable Taste of Orengedokuto To investigate the influence of the odour of the jellies on the disagreeable taste of Orengedokuto, the evoked image for the five fundamental tastes was determined after smelling the jellies. It has been reported that a sweet aroma can suppress the bitterness intensity of branched-chain amino acids.[11] As shown in Fig. 4, odour evoked various kinds of fundamental tastes in all samples. The odour of strawberry–chocolate and chocolate jelly in particular evoked sweetness, more than any other taste. However, there were no statistically significant differences between the sweetness intensities of the jellies, and any inhibitory effect of jelly odour on the bitterness of Orengedokuto was therefore thought to be small.

The Sourness and Sweetness Intensity of Orengedokuto with Jellies as Determined by Gustatory Sensation Testing To investigate the influence of the various jellies on suppression of the bitterness and astringency of Orengedokuto, the sourness and sweetness intensities of the various mixtures were determined in gustatory sensation tests. As shown in Fig. 5A, the sweetness intensities of chocolate and strawberry–chocolate jelly were significantly higher than that of control ($p<0.01$). As shown in Fig. 5B, the sourness inten-

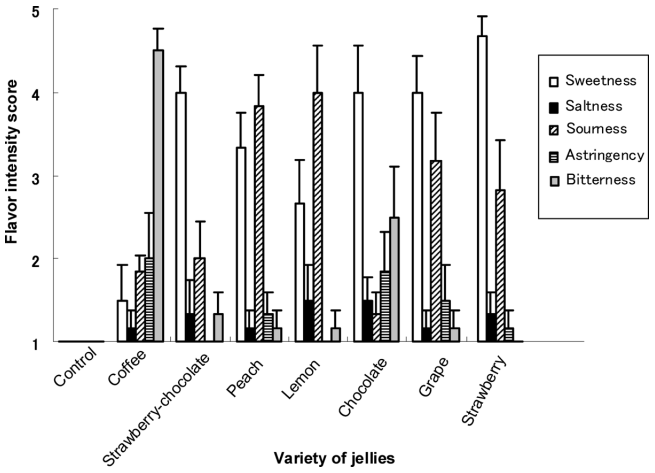

Fig. 4. The Odour-Evoked Image for Five Fundamental Tastes after Smelling Jellies

Each value is mean±S.E.M. of six subjects.

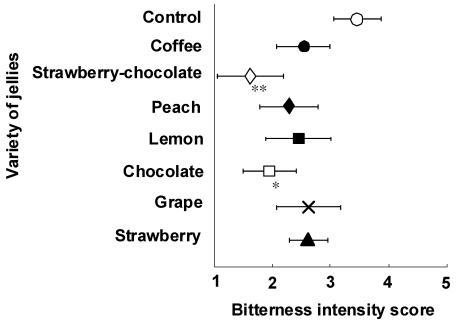

Fig. 3A. The Sweetness Intensity Scores of Various Jellies from Gustatory Sensation Tests

Each value is mean±S.E.M. of six subjects. $*p<0.05$; $**p<0.01$; compared with control using the Dunnet test.

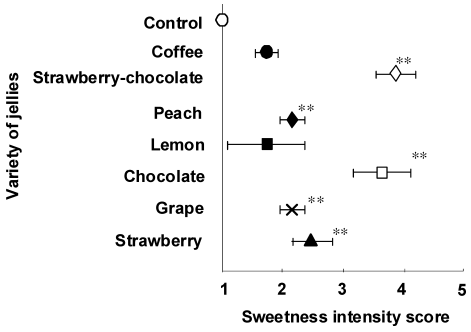

Fig. 5A. The Bitterness Intensity Scores of Various Jellies from Gustatory Sensation Tests

Each value is mean±S.E.M. of six subjects. $**p<0.01$; compared with control using the Dunnet test.

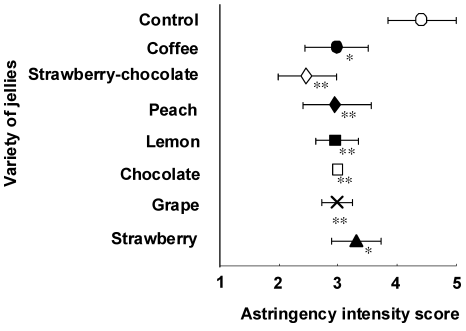

Fig. 3B. The Sourness Intensity Scores of Various Jellies from Gustatory Sensation Tests

Each value is mean±S.E.M. of six subjects. $*p<0.05$; $**p<0.01$; compared with control using the Dunnet test.

