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단 신

복합향으로부터 휘발성 유기성분의 신속한 분리

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A Rapid Sampling of Volatile Organic Components from Complex Flavors

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Flavors are complex mixtures of both the volatile and the nonvolatile organic components, many of which are sensitive to heat and traces of oxygen. Therefore, any satisfactory technique for extracting the volatile flavor components should be carried out at room temperature with minimal contact with air.

A recent steam distillation/column fractionation technique[1,2] may still destroy flavor components or create flavor artifacts, and low boiling volatiles will be lost during the solvent removal in the concentration step.

Headspace adsorption of volatiles[3~5] may ignore the contributions to the flavor of higher boiling materials.

By a simple microextraction technique[6], a relatively small quantity of a flavor essence was extracted with an even smaller quantity of solvent, thus eliminating the solvent removal step.

The purpose of the present work was to develop a simple and rapid method for the isolation of volatile flavor compounds from complex flavor matrices suitable for the direct analysis by glass capillary column gas chromatography. The technique is based on the principle of the column elution technique[7]: The elution is made on a micro-scale under the stream of inert gas to achieve the maximum yielding of volatile compounds with minimum contamination and artifact formation. The obtained GC chromatograms might be used as the fingerprinting pattern of each flavor.

EXPERIMENTAL

Flavors. Compound flavors, chocolate(Hanbul Chemical & Agricultural Co., Korea) and cherry (Heinr Bogwaldt Co., Germany), were chosen to test the efficacy of the present sampling technique.

Reagents. Dichloromethane (GR grade, E. Merck, Darmstadt, Germany). Ethylacetate(GR grade, Hayashi Pure Chemical Ind. Ltd., Japan). Carbopack B(Supelco, Inc.,

Bellefonte, Pa., USA). Chromosorb W, AW-DMCS, 80/100 mesh (Varian Aerograph, Walnut Creek, Ca, USA). Glass bead, 60/80 mesh (Gasukuro Kogyo Co., Ltd., Japan).

Isolation of Volatile Compounds. The isolation of volatile compounds were performed using the sampling system as shown in *Fig.* 1. Dimensions are as indicated. The system is composed of two parts: a glass tube and a microcolumn. The glass tube is a modified Kimax® culture tube with its plastic screw cap drilled a hole through which the isolation column can be tightened to the tube *via* a silicon O-ring. The microcolumn was packed with *ca.* 0.16 m*l* of an appropriate adsorbing material. After packing, the column was purified with a series of organic solvents such as methanol, acetone, ethylacetate, and dichloromethane, and then dried in the oven (80°C). The sampling procedure is illustrated in *Fig.* 2: A 20μl aliquot of a compound flavor was injected into the glass wool on the bottom of the microcolumn and then purified N_2 gas was allowed to flow through the column *via* the side arm until the one-third of the column was impregnated with the liquid. A 200μl of organic solvent was placed into the tube and was forced to go up through the column for the period of 8 minutes under the slow stream of the gas, and the eluate was collected.

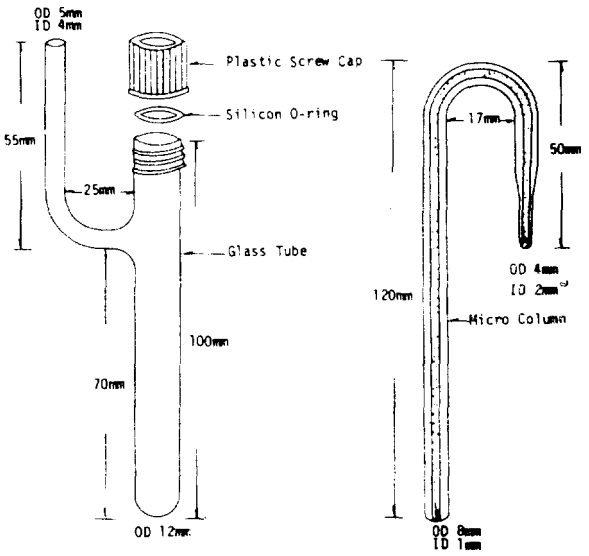

Fig. 1. Basic parts of the apparatus.

