EFFICIENT TRAPPING SYSTEM FOR VOLATILE COMPONENTS EVALUATION IN OILS AND FATS.

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INTRODUCTION

Methods of assessing deterioration in edible oils and fatty foods are based on sensory evaluation or chemical methods which measure one class of lipid oxidation products. Established procedures, such as peroxide value (Walker 1985), free fatty acids (Walker 1985), and thiobarbituric acid test (Tarladgis 1962) are generally indirectly related to quality in the tested products. Chromatographic methods, particularly gas chromatography (GC), are eminently suited for direct determination of trace amount of compounds responsible for off-flavour. Various GC methods have been reported to analyze volatile products from the oxidative deterioration of vegetable oils (Snyder 1988). The advantages and disadvantages of some of these methods have been discussed (Waltking 1983), but relatively few have proved to be quantitatively accurate. This paper reports the development of a simplified and improved dynamic headspace method for quantitative analysis of volatiles.

MATERIAL AND METHODS

Materials

All standards used in this study were purchased from Bedoukian Research. Each standard was individually chromatographed to check for purity. Packing material for precolumn, bonded Carbowax 20M (C20M) was obtained from Ultra Scientific. The analyzed oil samples were generously donated by CSP Foods Ltd. Regular glass wool was purchased from Supelco Canada Ltd. Before using, each batch of glass wool was heated overnight at 400C to removed any organic components trapped on it.

Purging System

All elements of this system were installed inside the gas chromatograph (GC). Injector was slightly modified to permit installation of an O-ring sealing for a glass insert tube. The other side of the tube was sealed by a regular septum used in GC and at this end a 2 mm diameter hole was made to direct carrier gas flow through the insert. A packed precolumn (15cm x 3mm

i.d.) was used for trapping volatiles. It was bent into a U-shape to allow for easy cooling with liquid nitrogen. The precolumn was deactivated inside by high temperature silylation (HTS) or backing with Carbowax 20M (Schieke 1977; Pretorius 1984). The deactivated precolumn was packed with C20M and attached with one end to the injector, and another to the splitter and through it to the capillary column. The carrier gas flow was controlled by a regular pressure regulator present in the GC and an additional backpressure regulator was installed to monitor split ratio at the outlet of the splitter.

Volatile Analysis Procedure

The glass wool plug was placed inside the glass liner with thermally cleaned tweezers. The oil sample was applied onto the wool plug with a pipetor. The sample volume was affected by volatile contents and was usually in a range of 50 - 200 µl. The injector was heated to 50, 75, 90, 110, 150, 175C to determine the optimum temperature. Before placing the sample into the injector, liquid nitrogen in a small Dewar flask was used to immerse the precolumn. The insert tube with oil sample was put inside the injector and the later was immediately closed. Purging of the volatiles was performed for 5, 10, 15, 20, 30 and 60 minutes to optimize the purging time. During purging a flow of carrier gas helium at 60 mL/min was maintained. When the assigned time passed, the tube with remaining oil was replaced by a clean one and the system pressurized to normal working pressure. Liquid nitrogen was removed prior to GC analysis. An internal standard solution of tridecane in a good quality oil (250 ng per sample) was added together with sample.

Gas Chromatography

The volatile components were separated on a fused silica capillary column 60m x 0.32 mm i.d.. The column was coated with 1.0 µm of BD-5 liquid phase (J&W, Rancho Cordova, CA). The flame ionization detector temperature was held at 265C, while column temperature was programmed as follows: 45 to 125C at rate 2C/min then to 185C at 5C/min and finally to 235C at 4C/min. Starting temperature was held for 5 min. and final for 26 min. Peak area was used for all calculations and multipoint calibration curve obtained by direct injection of different concentrations of compounds of interest.

RESULTS AND DISCUSSION

Trap Evaluation

The short precolumn used as the trap for volatile compounds was evaluated for peak broadening with a mixture of components with different functional groups. For this purpose the tailing factor (TF) was calculated for each peak as described by McNair et al.(1969). Measurements of peak half widths at 10% height was performed at fast chart speed (Table 1). A tailing factor of

100% indicated a symmetrical peak.

Table 1. Peak tailing and precolumn deactivation method (%).

Component	Treatment			
	NT	PC20M	HTS	C20M
Hexane	68	76	98	96
Cyclohexanol	52	69	96	98
Octanol	52	71	92	94
Hexanal	64	76	95	91
Hexenal	72	74	91	93
2,4-Heptadienal	69	73	91	90
2-Decenal	64	78	92	94
2,4-Decadienal	49	67	91	90

NT - no treatment and regular Chromosorb W coated with 5% Carbowax 20 ;
PC20M - column packed only with bonded Carbowax 20M
HTS - high temperature silylation and bonded Carbowax 20M
C20M - high temperature deactivation with Carbowax 20M and bonded Carbowax 20M.

The metal precolumn was chosen because of its mechanical strength over the wide range of temperatures. As shown in Table 1. using a precolumn tightly packed with classical packing material produced peaks with high tailing. This factor is particularly important if trace amount of volatiles are to be analyzed accurately. Peak shape also significantly affects the accuracy of integration. Peak broadening is caused by active sites present in the packing and metal wall. The tailing is particularly visible in highly polar components such as alcohols and unsaturated aldehydes. Packing this same column with bonded Carbowax 20M significantly reduced tailing (Schieke 1977). This type of packing is produced by removing metals from Chromosorb and high temperature coating of it with a monolayer of phase. The treatment eliminates any active centres in the packing. High temperature silylation or backing with Carbowax 20M of metal precolumn, almost totally eliminates tailing. Most peaks were symmetrical which allowed trace analysis to be performed. From a practical point of view, single silylation formed a very stable coat of silica derivative, withstanding a few loads of bonded C20M. The bonded forms of Carbowax 20M (packing and deactivation) are extremely susceptible to oxygen and moisture present in the carrier gas, necessitating purification of the latter. Based on laboratory experience one packing can be used for more than 300 purgings of the samples. Flowing helium during manipulations with sample allows it to form a protective blanket preventing oxygen and water from entering the system.

