Analysis of Vegetable Oils for Flavor Quality by Direct Gas Chromatography[1]

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ABSTRACT AND SUMMARY

Recently, a direct gas chromatographic method for examining volatiles in vegetable oils was reported [Dupuy et al., JAOCS 50:340 (1973); and Dupuy et al., Ibid. 53:628 (1976)]. The procedure stimulated the development of instrumental techniques for determining odor and flavor characteristics of vegetable oils. This symposium paper describes modifications of the original direct gas chromatographic procedure that substantially enhance its sensitivity and applicability. Profiles of volatiles for several experimental oils obtained by the modified system are presented, together with mass spectral data characterizing significant flavor-related peaks. Regression analysis of the instrumental data, with oil flavor scores, indicates that reliable flavor characteristics of high- and low-quality vegetable oils may be obtained rapidly and efficiently by instrumentation.

INTRODUCTION AND REVIEW

The detection and evaluation of volatile (flavor) components in food products has, until very recently, been limited to taste panel procedures. Such operations are complex (1). Establishing the necessary physical installations (2), training and selecting qualified sensory judges, and statistically treating the data involves much time, effort, and expense (3). Even under optimum conditions, sensory testing is limited by the taster's subjectivity. Early attempts to objectively measure volatile materials with the gas chromatograph (GC) centered on various enrichment techniques for providing volatiles in sufficient concentration to meet instrumental requirements. Solvent extraction and distillation (4,5) have been used frequently; however, solvent extraction is not quantitative and in some instances may introduce extraneous volatiles that originate in the solvent. Distillation procedures (6) are inherently slow and tedious, and the heat they require tends to induce chemical changes in the analytical sample. Although high-vacuum distillation (7-9) reduces the amount of heat required, it remains a complex and time-consuming operation. The analysis of headspace vapors for detecting vegetable and fruit aroma (10), and the direct vapor analysis of various food products (11) have been reported, but these methods require special preparation of the analytical sample and subsequent transfer of a vapor aliquot to the GC.

More recently, an innovative direct gas chromatographic method for the examination of volatiles in salad oils and shortenings was introduced (12). This method requires no preanalysis of the sample. The oil is simply placed directly into a GC inlet liner tube packed with glass wool and inserted into the heated inlet of the GC. When properly secured, the combination of heat and carrier gas sweeps the volatiles from the oil onto the chromatographic packing. The adsorbed volatiles are then resolved into a profile pattern by temperature programming. Oil flavor scores have been correlated with profiles of volatiles obtained by this method (13). These correlations indicate that the flavor characteristics of vegetable oils can be reliably assessed by this simple, direct technique. In addiiton, compounds associated with off-flavors can be identified. Other researchers have examined the potential of this procedure with positive results. Williams (14) substantiated the effectiveness of the method for evaluating flavor characteristics of fresh and light-treated soybean oils, stating that "extremely high correlation between the volatiles profile data and the flavor scores was found." Jackson (15) obtained increased sensitivity and improved reproducibility by adding the oil sample in one leg of a 2 ft U-tube packed loosely with volatile-free glass wool. The details of other useful variations of the basic, direct GC concept are contained in the symposium papers which follow.

This paper reports modifications of the original direct gas chromatographic method (13) for determining oil flavor quality. The described changes materially enhance sensitivity and resolution, yet the innate simplicity of application is retained. Profiles of volatiles obtained by the improved technique are given for several experimental oils. Mass spectral data are presented that characterize several significant flavor-related peaks. Regression analysis of the instrument data, with oil flavor scores, indicates that flavor characteristics of vegetable oils can be obtained objectively by instrumentation.

