A New Technique for the Isolation of Flavor Components from Fats and Oils

S. S. CHANG,[1] A. E. Staley Manufacturing Company, Decatur, Illinois

A new continuous process for the isolation of flavor compounds from fats and oils has been developed. This new technique involves 1) countercurrent contact of the oil with steam in a 30 plate Oldershaw column, 2) condensation of the steam carrying the flavor compounds in a trap cooled with liquid nitrogen, 3) continuous liquid-liquid extraction of the condensate with a small amount of ethyl ether, and 4) removal of the solvent with a 6 plate Oldershaw column. In this process the oil is heated to 80°C. for only 12 min. The peroxide number of a reverted soybean oil only decreased from 4.1 to 3.4 meq./kg. by this treatment.

Flavor compounds isolated from an oil by this new technique are characteristic of the oil. Very little decomposition occurs during the isolation process and therefore the flavor compounds isolated represent truly the flavor originally present in the oil. Flavor compounds isolated from a reverted-but-not-rancid soybean oil when added to a bland coconut oil at a concentration of less than 10 ppm make it taste exactly like a reverted soybean oil.

THE DEVELOPMENT of gas-liquid chromatography has offered a new approach to the study of flavor components of fats and oils. However, this new technique can only be applied when the flavor components have been isolated from the oil into a form that is suitable for gas chromatography. It is important that during the isolation process no additional oxidative and thermal decomposition products are formed, so that the products isolated are truly representative of the flavor in question. For example, soybean oil develops a beany and grassy flavor known as reversion when stored at room temperature under diffused daylight. The reverted soybean oil is not rancid and its peroxide number is usually only 1–3 meq./kg. In order to collect relatively large amounts of decomposition products for chemical characterization, previous investigators used two types of processes. Martin *et al.* repeatedly steam-distilled soybean oil at 200°C. and the oil was heated to 200°C. under atmospheric pressure after each deodorization to develop reversion flavor (1). Kawahara and Dutton bubbled air for 15 days at 37°C. through soybean oil that had been heated to 60°C. for 3 days with added ferric chloride and cuprous chloride (2). Undoubtedly, during these processes, volatile thermal and oxidative decomposition products were created in relatively large amounts. Such products may overshadow the micro amounts of flavor compounds which are truly responsible for the reversion flavor developed during storage. Recently, Bruyn and Schogt (3) reported a batch process for isolation of volatile constituents from fats and oils by degassing under high vacuum. The present paper reports a continuous process which isolates the flavor components from oil and fat without creating a significant amount of additional decomposition products. This new technique has been used successfully for the isolation of reversion flavor from soybean oil and undoubtedly can be adopted for isolating natural flavor as well as flavor developed during aging from oils and fats.

Apparatus

The isolation of the flavor components from an oil by the present process involves four steps, *viz.*: 1) continuous countercurrent contact of the oil with steam in a 30-plate Oldershaw column under vacuum at 80°C., 2) condensation of the steam which carries the flavor components with traps cooled with liquid nitrogen, 3) continuous liquid-liquid extraction of the condensate with an organic solvent, and 4) removal of the bulk of the organic solvent with a six-plate Oldershaw column to produce a concentrated solution of the flavor components that is suitable for

