Elek Science, London, 1973, p. 484.
4. Daghetta, A, and G. Amelotti, Riv. Ital. Sost. Grasse 45:735 (1968).
5. Olmedo, R.G., A. Carballido, and M. Borregon, An Bromatol. 19:67 (1967).
6. Lauber, E., Mitt. Geb. Lebenmittelunters. Hyg. 50:553 (1959).
7. Meduri, P., A. Rota, and N. Bino, Boll. Lab. Chim. Prov. 15:455 (1964).
8. Williams, K.A., "Oils, Fats and Fatty Foods," Fourth Edition, J.A. Churchill Ltd., London, 1966, p. 301.
9. Ewing, G.W., "Instrumental Methods of Chemical Analysis," Fourth Edition, McGraw-Hill, Kogakusha Ltd., Tokyo, 1975, pp. 34,35.

[Received April 11, 1979]

Spectrophotometric Studies of Rice Bran Oil and Mustard Oil Mixtures: II

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ABSTRACT

Visible spectra, between 400 to 500 nm range, of rice bran oil, mustard oil and their 7 mixtures, diluted 10 times in carbon tetrachloride, are reported. Mustard oil spectra shows three characteristic bands centered at wavelength 428, 453 and 482 nm, while no such band has been observed in rice bran oil spectra in this range. Intensity of 428 nm band increases as the rice bran oil percentage increases in the mixture of two oils. Five indices, R_1 to R_5, have been suggested for the approximate determination of rice bran oil adulteration in mustard oil. Plots of R_2 and R_3 against percentage of rice bran oil in the mixture have been found to be straight lines. The index R_3, equal to $1000(A_{428}-A_{482})$, has been found to be the most useful for this approximate estimation of rice bran oil in mustard oil.

INTRODUCTION

Rice bran oil is suspected to be one of the main adulterants of mustard oil. We have been engaged in the spectrophotometric study of the mixtures of rice bran and mustard oil with a view to have a quick method of its detection. It was reported earlier (1) that rice bran oil of high free fatty acid content (FFA) gives a C=O stretching band at 1710 cm^{-1}, not observed in mustard oil sample. This band emerges as a shoulder in the rice bran-mustard oil mixture, when rice bran oil content is 30% in the admixture. Experiments with neat oil samples as such in the cuvettes show that the presence of rice bran oil even at low concentration (5%) was sufficient to show a shoulder due to characteristic 564 nm rice bran oil band and their absorbance was found to be quite high between 400-500 nm region (2).

To understand the nature of spectra in this region it was thought desirable to extend these investigations using a suitable solvent. This paper reports the results of spectrophotometric study of mustard oil, rice bran oil and their mixtures using carbon tetrachloride as solvent.

EXPERIMENTAL

The samples of mustard (M) oil and rice bran (RB) oil were the same, used in earlier work, and their details have been already mentioned (2). Mixtures of mustard oil and rice bran oil were prepared in different ratios on volume basis at room temperature (20 C). All the samples were dissolved and diluted 10-fold with carbon tetrachloride. Pye Unicam SP8 – 100 UV Spectrophotometer was used to run the spectra of samples employing usual technique.

RESULTS AND DISCUSSION

Figure 1 shows visible spectra, between 400 to 500 nm, of mustard oil, rice bran oil and their mixtures. Since the oil samples are diluted in carbon tetrachloride, the intensity of various bands shown by neat oil samples (2), (500 to 760 nm) decreases to such an extent that the same almost merge in the main curve and so have not been shown in Figure 1.