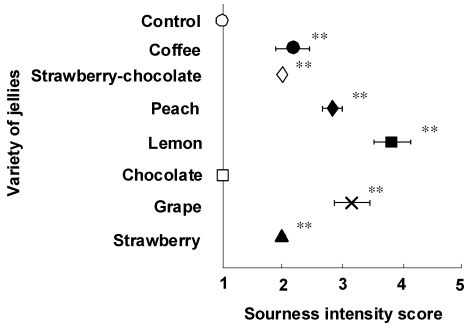

Fig. 5B. The Astringency Intensity Scores of Various Jellies from Gustatory Sensation Tests

Each value is mean±S.E.M. of six subjects. $**p<0.01$; compared with control using the Dunnet test.

sities of all the jelly mixtures were significantly higher than that of control ($p<0.01$).

Correlation between the Bitterness Intensity of Orengedokuto and the Sweetness Intensity of the Jellies To investigate the correlation between the bitterness and astringency intensities of Orengedokuto and the sweetness or sourness intensity of the jellies, a simple linear regression analysis was conducted, as shown in Figs. 6A—D. The regression equation $Y=-0.5479X+3.8825$ ($n=6$, $r=0.97158$, $r^2=0.94433$, $F=0.00006$, S.D.$=0.14138$, $p<0.001$) was obtained for the correlation between the bitterness and sweetness intensities. The bitterness intensity was significantly negatively correlated with sweetness intensity. The regression equation $Y=-0.060X+2.6350$ ($n=6$, $r=0.07494$, $r^2=0.00553$, $F=0.86101$, S.D.$=0.59755$) was obtained for the correlation between the bitterness and sourness intensities. There was no significant correlation between bitterness and sourness intensities. The regression equation $Y=-0.4139X+4.1533$ ($n=6$, $r=0.70269$, $r^2=0.49258$, $F=0.44652$, S.D.$=0.05233$) was obtained for the correlation between astringency and sweetness intensities. There was no significant correlation between astringency and sweetness intensities. The regression equation $Y=-0.2660X+3.786$ ($n=6$, $r=0.45678$, $r^2=0.20841$, $F=0.25552$, S.D.$=0.55771$) was obtained for the correlation between astringency and sourness intensities. There was no significant correlation between astringency and sourness intensities. These results suggest that it is the sweetness intensity of the jellies which suppresses the bitterness of Orengedokuto.

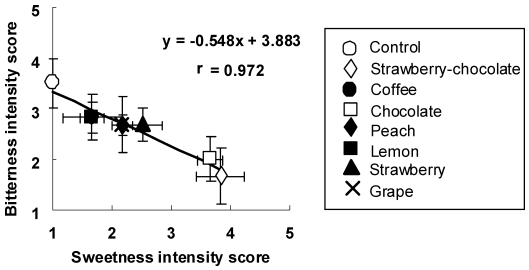

Fig. 6A. The Correlation between Bitterness and Sweetness Intensities Using a Simple Linear Regression Analysis

Each value is mean±S.E.M. of six subjects.

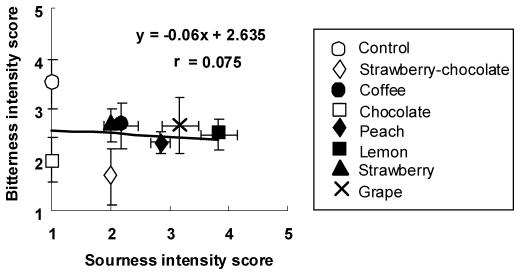

Fig. 6B. The Correlation between Bitterness and Sourness Intensities Using a Simple Linear Regression Analysis

Each value is mean±S.E.M. of six subjects.

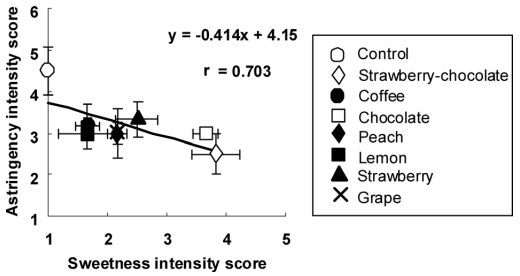

Fig. 6C. The Correlation between Astringency and Sweetness Intensities Using a Simple Linear Regression Analysis

Each value is mean±S.E.M. of six subjects.