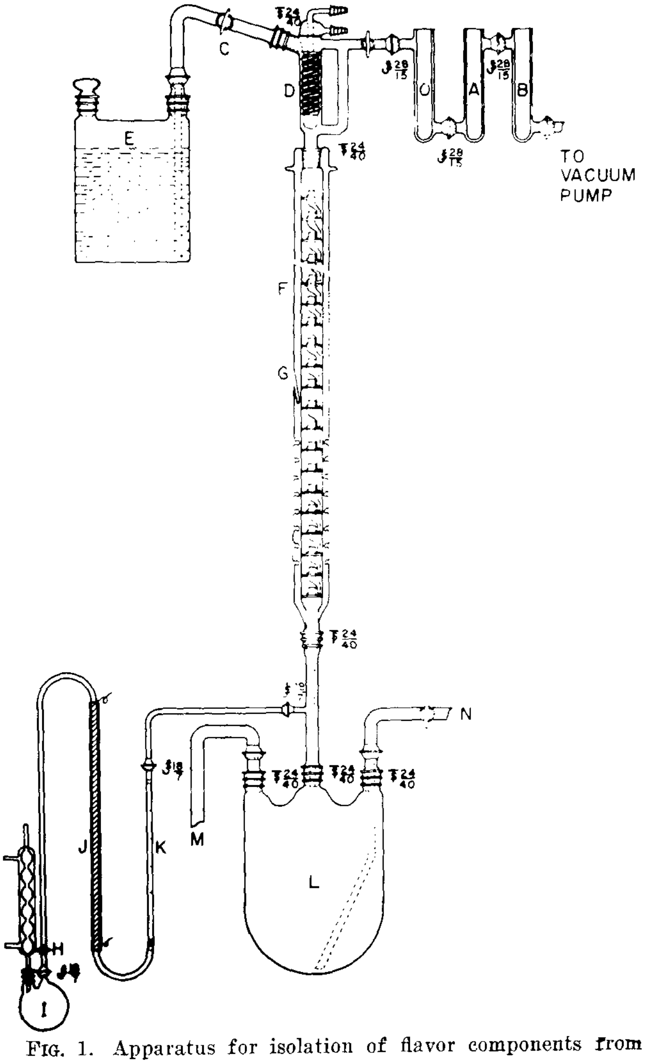

FIG. 1. Apparatus for isolation of flavor components from fats and oils.

---

[1] Present address: Department of Food Science, Rutgers, The State University, New Brunswick, New Jersey.

gas chromatography. The apparatus for each of the steps is described as follows:

Continuous Steam Stripping. The apparatus for transferring the flavor compounds from oil into water is shown in Fig. 1. After the all-glass apparatus is evacuated to a vacuum of 0.1 mm. Hg, the second cold trap A is filled with solid carbon dioxide and the third cold trap B is filled with liquid nitrogen. The stopcock C (K-84300, 2 mm. bore, high-vacuum stopcock, Kontes Glass Company, Vineland, N. J.) is then partially opened to let the oil suck into the heat exchanger D from the reservoir E. The heat exchanger is made from a Frederick Condenser, and is circulated with water maintained at 80 ± 2°C. in a constant-temperature bath. The oil is degassed in the heat exchanger and is heated to 80°C. as it flows down the condenser. The heated oil is dropped on the first plate of the Oldershaw column F (D-1, Glenlab Fractionating-Distillation Column, Braun-Knecht-Heimann Company, Belmont, California). This column is vacuum jacketed and silver plated to minimize heat loss (4). It consists of 30 precision drilled perforated plates sealed in a tube. Each plate is equipped with a baffle to direct the flow of liquid, a weir to maintain the level of liquid on the plate, and a reflux return tube as shown in Fig. 2. When the temperature of the column reached 70°C. as indicated by the thermocouple inserted into the column jacket at G, the stopcock H (K-84300, 2 mm. bore, high-vacuum stopcock, Kontes Glass Company, Vineland, N. J.) is opened to let steam flow into the column from the reservoir I, which is a two-necked round-bottom flask fitted with a reflux condenser and containing distilled water heated to vigorous boiling with a heating mantle. The steam is further heated by heating tape wrapped around the U-tube J to compensate the loss of heat due to expansion. The stopcock H is so adjusted that the rate of flow of the steam as shown by the flowmeter K is 5 g. of steam per hr. After all the bubble plates of the Oldershaw column are filled with the necessary amount of oil, the stopcock C is so adjusted that approximately 150 g. of oil flow into the column per hr. During the operation, the vacuum at the bottom of the column as indicated by a McLeod Gage connected to the flask L at M is maintained below 20 mm. Hg. The steam-stripped oil is accumulated in the flask L and may be withdrawn from the tube N when the flask is full. The steam carrying the flavor components passes through the empty trap O to remove any entrainments and then condenses on traps A and B.

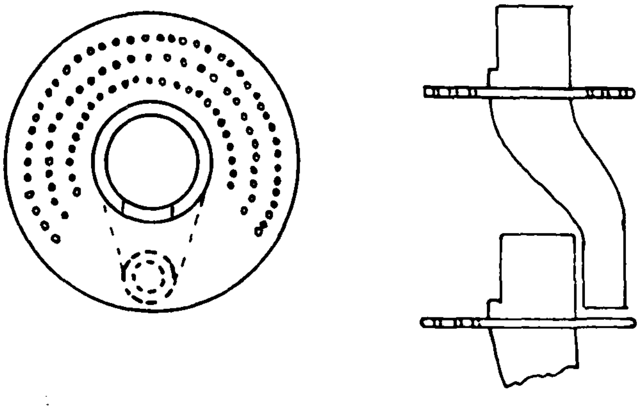

FIG. 2. Plate construction of Oldershaw column.