EXPERIMENTAL PROCEDURES

Sample Preparation and Analysis for GC

A 3-3/8 in. length of 3/8 in. OD borosilicate glass tubing was packed with volatile-free glass wool, loose enough to

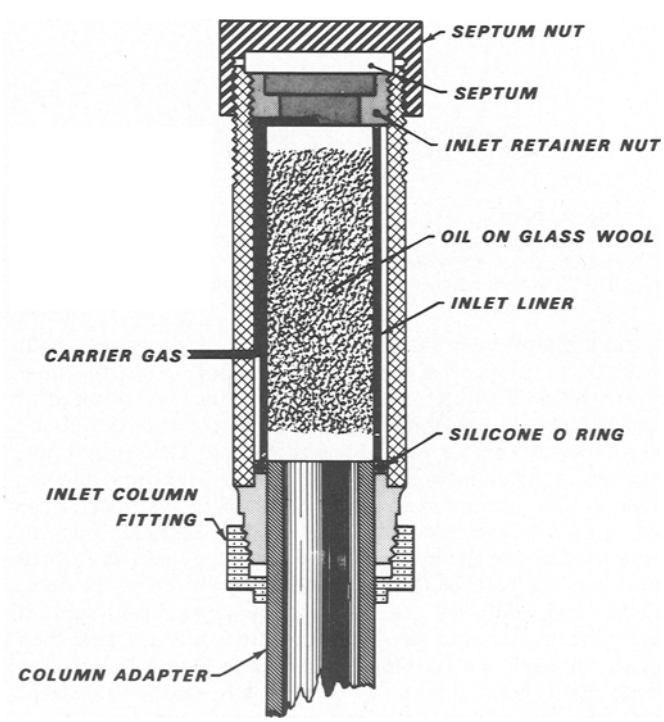

FIG. 1. Cross section of inlet of gas chromatograph showing inlet liner with sample.

[1]Presented at the AOCS Meeting, New Orleans, April 1976.

TABLE I

Mass Fragment Table

Compound	Fragment (m/e)[a]
Pentane	43-42-41-57-44-55-72-39
Hexane	57-43-41-56-42-71-86-39
Pentanal	44-41-58-43-57-71-86
Hexanal	44-41-56-57-43-72-82-67
Heptadienals	81-110-41-43-39
Decadienals	81-41-55-39-95-110-152

[a]Listed in decreasing relative intensities of standards and unknowns for first four compounds. Standards for heptadienals and decadienals were not available.

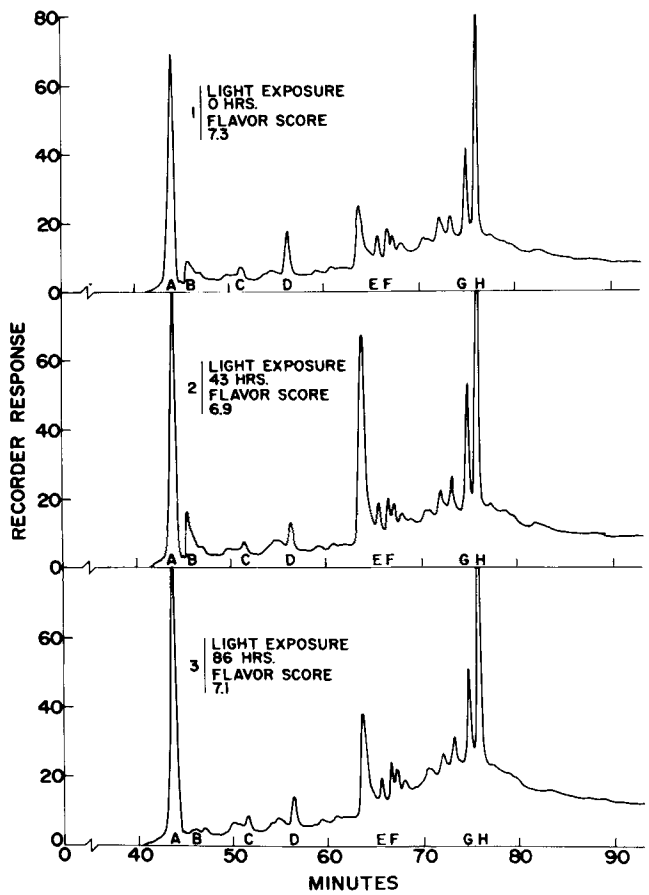

FIG. 2. Profiles of volatiles for three soybean oils obtained by direct gas chromatography. A, pentane; B, hexane; C, pentanal; D, hexanal; E, *trans*-2,*cis*-4-heptadienal; F, *trans*-2,*trans*-4-heptadienal; G, *trans*-2,*cis*-4-decadienal; H, *trans*-2,*trans*-4-decadienal.

