Isolation of Volatile Flavour Compounds from Fats and Oils by Vacuum Carbon Dioxide Distillation

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The volatile flavours of foodstuffs and other natural products occur in general in very low concentrations. Because most of them in addition are dissolved in the water or lipid phase, the vapour pressure of these compounds over the products is extremely low, which can make the detection of some important flavour compounds by, for example, the so-called "head space" methods quite impossible. The isolation of the volatile flavours is therefore very difficult, especially if a near-quantitative recovery of these compounds is desired. The isolation method commonly used in flavour analyses is vacuum steam distillation. The complete condensation of water in the cold traps and its low molecular weight make the distillation very simple and effective. The use of water has, however, some disadvantages, especially in the isolation of flavour compounds from fats. Firstly, the condensed flavour compounds must be extracted from the distillate, which requires considerable amounts of solvent, and during the concentration of this extract for gas chromatography losses of the flavour compounds occur. Secondly, the water hydrolyzes the fat to some extent, producing volatile free fatty acids which interfere with the gas chromatographic separation of the other volatile compounds. Likewise some flavour compounds, such as esters and lactones, are hydrolyzed by the influence of water. In the method used in this work, water (steam) was replaced with carbon dioxide, which was passed through the molten fat and condensed together with the volatilised flavour compounds in a liquid nitrogen (or liquid air) trap. Since the vapour pressure of carbon dioxide at this temperature is very low (*e.g.* only 1.3×10^{-5} mm Hg at -180°C[1]), it is possible to perform the distillation without difficulty at a pressure of 1–10 mm of mercury. A schematic diagram of this distillation apparatus is presented in Fig. 1. Since the accurate measurement of carbon dioxide fed directly from a gas cylinder into the vacuum distillation apparatus is somewhat inconvenient, 500–700 g solid carbon dioxide (dry-ice) which has been weighed into the reservoir D prior to the distillation was used. Commercially available dry-ice was found to be very pure, containing only traces of low molecular weight organic impurities. To

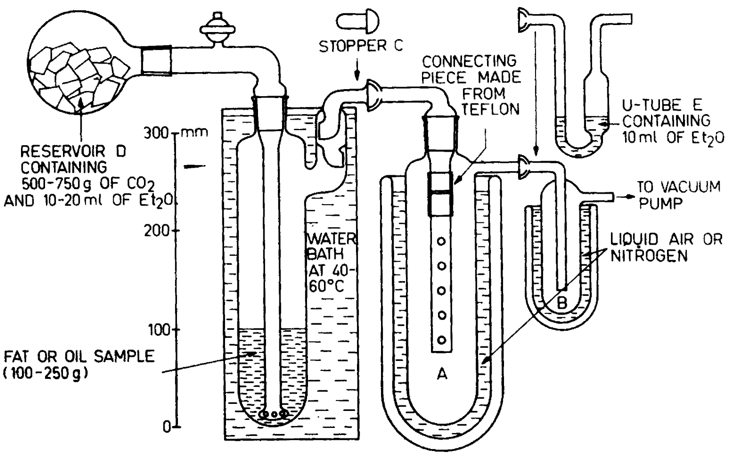

Fig. 1. A schematic diagram of the carbon dioxide distillation apparatus for the removal and collection of the flavour compounds from fats and oils.

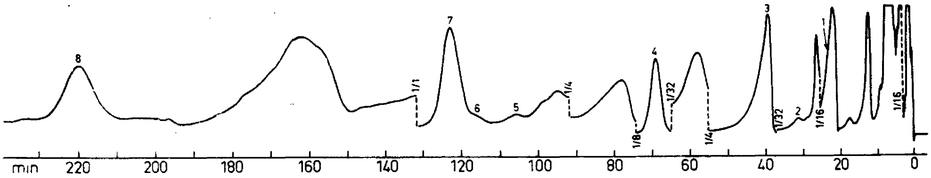

Fig. 2. Gas chromatogram of the flavour compounds of milk fat. (Ethylene glycol succinate column at 180°C, nitrogen flow 70 ml/min. The distillation was performed at 10 mm Hg). The peaks 1, 2, 3, 4, 7, and 8 have been identified as δ-C_6-, δ-C_7-, δ-C_8-, δ-C_{10}-, δ-C_{12}-, and δ-C_{14}-lactone, and peaks 5 and 6 as indole and skatole, respectively.

obtain an even distribution of the extraction solvent in the condensed carbon dioxide, 10—20 ml of a suitable low-boiling solvent, such as ethyl chloride, diethyl ether, or dichloromethane, was added to the reservoir D and passed as vapour together with the carbon dioxide through the fat. About 100—150 g of carbon dioxide per hour can be passed through the fat in this distillation system. The temperature of the fat was held at 40—60°C. After completion of the distillation the cold trap A was removed from the liquid nitrogen, closed with the stopper C and the carbon dioxide was allowed to sublime slowly during a period of 15—18 h at atmospheric pressure through the U-tube E (at −78°C) containing 5—10 ml of the extraction solvent, the temperature in the cold trap (A) rising to −78°C. The losses of the flavour compounds in the evaporating carbon dioxide at this stage were very small. The remaining solution in the trap was combined with the extraction solvent in the U-tube, dried and concentrated to a final volume of 250—500 µl in a special distillation apparatus[2] for the gas chromatographic runs. It is not possible to avoid completely the vaporization of some low-boiling flavour compounds along with the evaporating extraction solvent, but the losses are small, however, since the amount of the solvent to the evaporated is considerably smaller than in the case of vacuum steam distillation.

An approximate equation was derived earlier[3] by the theoretical treatment of the vaporization of a volatile compound from a nonvolatile liquid into a flow of carrier gas:

$$N \approx \frac{P}{p_0} n_1 \ln \frac{c_0}{c_1}$$

where N is the number of moles of gas required to lower the initial concentration (c_0) of the flavour compound to the final concentration (c_1), P the distillation pressure, P_0 the vapour pressure of the pure flavour compound at the distillation temperature and n_1 the number of moles of the involatile liquid. In practice the amount of the carrier gas is higher than the theoretical amount, since equilibrium between the gas and liquid phases in the distillation apparatus is not fully attained.

Since many of the higher-boiling flavour compounds in milk are more soluble in the lipid phase, it is possible to use milk fat in milk flavour studies. A clear milk fat with a low water content can be readily obtained from whole milk by means of a laboratory separator and centrifuge at a temperature of 30—35°C. The effectiveness of this distillation method for the isolation of higher-boiling flavour compounds from milk fat appears in Fig. 2. Even such high-boiling compounds as δ-dodeca- and δ-tetradecalactone (b.p. about 320—350°C at 760 mm Hg) are seen in this gas chromatogram. The free fatty acid content of the extract is also extremely low, permitting a complete separation of the individual flavour compounds.

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