Recovery of the Volatiles During Purging

To evaluate performance of the trapping system and select

the optimal parameters a mixture of 15 different standards was used. To assess the purging time a standard mixture of 2 ppm was analyzed with the results summarized in Table 2.

In all recovery experiments, an internal standard, solution in octane was injected directly into the GC shortly after the injector was closed to eliminate the influence of purging parameters. The coefficient of standard deviation for the internal standard peak area was only 1.4% for all experiments performed, indicative of good reproducibility.

Five minutes was sufficient with this set-up to transfer about half of the amount of the components into the GC system. Doubling the time, the recovery of most components was over 80%. This recovery is probably due to the very large area of evaporation formed by oil coated the glass wool plug. After 15 minutes of purging the recovery for almost every component was over 90% and the CSD of 3.2%. These results indicate excellent reproducibility and quantitative response of this method for the oil volatile analysis at very low concentrations. Increasing the purging time to 30 and 60 minutes did not significantly increase the recovery of the high boiling components, but decreased the recovery of the highly volatile components. As discussed above, 15 minutes appeared satisfactory for quantitative recovery and evaluation of volatile components in oils.

Table 2. Recovery of the standards during purging at 110C at 2 ppm of standards concentration

	Time in minutes					
Component	5	10	15	20	30	60
Pentane	60	87	95	96	90	81
1-Octene	54	89	93	93	90	83
Hexanal	56	86	95	94	90	87
2-Hexenal	53	82	93	91	89	90
Nonanal	55	79	91	90	94	90
2-Octenal	49	80	90	92	95	90
2,4-Heptadienal	48	81	91	93	92	94
Methyl Nonanoate	47	76	89	93	94	96
2-Decenal	50	74	90	92	94	92
2,4-Decadienal	52	72	90	89	94	91
2,4-Dodecadienal	41	67	89	92	96	90
1-Octen-3-ol	54	85	92	92	93	86
1-Octanol	58	87	93	91	95	84
Benzaldehyde	63	85	91	90	89	93
Acetophenon	59	81	90	94	90	92
CSD (%)	4.9	3.8	3.1	3.1	3.8	4.7

CSD - coefficient of standard deviation, calculated for all recovery results for each time separately;
All results are average of minimum three replications.

The effect of purging temperature on recovery is shown in Table 3. At the lowest temperature (50C) the recovery was very low for components analyzed. A significant improvement was observed using a purging temperature of 110C with high recovery for all components including high boiling compounds. Using higher temperatures could be impractical as elevated temperature accelerate decomposition of volatiles precursors, hydroxyperoxides, accumulated during oxidation of the oils and fats. Fat hydroxyperoxides are known to undergo rapid decomposition when temperature is raised above 100C (Selke 1987). Volatile profiles obtained at sampling temperature below 100C would, therefore, be representative of flavour compounds in oils at time of testing (Selke 1987). However, analyzed volatiles in oxidized canola oil, ranging in peroxide value from 15 to 20, showed only a 10-15% increase in volatiles when the temperature was raised from 90 to 110C.

Table 3. Recovery of the standards during purging at different temperatures for 15 minutes and 2 ppm concentration.

Component	Temperature (C)					
	50	75	90	110	150	175
Pentane	72	86	90	95	93	81
1-Octene	68	78	89	93	92	90
Hexanal	52	80	88	95	93	94
2-Hexenal	41	83	89	93	93	90
Nonanal	38	78	84	91	92	93
2-Octenal	34	68	81	90	90	96
2,4-Heptadienal	38	54	78	91	93	94
Methyl Nonanoate	30	68	82	89	96	90
2-Decenal	35	67	79	90	95	89
2,4-Decadienal	33	58	81	90	96	90
2,4-Dodecadienal	31	48	78	89	97	93
1-Octen-3-ol	42	65	82	92	91	93
1-Octanol	39	71	84	93	93	89
Benzaldehyde	29	67	85	91	92	90
Acetophenon	41	72	89	90	98	93
CSD (%)	7.9	4.6	3.8	3.1	3.5	3.7

For abbreviations see Table 2.

This increase should be interpreted as due to a temperature increase and not as result of the hydroxyperoxides decomposition. Selke et al.(1987) found a two fold increase in volatiles when the temperature was increased from 90 to 135C. In the experiment performed with oxidized canola oil, a similar increase was observed when the temperature was raised from 90 to 125C, which could be attributed to hydroxyperoxides decomposition. A very low coefficient of standard deviation confirmed that this dynamic headspace analysis is very high

reproducible.

Experiments with different concentrations of components were performed under optimal parameters, 15 minutes and 110C. The standards were diluted in a good quality canola oil at concentrations of 0.05, 0.1, 0.5, 2.0, 5.0, 10.0 ppm. Calculated recoveries for all compounds analyzed was over 90% and independent of concentration. Even at lower concentrations 0.05, 0.1, 0.5 ppm, only slightly higher coefficient of standard deviation were observed 4.8, 4.2, 3.8%, respectively.

In conclusion the method described has proved extremely reliable and reproducible for the analysis of volatiles of oil and fat samples. The average CSD for the internal standard peak calculated each time for each series of samples analyzed was very low at 2.1%.

This method eliminates manipulations with volatiles and makes it very easy to operate. The analytical method described also permits quantitative analysis to be performed at low cost per analysis.

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