permit diffusion of oil throughout the packing, yet tight enough to prevent seepage of the sample from the liner onto the GC column. Clearance of 1/4 in. was allowed at the bottom of the liner and 1/2 in. at the top. A 600 mg vegetable oil sample was added at the top. The septum nut, septum, and retainer nut of the GC were removed, and the liner containing the sample was inserted in the inlet of the GC on top of the silicone O-ring. When the retainer nut was tightened above the upper lip of the liner, a seal was formed between the base of the inlet and the lower lip of the liner. On closing the inlet system with the septum and septum nut, the carrier gas was forced to flow upward and then down through the sample, as is shown in Figure 1. Volatiles were rapidly eluted from the sample as the carrier gas swept through the heated liner and were adsorbed on the top portion of the column, which was maintained at 30 C during the initial hold period of 40 min. The liner containing the spent sample was removed from the inlet, the

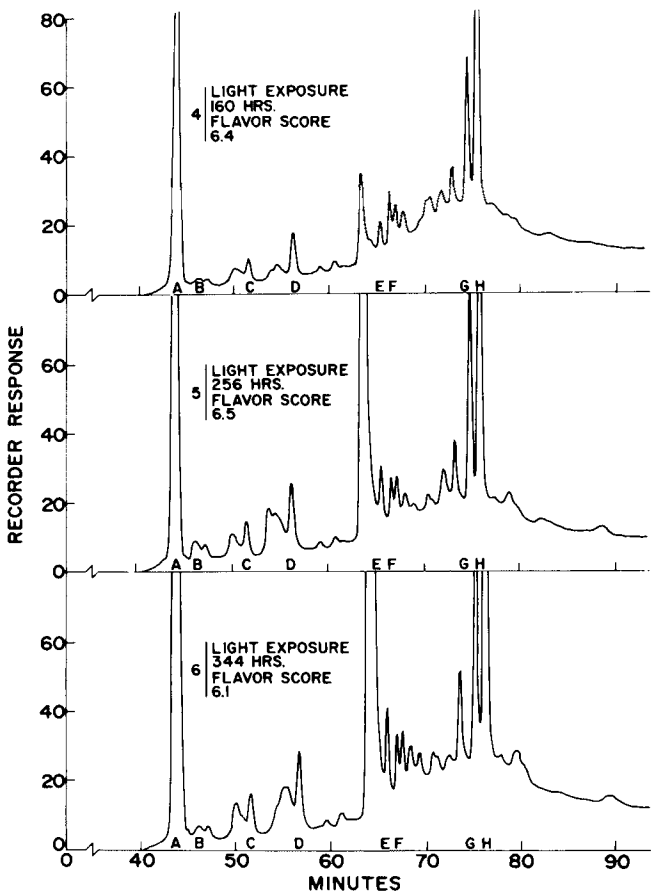

FIG. 3. Profile of volatiles for three soybean oils obtained by direct gas chromatography. A, pentane; B, hexane; C, pentanal; D, hexanal; E, *trans*-2,*cis*-4-heptadienal; F, *trans*-2,*trans*-4-heptadienal; G, *trans*-2,*cis*-4-decadienal; H, *trans*-2,*trans*-4-decadienal.

integrator and programmer were turned on immediately, and the temperature was raised to 100 C in 5 min. Temperature programming was then begun; when complete, the temperature was maintained on final hold to elute and resolve all of the volatiles adsorbed on the column. The oven was then cooled to 30 C in preparation for the next sample.

