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Application News

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Liquid Chromatography Mass Spectrometry

Multi-Component Analysis of Five Beers

Sensory test methods that present flavor and taste in numerical terms are widely used techniques for the quantitative evaluation of food quality. Recently, attempts have been made to obtain a greater amount of data by combining the results of sensory tests with metabolomics data that provides a comprehensive analysis of food constituents. Combining comprehensive analytical data with use of multivariate analysis has become an effective option for elucidating the differences in a wide range of constituents. This combination of multivariate analysis and metabolomics promises to see application in areas of quality evaluation and quality improvement, as well as in the development of foods with improved functional properties. In this article, we describe the simultaneous analysis of five commercially available beers with a triple quadrupole mass spectrometer (LCMS-8060) using a primary metabolite method package, and performing principal component analysis on the data set obtained using Traverse™ MS software. We classified the five beers based on differences in constituent compounds, and confirmed characteristic components in each beer. We then performed hierarchical clustering analysis to show how these five beers are classified.

Table 1 Analytical Conditions

LC Analytical Conditions

Column	: RP column
Mobile Phase A	: 0.1 % Formic acid - Water
Mobile Phase B	: 0.1 % Formic acid - Acetonitrile
Flowrate	: 0.25 mL/min
Mode	: Gradient elution

MS Analytical Conditions

Ionization Method	: ESI(+)/(-)
Nebulizing Gas Flow	: 2 L/min
Heating Gas Flow	: 10 L/min
Drying Gas Flow	: 10 L/min
Probe Voltage	: 4 kV(+)/-3 kV(-)
Interface Temperature	: 300 ˚C
DL Temperature	: 250 ˚C
Block Heater Temperature	: 400 ˚C

Beer Sample Preparation and Analysis

An internal standard was added to 0.2 mL of degassed beer, and ultrafiltration was performed on this mixture using a molecular weight cut off filter. The filtrate was recovered, diluted 200 times, and then used for simultaneous analysis by LC/MS. LC/MS analysis was performed according to analytical conditions in the primary metabolite method package. The analytical conditions used are shown in Table 1. The MRM chromatograms for three of the five beers (lager beer A, *happoshu* [low-malt beer], and non-alcoholic beer) are shown in Fig. 1. These results show the types of various constituents, such as amino acids, found in the five beers differ greatly between the beers.

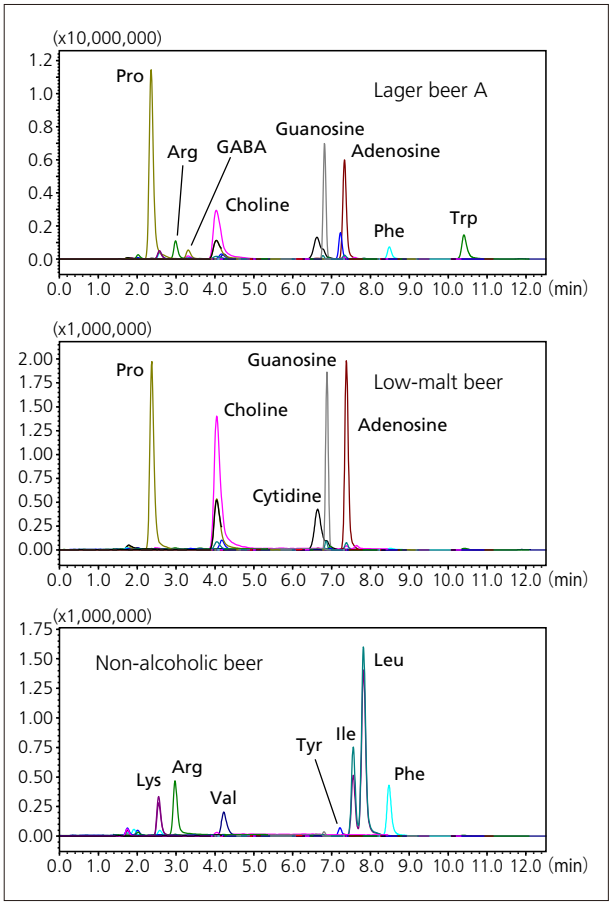

Fig. 1 MRM Chromatograms of Three Beers

Multivariate Analysis with Traverse™ MS Software

Using the area ratio of each constituent (58 constituents) compared against the internal standard, a principal component analysis (PCA) was performed for the five beers using the Traverse™ MS software. PCA results showing different quantities of constituents in each beer are presented in Fig. 2. Score plot and loading plot results are also shown in Fig. 2. The five beers can be seen to appear fully separated on the score plot. From the loading plot, we also determined constituents that were characteristic to each beer. We confirmed that a number of amino acids and nucleosides differed markedly between the beers.

Next, the area ratio of detected constituents was used to perform hierarchical clustering analysis of the five beers using the Traverse™ MS software. The results of this analysis are shown in Fig. 3. Autoscaling was used to normalize area ratios between samples. As shown in Fig. 3, the lager beers and ale beer are grouped relatively close to each other, and the low-malt beer and non-alcoholic beer are also grouped close to each other. Hierarchical clustering analysis provides a visual representation of the degree of similarity between the beers in terms of their constituents. Performing a comprehensive analysis of food constituents in this way and combining it with multivariate analysis makes it easy to evaluate which constituents affect food quality and function. This combination of comprehensive simultaneous analysis and multivariate analysis is expected to become more commonly used for quality evaluation of food in the future.