Liquid-Liquid Extraction. The condensate collected in the cold traps A and B is saturated with sodium chloride and extracted with an organic solvent, such as ethyl ether with a liquid-liquid continuous extractor fitted with a high-efficiency condenser and a fritted glass disk dispersion tube (JE-3150, Extraction Apparatus, Ether, Scientific Glass Apparatus Company, Bloomfield, N. J.) The top of the condenser is connected to a trap cooled with solid carbon dioxide to collect any vapors which are not completely condensed by the condenser. Both the condenser and the extractor are cooled with ice water. The solution is continuously extracted for 8 hr. The extraction is repeated with a fresh portion of ethyl ether. The apparatus is of all-glass construction. It is important that silicone grease is not used on the ground joints. Standard tapered Teflon sleeves may be used.

Concentration of Ethyl Ether Extract. The ethyl ether extract is concentrated to a volume suitable for gas chromatography with a six-plate Oldershaw column fitted with a distilling head with adjustable reflux ratio (6634, distilling head, Hennion Design, Ace Glass Company, Vineland, N. J.). The distillate is checked during suitable intervals by gas chromatography and organoleptic means to determine if any compound besides ethyl ether is distilled. Since the distillation is started with a few hundred ml. of distillant and ended with less than 5 ml., it is necessary to transfer the distillant from a 250-ml. round-bottom flask to a 10-ml. pear-shaped micro flask (ME-501, Metro Industries, Long Island City, N. Y.), near the end of the distillation. To avoid contamination by silicone grease, standard tapered Teflon sleeves are used with the all-glass distillation apparatus.

Experimental

Oils Used. Reverted-but-not-rancid soybean oil was obtained by aging soybean oil refined with acetic anhydride and deodorized at 204°C. Pyrex glass gallon jugs with screw caps were filled with only a minimum of head space left, then closed tightly and set at room temperature for 2 weeks under diffused daylight.

Bland corn oil was obtained by bleaching refined corn oil with 0.5% active carbon and then deodorized at 204°C.

Aged corn oil was prepared by aging corn oil, as obtained from a supermarket, in Pyrex glass gallon jugs at room temperature for one week.

Isolation of Flavor Components. Twenty gal. of reverted-but-not-rancid soybean oil were stripped with 2,200 g. of steam. The condensate collected was extracted twice with 300-ml. portions of ethyl ether. The extract was concentrated to 4 ml. and was then ready for gas chromatography.

Results and Discussion

Reversion flavor of soybean oil has been isolated. Approximately 0.4 g. of a viscous liquid was isolated from 20 gal. of reverted-but-not-rancid soybean oil. Addition of less than 10 ppm of this isolated flavor to a bland coconut oil makes it taste exactly like a reverted soybean oil. The panel also identified a bland corn oil containing this isolated reversion flavor as typical reverted soybean oil. It is interesting to note that the volatile decomposition products obtained by bubbling air into soybean oil when added to coconut oil or corn oil impart a rancid and painty flavor to the oil but do not make it taste like reverted soybean oil.

The flavor compounds isolated by the present technique are characteristic of each oil. The expert flavor panel identified a bland coconut oil containing the volatile components isolated from one-week-old corn oil as corn oil.

During the present process for the isolation of flavor compounds, the oil is only heated to 80°C. for 12 min. Therefore no significant amount of flavor components is produced during the process, and the flavor isolated represents that originally present in the oil. This is further evidenced by two observations. One, when 5 gal. of freshly deodorized soybean oil were treated by the present process, no detectable amount of flavor compounds was obtained. Two, the decrease of peroxide number of the oil during the present process is less than one meq. per kg. The reverted soybean oils subjected to the present process had peroxide numbers ranging from 4.1–7.3 meq. per kg.; after passing through the column, the peroxide numbers were 3.4–6.6, respectively.

Due to the low temperature used in this process, the oil is not completely deodorized. Reverted soybean oil, after passing through the Oldershaw column of the present process, still has a reversion flavor but it is less intense.