Sample Preparation for Mass Spectrometry

A silicone membrane separator was used to interface the GC with the mass spectrometer. The quantity of volatiles that permeates this membrane and enters the mass spectrometer varies considerably, depending on the temperature and the polarity of the compound. When small peaks from a profile of volatiles are to be identified by mass spectrometry (MS), it is sometimes necessary to increase the oil sample size to adequately supply the mass spectrometer with detectable quantities of the volatiles. This is achieved by following the method described under "Sample Preparation and Analysis for GC" to the stage where the inlet liner is discarded. At this time, a second inlet liner (prepared in a manner similar to the first) is inserted in the GC inlet, and the process of elution is repeated. When complete, normal temperature programming is carried out. Using this technique, the quantity of oil sample and the volatiles eluted are effectively doubled. This "double liner" technique is useful not only for mass spectral application, but also for substantially increasing the sensitivity of the GC method for obtaining profiles of volatiles of high quality oils whose low volatiles concentration require maximum detection potential.

Materials: Tenax GC, 60-80 mesh; a thermostable porous polymer (2,6-diphenyl-paraphenylene oxide); Poly MPE

(poly-metaphenoxylene) (16), and silicone O-rings were obtained from Applied Science Laboratories, State College, PA. The silicone O-rings were heated at 200 C for 2 hr to remove volatiles. Pyrex glass wool, obtained from Corning Glass Works, Corning, NY, was heated at 200 C for 16 hr to remove volatiles. The partially hydrogenated, flavor-scored, light-treated soybean oils were provided by the AOCS Flavor and Nomenclature Committee (AOCS-FNC). The oils were flavor scored by twelve industrial taste panels on a 1 to 10 flavor scale. The number of sensory judges in an individual panel varied from 5 to 18, and in all, comprised 150 panelists. These are considered to be the most accurately flavor-scored oils available and thus, very suitable for this type of study.

Gas Chromatography: The following conditions were used to obtain profiles of volatiles from the oils.

Instrument: Tracor MT-220 gas chromatograph with dual independent hydrogen flame detectors, a Westronics MT22 recorder, and a Hewlett-Packard Integrator, Model 3380A.

Columns: Stainless steel U-tubes, 1/8 in. OD, 9 ft long, packed with Tenax GC which had been coated with 10% Poly MPE.

Flow Rates: Nitrogen carrier gas, 60 ml/min in each column; hydrogen 60 ml/min to each flame; air, 1.2 ft^3/hr (fuel and scavenger gas for both flames).

Temperatures: Inlet temperature was 170 C. Detector was at 250 C. Column oven maintained at 30 C during initial 40 min hold period. After removal of inlet liner, colunn heated to 100 C within 5 min, then programmed 3 C/min for 30 min. Final hold at 190 C for 30 min.

Special seal: A silicone O-ring was positioned around the 1/4 in. stainless steel adaptor, which projected into the bottom of the inlet of the GC.

Attenuation: 10 x 4.

Mass Spectrometry:

Instruments: Tracor Model 222 GC interfaced with a Hewlett-Packard (Quadrapole) spectrometer, Model No. 5930A.

Ionization Potential: 70 eV.

Scan range: 31 to 235.

Data Processing: INCOS 2000 mass spectrometer data system.

GC Conditions: For GC/MS analysis, the same conditions were used as for GC described earlier, except that helium was the carrier gas.

RESULTS AND DISCUSSION

Lightly hydrogenated soybean oils exposed to light for 0, 43, 86, 160, 256, and 344 hr, were evaluated by direct GC. These oils were also examined by direct GC/MS, and prominent volatiles such as pentane, hexane, pentanal, hexanal, heptadienals, and decadienals were characterized by their mass fragmentation patterns, as shown in Table I. Based on GC retention times, the heptadienals and decadienals were further characterized as *trans*-2,*cis*-4 and *trans*-2,*trans*-4 geometric isomers.