Curves A and I show the spectra of mustard oil and rice

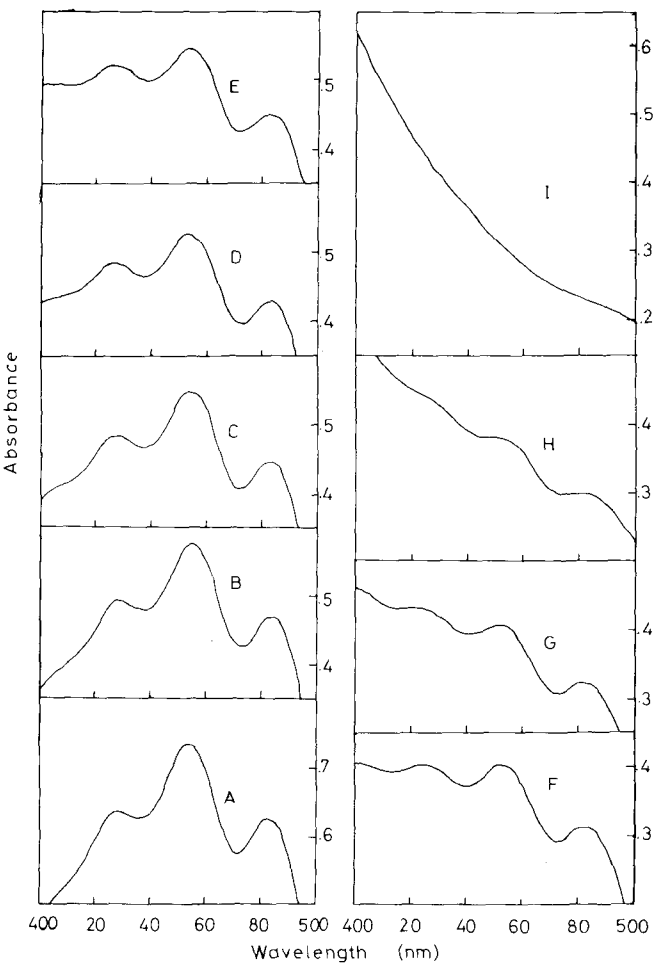

FIG. 1. Spectra of mustard oil, rice bran oil and their mixtures at 1/10th dilution in carbon tetrachloride. (A) 100M:0RB; (B) 90M:10RB; (C) 80M:20RB; (D) 70M:30RB; (E) 60M:40RB; (F) 50M:50RB; (G) 40M:60RB; (H) 20M:80RB; (I) OM:100RB; oil mixtures.

TABLE I

Some Indices of Rice Bran and Mustard Oil Mixtures

Sample	Ratio of RB and M oil	FFA content as oleic acid	R_1	R_2	R_3	R_4	R_5
A	0RB : 100 M	1.8%	0.868	96	9	3.183	1.09
B	10RB : 90 M	3.97%	0.855	83	22	3.270	1.26
C	20RB : 80 M	6.14%	0.877	67	37	3.318	1.55
D	30RB : 70 M	8.31%	0.918	43	55	3.357	2.27
E	40RB : 60 M	10.48%	0.953	24	72	3.375	4.0
F	50RB : 50 M	12.65	0.997	1	92	3.586	92.0
G	60RB : 40 M	14.82%	1.067	-27	110	3.603	—
H	80RB : 20 M	19.16%	1.143	-55	135	3.716	—
I	100RB : 0 M	23.5%	1.377	—	190	—	—

bran oil, respectively. The mustard oil spectrum shows three characteristic bands having centers at ca. 428,453 and 482 nm. The central band at 453 nm is of maximum intensity. Bands at 428 and 482 nm seem almost of equal intensity. However, the former is slightly greater. Since the apparent color of a solution is always the complement of the color absorbed, the 428, 453 and 482 nm bands seem to be in accordance with the yellow colored mustard oil which absorbs at these wavelengths of violet and blue region (3). The spectrum of rice bran oil does not show any band in this region. Its absorbance value continuously falls as wave-length increases.

It can be observed by the study of curves B to H that as the rice bran oil ratio increases in the mixture the intensity of 428 nm band also increases correspondingly and becomes wider. When the rice bran oil content is 60%, it appears as a shoulder, and at 80% rice bran oil content this band seems to be lost in the main curve. It is noteworthy that the 453 and 482 nm bands can be identified as broad shoulders even down to 20% mustard oil in the mixture.

A method, like one suggested by Meduri et al. (4), is being proposed here for the approximate estimation of the percentage of rice bran oil in admixtures with mustard oil, by taking absorbance reading at three wavelengths 428, 453 and 482 nm and determining a number of indices as given below.

$$R_1 = A_{428}/A_{453},\ R_2 = 1000\,(A_{453} - A_{428})$$

$$R_3 = 1000\,(A_{428} - A_{482}),\ R_4 = (A_{428} + A_{453} + A_{482})/A_{482}$$

$$R_5 = (A_{453} - A_{482})/(A_{453} - A_{428}).$$

The values of these indices for various mixtures of rice bran oil and mustard oil are tabulated in Table I.