The flavor compounds isolated by the present process are free from entrainment of oils. One-tenth ml. of the concentrated ethyl ether solution of reversion flavor of soybean oil as obtained by the present technique was spread on a rock salt plate. The plate was set in a vacuum desiccator at room temperature for 4 min. The residue left on the rock salt plate as a thin film had a strong typical reversion odor. After the plate was kept under vacuum overnight at room temperature, all the reversion odor disappeared and there was no oily residue left on the rock salt plate.

It was found that a 10-plate Oldershaw column can be used to replace the 30-plate column without seriously affecting the isolation of flavor compounds. When the short column is used, the steam stripping can be operated under a vacuum of 10 mm. Hg at the bottom of the column. It was also found that carbon dioxide can be used to replace steam. A high purity carbon dioxide gas cylinder was attached to the flowmeter K (Fig. 1). The carbon dioxide gas, after passing through the Oldershaw column, was condensed in the liquid nitrogen trap B as solid carbon dioxide. After the stripping operation the traps A and B were taken off and the liquid nitrogen in trap B was removed. The solid carbon dioxide condensed on trap B was allowed to evaporate slowly and the gas was led to pass through trap A, still cooled with dry ice. After all the solid carbon dioxide in trap B was evaporated, the flavor compounds left in both traps A and B were rinsed out with ethyl ether. This process is more time-consuming and is less efficient than using steam.

Acknowledgment

The author acknowledges the technical assistance of Paul A. Seaberg who carried out the tedious work of processing large amounts of oils with the reported technique.

REFERENCES

1. **Martin**, C., Schepartz, A. I., and Daubert, B. F., J. Am. Oil Chemists' Soc., *25*, 113–117 (1948).
2. **Kawahara**, F. K., and Dutton, H. J., J. Am. Oil Chemists' Soc., *29*, 372–377 (1952).
3. **Bruyn**, J. de, and Schogt, J. C. M., J. Am. Oil Chemists' Soc., *38*, 40–44 (1961).
4. **Collins**, F. C., and Lantz, Vernon, Ind. Eng. Chem., *18*, 673–677 (1946).

[Received May 26, 1961]

Characterization of the Reversion Flavor of Soybean Oil

S. S. CHANG,[1] K. M. BROBST, H. TAI, and C. E. IRELAND,
A. E. Staley Manufacturing Company, Decatur, Illinois

The reversion flavor isolated from reverted-but-not-rancid soybean oil was separated into 14 fractions by gas-liquid chromatography. The gas chromatographic fractions were collected in specially designed gas cells and polyethylene capillary traps for the determination of their infrared spectra by micro techniques. Ethyl formate, ethyl acetate, ethyl alcohol, n-butyraldehyde, 2-heptanone, and 2-heptenal were positively identified. Presence of alcohol, ester, and possibly dimethyl amino compounds in the fractions with higher chromatographic retention times was indicated by infrared analyses. Ultimate analyses of the flavor compounds isolated from reverted-but-not-rancid soybean oil also indicated the presence of nitrogen compounds.

THE DEVELOPMENT of a characteristic beany and grassy flavor known as reversion during the storage of refined and deodorized soybean oil is a classical problem of the soybean oil industry. A number of mechanisms have been postulated for the formation of reversion compounds. None of them has been unequivocally accepted. Mattil (1) found that addition of the nonsaponifiable extract of hydrogenated soybean oil to either refined cottonseed oil or refined peanut oil caused these oils to develop flavors characteristic of reverted soybean oil. Chang and Kummerow (2) reported that oxidative polymers of polyunsaturated fatty esters, if formed during the processing of soybean oil, may serve as one of the precursors of the reversion compounds. This theory appears to have been substantiated by Evans *et al.* (3), who prepared oxidative polymers by heating peroxides of soybean oil to conditions approximating those of deodorization and found that addition of the dimers to soybean oil significantly decreases its flavor stability. But the most extensively studied hypothesis (4, 5, 6) is that the reversion compounds are oxidative decomposition products of linolenic acid. Recently 3-hexenal was reported to have a green bean odor (7). This seems to indicate that linolenic acid is at least one of the precursors of the reversion flavor of soybean oil.

On the other hand, there are a number of experimental observations which cannot be explained by the linolenic acid theory. The development of reversion

---

[1] Present address: Department of Food Science, Rutgers, The State University, New Brunswick, N. J.