The method was found to be practical and gave reliable results with sufficient precision (considering the low levels involved) if enough attention is given to detail.

REFERENCES

1. Kodicek, E., and D.E.M. Lawson, in "The vitamins," Vol. VI, edited by P. György and W.N. Pearson, Academic Press, New York, 1967, p. 211.
2. Mulder, F.J., E.J. deVries and B. Borsje, J. Assoc. Off. Anal. Chem. 54:1168 (1971).
3. Nabholz, A., Trav. Chim. Aliment. Hyg. 68:86 (1977).
4. Bell, J.G., and A.A. Christie, Analyst 99:385 (1974).
5. Hofsass, H., A. Grant, N.J. Alicino and S.B. Greenbaum, J. Assoc. Off. Anal. Chem. 61:735 (1978).
6. Ray, A.C., J.N. Dwyer and J.C. Reagor, J. Assoc. Off. Anal. Chem. 60:1296 (1977).
7. Henderson, S.K., and A.F. Wickroski, Ibid. 61:1130 (1978).
8. Antalick, J.-P., H. Debruyne and J.-G. Faugere, Ann. Falsif. Chim. 70:497 (1977).
9. Mankel, A., Dtsch. Lebensm. Rundsch. 75:77 (1979).
10. Asshauer, J., and I. Halász, J. Chromatogr. Sci. 12:139 (1974).
11. van Niekerk, P.J., and L.M. du Plessis, S.A. Food Rev. 3:167 (1976).
12. Egaas, E., and G. Lambertsen, Internat. J. Vit. Nutr. Res. 49:35 (1979).
13. Hanewald, K.H., F.J. Mulder and K.J. Keuning, J. Pharm. Sci. 57:1308 (1968).
14. Green, J., in "Fat-Soluble Vitamins," edited by R.A. Morton, Pergammon Press, Oxford, 1970, p. 81.
15. Mounsey, J. "Introduction to Statistical Calculations," English Universities Press, London, 1958, p. 280.

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✂*Betula platyphylla* var. *japonica* Seed Oil: A Rich Source of Linoleic Acid

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ABSTRACT

Component fatty acids of the oil extracted from *Betula platyphylla* Sukatchev var. *japonica* Hara (Betulaceae) seeds were analyzed by gas liquid chromatography. The predominant fatty acid was linoleic acid (87%), and together with oleic and linolenic acids the 18-carbon unsaturated acids amounted to 97% of the total acids.

INTRODUCTION

Betula platyphylla Sukatchev var. *japonica* Hara, a member of the family Betulaceae, is a deciduous tree with white bark and is distributed from the middle to northern regions of Japan. No information is available on the fatty acid composition of the seed oil.

In this investigation, oils were extracted from 2 seed samples of different origins and examined for their characteristics and fatty acid composition.

EXPERIMENTAL PROCEDURES

Material

Two seed samples were used in the investigation. One sample was collected in September 1978 from trees grown at Kawagoe-shi, Saitama-ken and the other was collected in the same season from trees grown at Sapporo-shi by the Hokkai-do Forestry Cooperative Association, Sapporo-shi, Hokkai-do, Japan.

Extraction of Oil

Winged nutlets separated from each air-dried sample were dissected into seed and wing fractions by abrasion between the hands. The seed fraction (nutlets) was sorted to remove the loosened wings and trash by a combination of winnowing, sieving and hand-picking. The cleaned seeds were crushed in a mortar and placed in a Soxhlet extractor for reflux with ethyl ether solvent. The ether-extract was treated with hexane in the manner previously reported (1), and the resulting hexane-soluble oil was analyzed for its characteristics and fatty acid composition.

Preparation and Gas Liquid Chromatography (GLC) of Methyl Esters

Each sample oil was saponified in the usual manner. After removal of the unsaponifiable matter, the mixed fatty acids were refluxed with H_2SO_4/CH_3OH to give methyl esters.

The methyl esters were analyzed using a Hitachi 163 gas chromatograph equipped with a hydrogen flame ionization detector and a Takeda Riken TR-2217 automatic integrator. A 4 m x 3 mm stainless steel column packed with 5% diethylene glycol succinate polyester coated on 60/80 mesh Chromosorb G AW was used under the conditions: flow rate of nitrogen 20 ml/min; temperature of column, 200 C; temperature of injection port and detector, 300 C. Identification of each component was made by comparing its retention time with that of a reference sample.

RESULTS AND DISCUSSION

Sample seeds grown in Kawagoe (sample A, 51.3 g) and in Sapporo (sample B, 28.1 g) yielded 9.8g (19.1%) and 7.9g (28.1%) of greenish yellow oils, respectively. Geographical location influenced the oil content. Characteristics of the oils are listed in Table I.