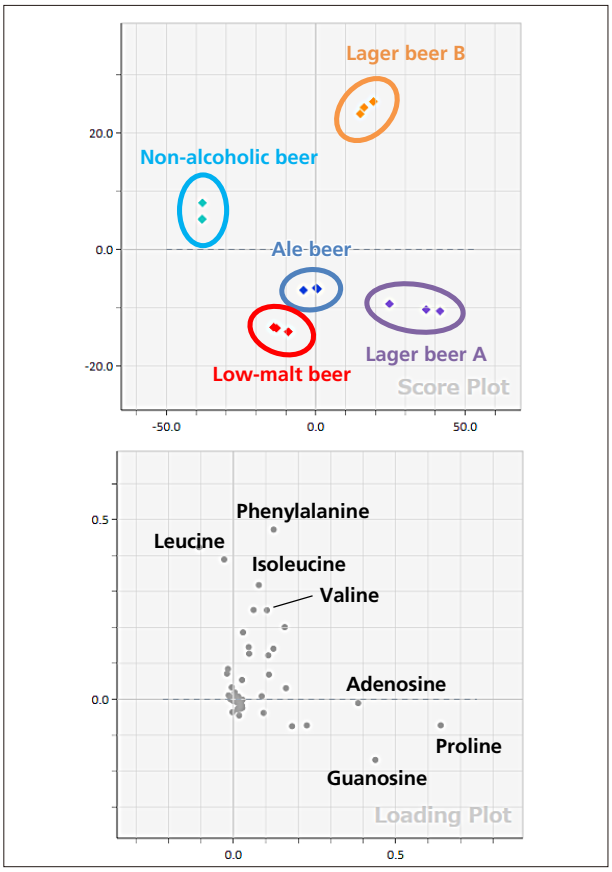

Fig. 2 Principal Component Analysis of Five Beers

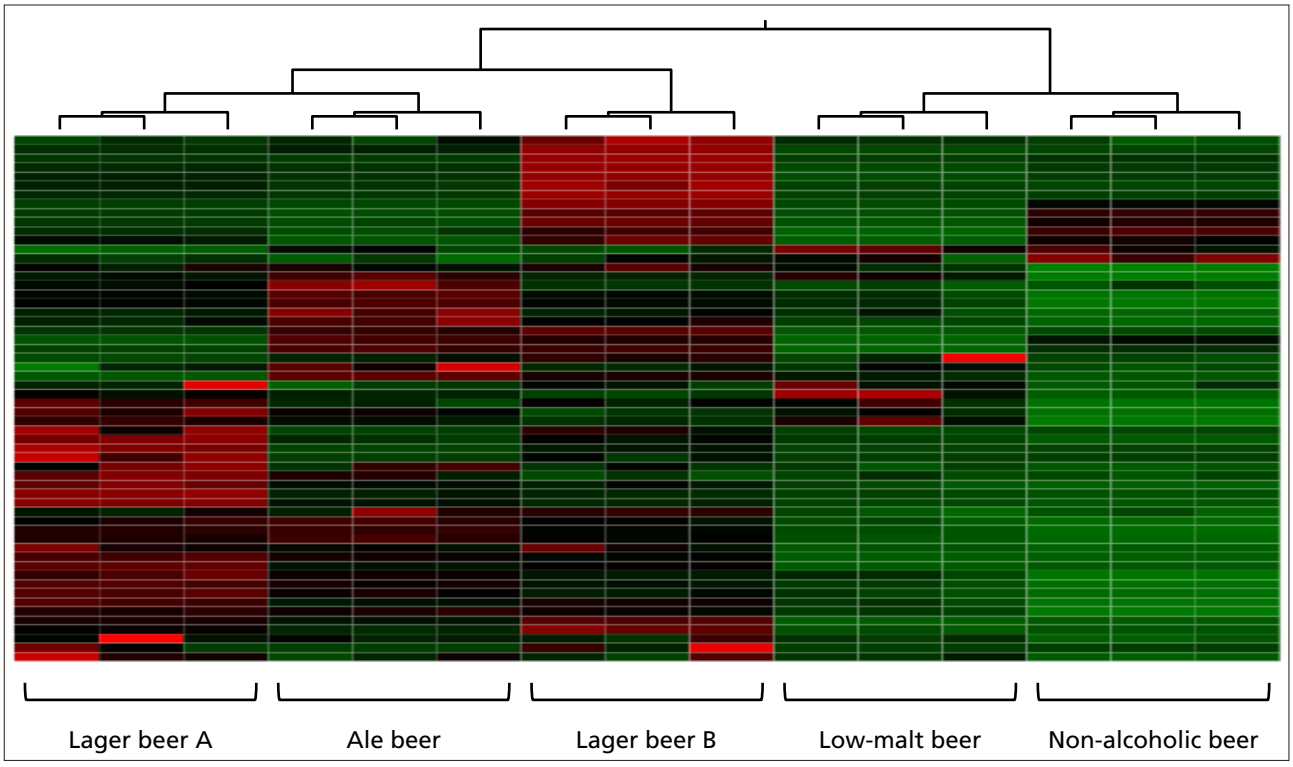

Fig. 3 Hierarchical Clustering Analysis of Five Beers

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