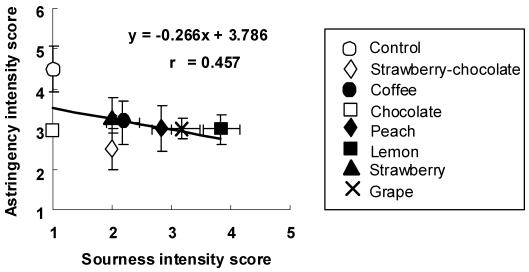

Fig. 6D. The Correlation between Astringency and Sourness Intensities Using a Simple Linear Regression Analysis

Each value is mean±S.E.M. of six subjects.

Keast and Breslin have reviewed the interaction between bitterness and sweetness.[12] Mixtures of bitter and sweet tastes were variably affected at low intensities/concentrations, while mixtures at moderate and high intensities/concentrations were mutually suppressive. In this study, the bitterness of Orengedokuto had a moderate intensity (control $\tau=3.5$), and the jellies with moderate to high sweetness intensities (strawberry–chocolate $\tau=3.8$, chocolate $\tau=3.7$, respectively) most strongly masked the bitterness of Orengedokuto. This result is in agreement with the above review.

It has well known that flavored powder of chocolate is shown possibly useful for suppression of bitter taste of drugs.[13,14] Flavor of chocolate jelly may have some effects on inhibition of bitterness of Orengedokuto. However, from the results and statistics in this study, high sweetness intensities of strawberry–chocolate jelly and chocolate jelly (strawberry–chocolate $\tau=3.8$, chocolate $\tau=3.7$, respectively) are suggested to be more potent factor of inhibition of bitterness of Orengedokuto than flavor of chocolate.

There are few papers on masking the bitterness of Kampo medicines. Kampo medicines have many constituents, making it difficult to determine the main cause of bitterness. One of the bitter constituents of Orengedokuto is berberine. The sweetness intensity of the flavoured jellies may therefore suppress the bitterness of Kampo medicines containing berberine.

Conclusion

In the present study, the effects of various flavoured jellies on the palatability and bitterness of the Kampo medicine 'Orengedokuto' were evaluated using human gustatory sensation testing. 'Agreeable taste' and 'odour' were the factors which were considered to determine the overall palatability of 'Orengedokuto' using the semantic differential (SD) method and factor analysis. However, the inhibitory effect of jelly odour on the bitterness of Orengedokuto was found to be small. The bitterness of Orengedokuto was significantly negatively correlated with the sweetness intensity of the jellies in a simple linear regression analysis. These results suggested that jellies with a high sweetness intensity can inhibit the high bitterness intensity of Orengedokuto.

References

1) Otsuka K., Yakazu D., Shimizu T., "Standard Practice of Kampo Medicine," Nanzando, Tokyo, 1986, p. 86.
2) Wakita H., Suzuki N., Miyamoto K., *J. Traditional Med.*, **19**, 230—237 (2002).
3) Hanawa T., Watanabe A., Tsuchiya T., Ikoma R., Hidaka M., Sugihara M., *Chem. Pharm. Bull.*, **43**, 872—876 (1995).
4) Kaneuchi M., Kohri N., Senbongi K., Sakurai H., Iseki K., *Yakugaku Zasshi*, **125**, 187—196 (2005).
5) Ando M., Kouzai T., Higuchi F., Ichihashi T., Takeda M., Mizutani Y., Uesaka H., *Yakuzaigaku*, **60**, 261—270 (2000).
6) Fukui A., Fujii R., Yonezawa Y., Sunada H., *Chem. Pharm. Bull.*, **52**, 298—302 (2004).
7) Fukui A., Fujii R., Sunada H., *Yakuzaigaku*, **60**, 62—70 (2000).
8) Tsuji E., Uchida T., Fukui A., Fujii R., Sunada H., *Chem. Pharm. Bull.*, **54**, 310—314 (2006).
9) Osgood E. C., Suci G. J., Tannenbaum P. H., "The Measurement of Meaning," University of Illinois Press, Urbana, 1957.
10) Katsuragi Y., Mitsui Y., Tanaka H., Wada K., Kurosaki S., Ohki T., Yoshida M., Matsuyama K., *Pharm. Res.*, **14**, 720—724 (1997).
11) Mukai J., Tokuyama E., Ishizaka T., Okada S., Uchida T., *Chem. Pharm. Bull.*, **55**, 1581—1584 (2007).
12) Keast R. S., Breslin P. A. S., *Food Qual. Pref.*, **14**, 111—124 (2002).
13) Namiki N., Sasaki T., Takano H., Sakurai S., Yamamoto T., *Yakuzaigaku*, **64**, 308—315 (2004).
14) Namiki N., Sasaki T., *PHARM TECH JAPAN*, **20**, 2267—2274 (2004).