The UV and IR spectra of the oils gave no definite evi-

TABLE I

Properties of Oils and Their Mixed Fatty Acids

	Samples	
	A	B
Oil		
Sp grav (20 C/20 C)	0.9268	0.9241
Saponification value	191.2	192.2
Iodine value (Wijs)	158.3	159.1
Unsaponifiable matter (%)	3.54	1.93
Mixed fatty acids		
Neutralization value	199.0	198.9
Iodine value	167.1	168.1

TABLE II

Fatty Acid Composition of Oils Determined by Gas Liquid Chromatography

Sample	Component acids (% by wt)										
	14:0	16:0	16:1	18:0	18:1	18:2	18:3	20:0	21:0	22:0	24:0
A	0.1	1.3	0.2	0.3	8.5	87.8	1.4	trace	0.1	0.2	0.1
B	trace	1.6	0.2	0.5	9.1	87.2	1.4	trace	trace	trace	–

dence of conjugated unsaturation or of unusual functional groups.

Fatty acid compositions of the oils, based on GLC analyses of their methyl esters, are shown in Table II. Linoleic acid was present in extremely high levels (87.8% and 87.2%, respectively), and together with oleic and linolenic acids the 18-carbon unsaturated acids amounted to 97.7% of the total acids in individual oils. Probably this concentration of linoleic acid (87.8%) is one of the highest so far reported in any seed oil.

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REFERENCE

1. Tanaka, T., S. Ihara and Y. Koyama, JAOCS 54:269 (1977).

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A Rapid Method for Analysis of Refined Vegetable Oils for TBHQ by Gas Chromatography[1]

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ABSTRACT

A simple and rapid extraction technique using acetonitrile solvent has been used to extract TBHQ antioxidant from refined soybean and cottonseed oils. The extracts have been analyzed by a gas chromatographic technique, using silyl derivatization, to quantitate the TBHQ content of the oil. The method has been calibrated using 14 data points for linear regression, showing a coefficient of determination of 0.9587. Average recoveries over a 14-day period range from 102 to 106.6% at the 50-150 ppm levels. The overall test standard deviation, which includes the effects of time, is 5.9.

INTRODUCTION

Gas chromatographic procedures have been proposed to analyze for tertiarybutylhydroquinone (TBHQ) antioxidant in food oils. These procedures employ direct injection of the diluted oil sample (1) or time-consuming extractions, followed by concentration steps which treat the antioxidant severely by drying under vacuum (1,2). While we have sometimes used the direct gas chromatographic injection approach successfully, we also have experienced severe difficulty with the breakdown of TBHQ in the injection port of the chromatograph (D.M. Wyatt, unpublished results).

In the method described in this paper, use of the N,O-bis-(trimethylsilyl)-trifluoroacetamide (BSTFA) derivative of TBHQ conveniently bypasses this problem. There also is no need for tedious extraction methods to remove TBHQ from food oils. A simple agitation of the oil with acetonitrile, followed by slight concentration of an aliquot of the acetonitrile layer, are sufficient for acceptable TBHQ recoveries. Published methods for TBHQ analysis generally have not included the effect of time on analytical precision; this aspect has been included in our method.

[1]Presented at the JOCS/AOCS Annual Meeting, 1979, San Francisco.

METHODS

Gas Chromatographic Conditions

The column used was a 6 ft, 1/4 in. od glass column packed with 10% Versilube F-50 on 100-200 mesh Gas Chrom Q. The chromatograph was operated with a column temperature of 190 C, injection temperature 200 C, and flame ionization detector temperature 240 C. The carrier gas was nitrogen at a flow rate of 53 ml/min, and the hydrogen and air flow rates to the detector were set for optimal response for the specified chromatographic conditions. The injection port was glass-lined. The injected volume was 1.5 μl. The detector output was 5 mV, FSD with electrometer sensitivity of 5×10^{-12} amps.

Calibration

Separate acetonitrile stock solutions of TBHQ and the internal standard, propylparahydroxy benzoate (propyl paraben), were accurately prepared and diluted to give a final concentration of 102 μg/ml for TBHQ and 156 μg/ml for the propyl paraben. This allowed convenient combination of either 0.5, 1.0, or 1.5 ml of TBHQ standard solution to 1 ml of propyl paraben standard solution, thus approximating the concentration range of interest, 50-150 μg TBHQ, with the amount of internal standard being held constant at 156 μg in any combination. One ml of BSTFA was added to each solution and the combined solution was concentrated by evaporation under a nitrogen stream to ca. 1 ml just