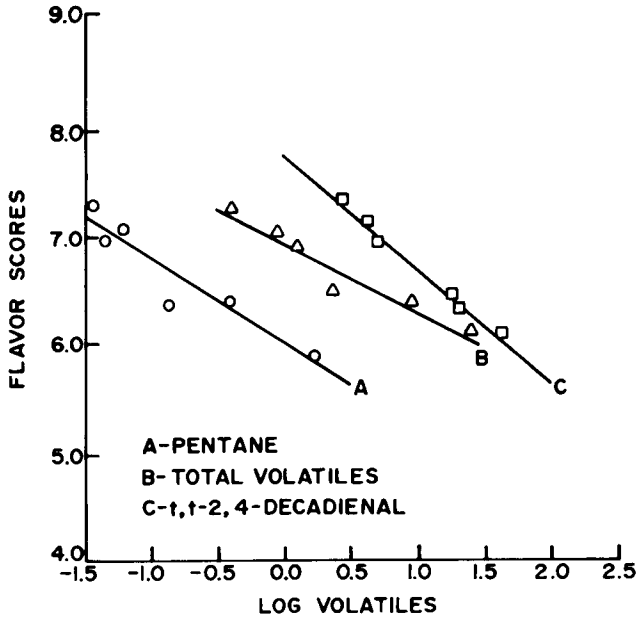

FIG. 4. Regression of the flavor scores on the log of the gas chromatographic volatile peaks: A, pentane; B, total volatiles; C, *trans*-2,*trans*-4-decadienal.

Although the light treatments resulted in only slight deterioration of these oils as indicated by the AOCS-FNC flavor scores, the profiles of volatiles shown in Figures 2 and 3 indicate a gradual increase in the intensity of individual and total volatiles (TV) with prolonged light exposure. Certain volatiles appear to be uniquely sensitive in this regard, hence they are good indicators of changes that occur in oils exposed to light. As shown in Figure 4, regression plots were obtained for the AOCS-FNC flavor score on logarithms of the GC volatile peaks of pentane, TV, and *trans*-2,*trans*-4-decadienal. Although all three of these volatile components correlate well with flavor scores, the *trans*-2,*trans*-4-decadienal appears to be the most sensitive and accurate.

Predictions of oil flavor scores were made by regression for each oil, using the GC data obtained on the pentane, TV, and *trans*-2,*trans*-4-decadienal peaks. The predicted scores are listed in Table II. Of the three peak indicators,

TABLE II

Flavor Scores of Soybean Oils Subjected to Light Exposure

Oil sample	Light exposure (hr)	GC flavor score predictions (1)[a]	(2)[b]	(3)[c]	Taste panel flavor score (4)[d]
A	0	7.1	7.2	7.3	7.3
B	43	7.0	6.9	7.0	6.9
C	86	6.9	7.0	7.0	7.1
D	160	6.7	6.8	6.4	6.4
E	256	6.4	6.4	6.5	6.5
F	344	6.0	6.1	6.1	6.1

[a]Based on log of pentane peak concentration.

[b]Based on log of total area under the curve.

[c]Based on log of *trans*-2,*trans*-4-decadienal peak concentration.

[d]Average of 150 panelists.

TABLE III

Correlation Coefficients of Soybean Oil Volatiles with Flavor Scores or Light Exposure

Volatiles	Volatiles and flavor scores[a]		Volatiles and light exposure	
	linear	log	linear	log
Total volatiles	-0.82*[b]	-0.89*	-0.95**[c]	-0.98**
Pentane	-0.83*	-0.90**	-0.95**	-0.99**
Pentanal	-0.85*	-0.86*	-0.84*	-0.78
Hexanal	-0.71	-0.66	-0.90**	-0.83*
Pentanal & Hexanal	-0.82*	-0.83*	-0.94**	-0.91**
trans-2,*cis*-4-heptadienal	-0.62	-0.64	-0.80*	-0.80*
trans-2,*trans*-4-heptadienal	-0.93**	-0.93**	-0.85*	-0.86*
trans-2,*cis*-4-decadienal	-0.89*	-0.94**	-0.98**	-0.99**
trans-2,*trans*-4-decadienal	-0.97**	-0.99**	-0.96**	-0.95**

[a]Overall average of all accepted scores.
[b]*Significant at the 95% confidence level.
[c]**Significant at the 99% confidence level.