It is clear from Table I that values of R_1, R_3, R_4 and R_5 increase as the rice bran oil percentage increases in the mixture. The value of R_2 decreases as the rice bran oil ratio increases and becomes negative when RB is more than 50%.

In order to test the applicability of the indices in Table I for the estimation of rice bran oil in mustard oil, the absorbance values of other admixtures were also measured in similar conditions; i.e., after diluting 10 times in CCl_4. It was observed that amounts of rice bran oil in mustard oil indicated by R_3 and R_4 generally agree quite well. In case the agreement is not good, then the consultation with other indices is found to be helpful. A plot of R_2 and R_3 against percentage of rice bran oil is shown in Figure 2. It is interesting to observe that both give straight lines with the difference that R_3 gives positive slope while R_2 gives negative slope. Testing a mixture of oils of unknown composition with the help of these plots suggested that R_3 is the most useful index for the approximate determination of rice bran oil in mustard oil. All these indices give dependable results up to 50% presence of rice bran oil in mustard oil.

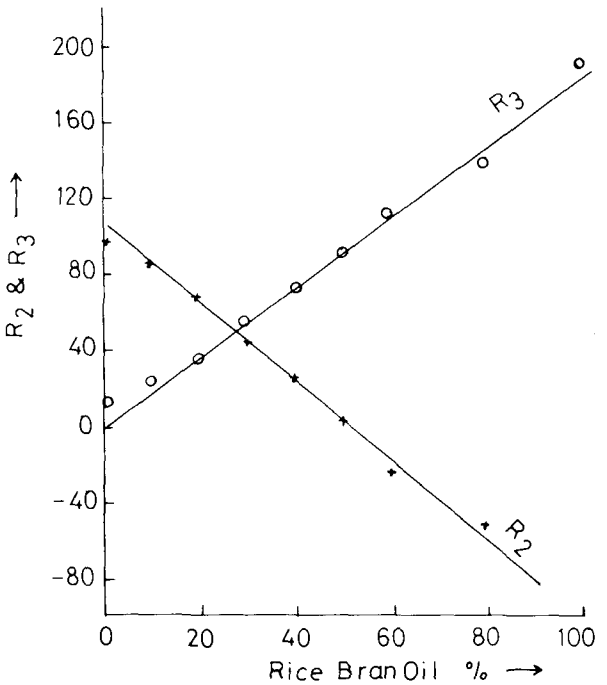

FIG. 2. Plot of indices R_2 and R_3 against percentage of rice bran oil in the rice bran-mustard oil mixture.

Since rice bran oil can have a high FFA content up to 80% as oleic acid (5), it is possible that, due to this difference of FFA contents from sample to sample, the absorbance of rice bran oil sample may be different, and thus it may have some effect on the values of these indices. However, the overall nature of these indices should remain the same. FFA content of all these samples were determined (Table I) to study the correlation of these indices with it, but no satisfactory relation could be established.

It is to be noted here that these indices suggest simply adulteration in mustard oil more confirmatively rather than the presence of rice bran oil, because in the range 400-500 nm no characteristic peak of rice bran oil is observed. The positive indication of rice bran oil's presence can be obtained by the 500-700 nm spectra of neat oil, as mentioned in earlier communication from this centre (2).

ACKNOWLEDGMENT

Dr. A.B. Shrestha, ex-Executive Director of the Research Centre, provided necessary lab facilities, and the present Director Dr. K.L. Shrestha granted permission to communicate this paper.

REFERENCES

1. Jha, J.S., J. Inst. Sc., Tribhuvan University (submitted).
2. Jha, J.S., JAOCS, resubmitted after revision.
3. Ewing, G.W., "Instrumental Methods of Chemical Analysis", McGraw-Hill Kogakusha Ltd., Tokyo, (Fourth Edition,) 1975, pp. 34-35.
4. Meduri, P., A. Rota, and N. Bino, Boll. Lab. Chim. Provinciali, 15:455 (1964).
5. Williams, K.A., "Oil, Fats and Fatty Foods," J.A. Churchill Ltd. London, (Fourth Edition) 1966 p. 258.