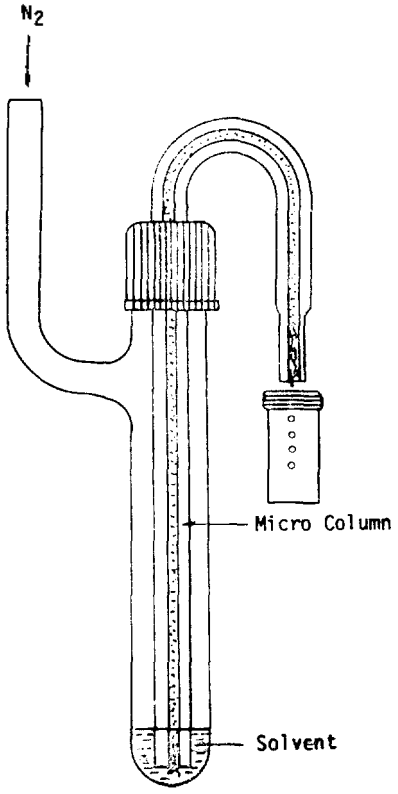

Fig. 2. Sampling system.

Gas chromatographic separation. Each flavor eluate was analyzed directly using a Hewlett-Packard Model 5840A gas chromatograph equipped with a flame ionization detector and a Model 18835 B capillary inlet system (Hewlett-Packard, Avondale, Pa, USA). A glass capillary column (16m×0.25mm I.D.;

coated with Carbowax 20M; Supelco, Inc., Bellefonte, Pa, USA) was employed. Capillary inlet system maintained at 250°C was operated either at splitless injection mode with inlet purge flow of 50 m*l*/min and purge delay time of 0.5 min, or at split injection mode with split ratio set to 60:1. The flow of nitrogen gas through the column was 0.8m*l*/min, and the flow through the detector kept at 280°C was 30m*l*/min furnished with make-up gas.

RESULTS AND DISCUSSION

The compound flavors under the investigation are red-brown opaque viscous liquids of little known compositions, which are soluble both in water and alcohol. Information on changes in their qualities is quite limited.

The present sampling device allows the microcolumn elution to effect a separation of the volatile flavor components from the nonvolatile materials, which may be detrimental to GC system. The colorless eluates obtained were suitable for direct analyses by GC. The micro-scale procedure required very little quantities of the flavor and the eluting solvent. Therefore, the whole sampling could be completed within 10 minutes. The elution was performed under the inert gas stream at room temperature, thus minimizing the possible contamination or artifact formation.

The elimination of the concentra-

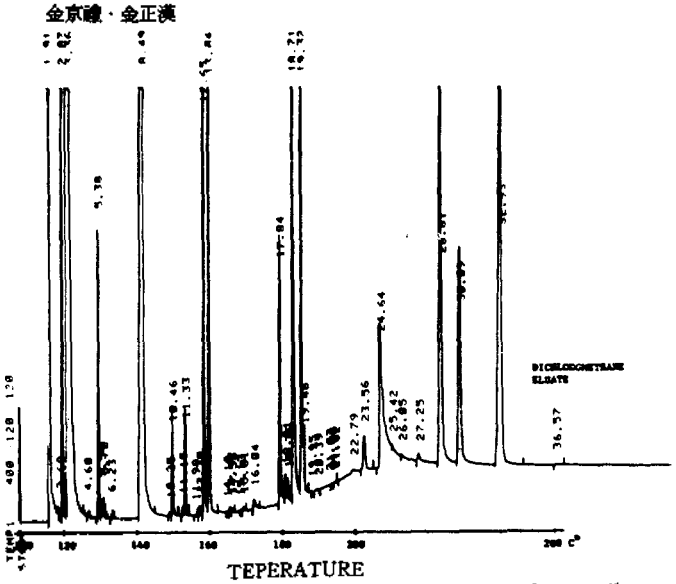

Fig. 3. GC profile of cherry flavor. Column, glass capillary (16 m×0.25 mm I.D. coated with Carbowax 20 M; N_2 carrier flow rate, 0.8 m*l*/min; septum/seal flow, 0.5 m*l*/min; 3μl injection in split mode operation; split ratio, 60:1; column temperature, 120°C for 3 min, then programmed to 200°C at 4 °C/min; injector, 250 °C; detector, 280 °C.