TABLE IV

Comparison of Effectiveness of a Typical Industrial Panel and the Direct Gas Chromatographic (GC) Method for Determining Oil Flavor Quality

	Product moment correlation coeffcient		Spearman Ranking correlation coefficient	
	AOCS-FNC flavor score	Light exposure time	AOCS-FNC flavor score[a]	Light exposure time
Industrial taste panel[b]	+0.62	−0.64	+0.59	−0.59
GC volatile peak, pentane	−0.83	0.95	−0.98	1.00
GC total volatiles	−0.82	0.95	−0.94	0.94
GC volatile peak, *trans*-2,*trans*-4-decadienal	−0.97	0.95	−0.94	0.94

[a]Ranking based on the results of Duncan's multiple range test.
[b]Average of the 12 correlation coefficients for the 12 individual panels.

the *trans*-2,*trans*-4-decadienal values are essentially the same as those reported by the AOCS-FNC taste panels. In addition, it is evident from the profiles of volatiles that this is a prominent peak, that can be detected quite easily using the Tenax GC coated to a level of 10% with Poly MPE (17). This column packing produces better resolution of volatiles with less column bleed than was experienced with the Porapak P packing cited in our previous work (12,13). The resolution of the *trans*-2,*trans*-4-decadienal peak is an important consideration in the analysis of high quality oils because it is a peak of considerable magnitude, easily detectable when volatiles are likely to be present in low concentration.

The correlation of oil flavor scores determined by various GC profile peaks with actual taste panel flavor scores should provide useful information on the validity of the GC method. Such data, however, may not always be completely reliable because taste panels are themselves subject to the inherent weakness of subjective variation. Thus, since taste panel values are not necessarily absolute indicators of actual flavor score, the GC correlations so derived are thereby limited by the proficiency of the taste panels employed.

Oil flavor quality is known to deteriorate progressively with exposure to light; therefore, a correlation of the various GC volatile profile indicators with light exposure time would seem to be more meaningful in estimating the effectiveness of the direct GC method for assessing flavor quality of oils. Table III lists the correlation coefficients of nine GC volatile profile indicators for the six experimental oils, on a linear and log basis, using both taste panel flavor scores and oil light exposure indices of flavor quality. The correlations derived using light exposure times are better than those based on the AOCS-FNC flavor scores. This would suggest that flavor scores obtained with the direct GC method are as good as, if not better, than those derived by taste panel evaluation.

The AOCS-FNC flavor scores supplied for these oils were the combined results of 12 industrial taste panels. Analysis of the flavor scores from the individual taste panels and of the GC peak values enables a direct comparison to be made between the capabilities of each to detect small differences in oil flavor quality. Using the flavor score data from each of the 12 taste panels, correlation coefficients were calculated between the panels' scores and the overall combined scores and between the panels' scores and the oil light exposure time. Two types of correlation coefficients were calculated: (a) the simple product-moment coefficient of correlation; and (b) the Spearman ranking correlation coefficient. The results, summarized in Table IV, are compared with similar correlation coefficients calculated for the GC pentane, TV, and *trans*-2,*trans*-4-decadienal peak. In all cases the GC results produced higher correlation coefficients. Taste panels such as those currently being used in industry were not as effective as GC for either estimating flavor score or determining the relative ranking of flavor quality of the oils examined in this study.

The results of the work with these six oils indicate that flavor related volatiles can be eluted effectively from high quality as well as poorer quality oils, but more work is still needed. A larger number of oil samples and different types of oils should be studied, and different column packings and techniques should be evaluated to improve resolution and assessment of the volatiles eluted.

ACKNOWLEDGMENTS

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