[Received October 4, 1979]

The Fatty Acid Composition of Seed Oils from Ten Plant Families with Particular Reference to Cyclopropene and Dihydrosterculic Acids

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ABSTRACT

Oil contents and fatty acid compositions of 40 seed oils of the plant families Elaeocarpaceae, Thymelaeceae, Malvaceae, Sterculiaceae (order Malvales); Anacardiaceae, Celestraceae, Sapindaceae (Sapindales); Ebenaceae, Sapotaceae (Ebenales) and Rhamnaceae (Rhamnales) are presented. Cyclopropene fatty acids (CPFA) occur in two families in the order Malvales not hitherto assayed. CPFA contents of seed oils of 12 Australian and Pacific species of Malvaceae and Sterculiaceae are given. CPFA occur randomly in small amounts in at least six families not in the order Malvales. Dehydrosterculic acid (DHS) occurs in small amounts in many species of Anacardiaceae, Celestraceae, Elaeocarpaceae, Malvaceae, Sapindaceae, Sapotaceae and Sterculiaceae. Linoleic acid was predominant in 28 of 40 seed oils, being as high as 63.9% in two species. The sum of 18:1 and 18:2 esters exceeded 70% in 20 oils.

INTRODUCTION

The occurrence of cyclopropene fatty acids (CPFA), malvalic and sterculic, in many species of the plant families Bombacaceae, Malvaceae, Sterculiaceae and Tiliaceae was summarized up to 1969 by Phelps et al. (1), Recourt et al. (2) and Christie (3). Subsequently, 30 other species in these families have been analyzed for CPFA (4-10).

Assays of seed oils for CPFA in other families in the order Malvales have not recently been reported. Analyses on some species of Thymelaeaceae and Elaeocarpaceae are reported in this paper as well as a number of Australian and New Caledonian species of Malvaceae and Sterculiaceae not hitherto studied. In their study of seed oils of species from 113 plant families, Earle and Jones (11) reported the occurrences of CPFA in plants not in the order Malvales. These included *Styrax americana* (order Ebenales, family Styracaceae), a *Randia* sp. (order Rubiales, family Rubiaceae) and *Astragalus brazoensis* (order Rosales, family Leguminosae). Assays were conducted on some seed oils in Ebenaceae and Sapotaceae (order Ebenales), in Anacardiaceae, Celastraceae, and Sapindaceae (order Sapindales) and in Rhamnaceae (Rhamnales).

CPFA were detected in the seed oils by the Halphen color test (12). The assays on 20 Halphen-positive and 20 Halphen-negative oils are reported.

The cyclopropane fatty acid, dihydrosterculic (DHS), frequently occurs in oils containing CPFA (8). It also occurs as a major component (41%) in *Litchi chinensis* seed oil (13) and comprises 17% in the oil of *Euphoria longan* (14), both species being in the family Sapindaceae. In the present investigations DHS was assayed in each oil.

This work is part of a continuing survey of seed oils from the Australian and South Pacific regions.

EXPERIMENTAL PROCEDURES

Material

Most seeds were collected by officers of the Royal Botanic Gardens, Sydney, H.S. McKee of New Caledonia and the author.

Treatment of Oils

After drying at room temperature, the seeds were disintegrated and extracted with cold hexane (b.p. 65-67 C) in an "Omni-Mixer." The methyl esters were prepared by the rapid method of Glass and Christopherson (15) using sodium methoxide. The esters were purified by dissolving them in hexane and placing up to 100 mg on a column comprising 12 g Florisil containing 7% water, and eluting with 40 ml hexane. The methyl esters were eluted with 70 ml diethyl ether in hexane (5% v/v).

Halphen Color Test on Oils

The AOAC method (12) was used, but, when the amount of oil available was less than 0.5 g, only 1-2 ml of reagent was used; less than 50 mg of two oils were available and dilutions of 1 in 20 and 35 were necessary. Dilution rather than large reduction of the reagent volume was chosen, because at least 1 ml of reagent was necessary to perceive the development of a faint pink color.

Argentation of Methyl Esters

Samples of esters from oils having a positive Halphen Color test were analyzed for cyclopropene and other acids by the method of Schneider et al. (16), which involves the reaction of silver nitrate in methanol with CPFA esters to form ether and ketone derivatives. The recovered esters and ether and ketone derivatives were submitted to analysis by gas chromatography.

When CPFA concentrations were less than ca. 3%, the derivatives were separated from the methyl esters by column chromatography with neutral alumina (16). The methyl ester fraction did not contain derivatives, but the derivative fraction usually contained small amounts of methyl esters. In the subsequent analysis of the reaction products by gas chromatography, a known weight of the methyl ester 20:0 was added as an internal standard.