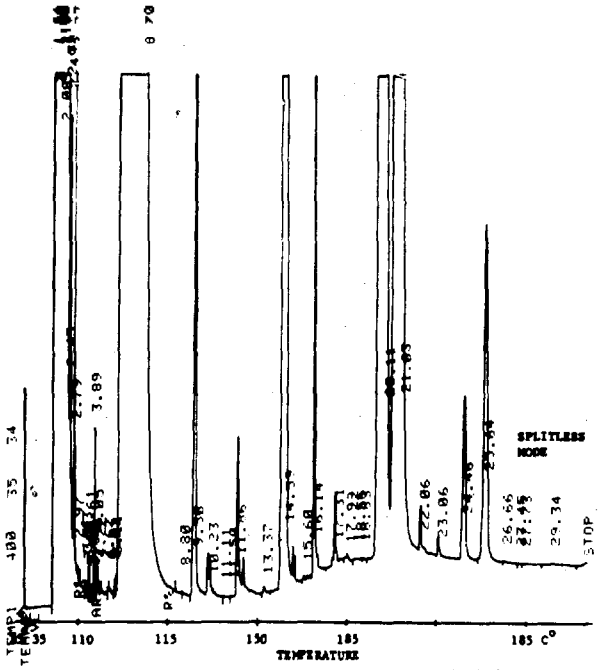

Fig. 4. GC profile of chocolate flavor (splitless mode) 0.8 μl injection in splitless mode operation; purge flow, 50 m*l*/min; purge delay time, 0.5 min; column temperature, 35 °C for 0.5 min, then programmed to 110 °C at 30 °C/min, and then to 185 °C at 4 °C/min; other conditions are the same as in *Fig.* 3.

tion step kept volatiles from being lost.

In order to optimize the sampling procedure, preliminary experiments were conducted for choosing the appropriate sorbent and solvent like Carbopack B, glass bead and Chromosorb W, and ethylacetate and dichloromethane. Chromosorb W as the adsorbing medium and dichloromethane as the solvent were chosen.

Wall-coated open tubular glass capillary columns were utilized to achieve good resolutions of the volatile flavor components. The inertness and high resolving power make the glass capillary columns in GC[8] have now a wide ranging applications to biomedical, petrochemical, environmental, flavor and fragrances areas. They can give fingerprint analyses, to determine if the sample has been changed in its quality or contaminated, thus being used for the quantification of virtually all of the major and minor peaks.

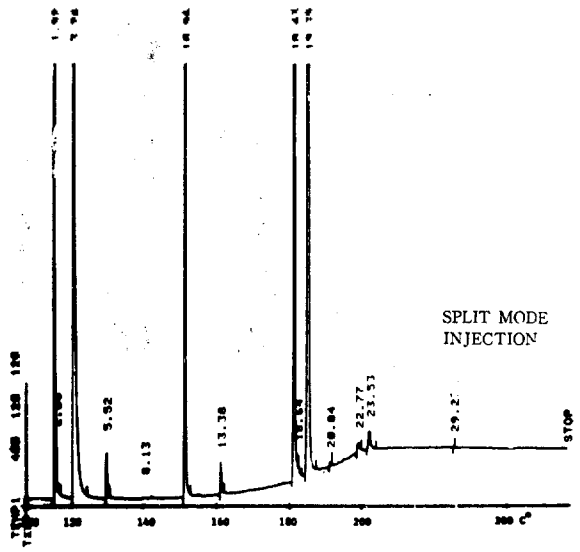

Fig. 5. GC profile of chocolate flavor (split mode). Conditions are the same as in *Fig.* 3.

Due to the polar nature of the flavor components, Carbowax 20M could only give a good resolution. *Fig.* 3C shows the well-resolved profile of the cherry flavor.

"Grob" splitless injection technique[9] was required for the chocolate flavor because of the concentration problem. The splitless injection technique is now best suited for the trace analyses[10,11]. The splitless mode as in *Fig.* 4 yielded a chromatogram, composed of over 20 peaks wih peak areas exceeding 1000 counts. Note that minor peaks were not detected in the split mode evidenced by *Fig.* 5.

The differences of the two compound flavors in their flavor quality are clearly reflected by the composition differences as seen in chromatograms of *Fig.* 3 and *Fig.* 4.

The sampling procedure described here will need further refinements in terms of the sampling variables to meet the requirement for the desired analytical information from flavors of interest.

The sampling technique may prove useful in the rapid evaluation of the various types of flavor products for quality control on a routine basis.

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