Triglyceride Composition of *Amoora rohituka* Seed Oil

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ABSTRACT

The fatty acid composition of *Amoora rohituka* seed oil was determined by gas-liquid chromatography. The mole percentages of individual acids were found to be palmitic, 24.8; stearic, 12.4; oleic, 20.9, linoleic, 28.5; and linolenic, 13.4. Triglyceride composition was calculated from the fatty acid composition of the native oil and of the monoglyceride produced from it by pancreatic lipase hydrolysis. This calculation gave 2.2, 28.6, 48.1, and 21.1% GS_3, GS_2U, GSU_2, and GU_3, respectively.

INTRODUCTION

The plant *Amoora rohituka*, of the Meliaceae family, is a large evergreen tree of India with a straight cylindrical trunk 50 ft long and 5-6 in. in girth and is distributed over the sub-Himalayan tract of that country. According to earlier reports (1,2), the seeds of this plant (78% kernel) yield ca. 47% of a reddish brown oil constituted of 57.4% linoleic and 11.2% oleic acids. One of the seed oils of the Meliaceae family, *Melia azadirachta*, which has already gained considerable commercial importance in this country, is composed of 49.1-61.9% oleic acid and 9.0-15.8% linoleic acid (3-6). Such wide variations in the compositions of the seed oils from two species of the same family warranted further investigation of the former by modern techniques. Also, the seed oil of *A. rohituka* appeared to be of interest to us in our study of the triglyceride composition of linoleic rich oils (7,8).

The present knowledge of the triglyceride composition of linoleic rich oils is based mostly on the data obtained by the low temperature crystallization technique, now considered to be inadequate (9-11) for the purpose, and also by the rather tedious countercurrent distribution technique of Dutton and Scholfield et al. (12,13). The method of determination of triglyceride composition based on selective hydrolysis by pancreatic lipase (14,15) has not been extensively used in the investigation of linoleic rich oils.

This paper reports the findings on the triglyceride composition of the seed oil of *A. rohituka* determined by a combination of enzymatic hydrolysis, thin layer chromatography, and gas-liquid chromatography (GLC).

TABLE I

Characteristics of the Seed Oil and Mixed Fatty Acids (MFA) of *Amoora rohituka*

Characteristics	Oil	MFA
Percentage free fatty acids as oleic	0.4	-
Specific gravity	0.9	-
Saponification equivalent	291.2	274.8
Iodine value Wij's 60 min	103.2	107.5
Unsaponifiable (%)	5.8	-

EXPERIMENTAL PROCEDURES

Seeds of *A. rohituka* were procured from a local commercial supplier.

Lipolysis was carried out as suggested by Coleman (16) at pH 8.5 and at 37.5 C using a purified pork pancreatic lipase preparation with the addition of Ca^{2+} ions and bile salts. The partial glycerides were separated on a thin layer of silica by development with a solvent system of *n*-hexane, diethyl ether and acetic acid (80:20:0.25). The monoglycerides thus isolated and the original triglyceride sample were next converted into methyl esters, the former by the method developed by Luddy et al. (17) and the latter by the acid catalyzed esterification process. GLC of the methyl esters was carried out with an F and M analytical gas chromatograph (Model 700 R-12) equipped with a flame ionization detector. The column (6 ft x 1/4 in.), packed with 10% polyester of diethylene glycol adipate on 60-80 mesh Gas chrom Z, was operated at 166 C with a carrier gas flow of 40 ml/min. Peak areas were determined as the product of peak height and the width at half height. The weight percentages obtained were converted to mole percentages.

RESULTS

On extraction with petroleum ether (bp 60-80 C), seed kernels of *A. rohituka* yielded 35% of deep yellow colored oil (18.4% free fatty acid) which on refining became light yellow in color. On analysis by standard procedures, the refined oil and the mixed fatty acids obtained from it showed the characteristics given in Table I.

The fatty acid composition of the 2-monoglyceride obtained from lipolysis of the *A. rohituka* seed oil was determined by GLC analysis of its methyl esters. The results, along with fatty acid composition (mol %) of the triglyceride, are given in Table II.

The triglyceride composition of *A. rohituka* seed oil calculated from the fatty acid composition of the original triglyceride and the 2-monoglyceride using the assumptions of Vanderwal (18) and Coleman (16) was found to be GS_3 2.2, GS_2U 28.6, GSU_2 48.1, and GU_3 21.1 percents mole, respectively.

DISCUSSION

Fatty acid composition of the seed oil of *A. rohituka* as determined in the present investigation is shown in Table III, along with some of the findings reported earlier on the fatty acid compositions of the seed oils of the Meliaceae family. Present findings on the fatty acid composition of *A. rohituka* seed oil show wide deviations from that reported earlier by Ayyar and Patwardhan (1). This difference may be explained by climatic and geographic effects or by the greater accuracy of the results obtained by modern techniques in comparison to the hexabromide method followed

TABLE II

Fatty Acid Composition (mol %)

Sample	Fatty acids				
	$C_{16:0}$	$C_{18:0}$	$C_{18:1}$	$C_{18:2}$	$C_{18:3}$
Amoora rohituka triglyceride	24.8	12.4	20.9	28.5	13.4
2-Monoglyceride from native seed oil	7.7	-	44.5	35.4	12.4

TABLE III

Fatty Acid Compositions (% by wt) of Seed Fats of the *Meliaceae* Family[a]

Sample	Habitat	Component fatty acids					
		$C_{16:0}$	$C_{18:0}$	$C_{20:0}$	$C_{18:1}$	$C_{18:2}$	$C_{18:3}$
Azadirachta indica (3)[b]	India	14.9	14.4	1.3	61.9	7.5	-
Azadirachta indica (4)	India	16.2	14.6	3.4	56.6	9.0	-
Azadirachta indica (5)	India	15.9	17.7	2.1	52.9	10.5	-
Azadirachta indica (6)	India	20.1	19.2	-	44.4	16.3	-
Carapa procera (19)	W. Africa	31.3	5.0	0.9	49.3	11.9	0.4
Melia azedarach (20)	Argentina	8.1	1.2	0.6	20.8	67.7	-
Swietenia macrophylla (21,22)	India	12.5	16.4	0.6	25.3	33.9	11.9
Amoora rohituka (1)	India	7.5	15.1	-	11.2	57.4	7.8
Amoora rohituka (present work)	India	23.1	12.8	-	21.5	29.0	13.6

[a]Minor components are not shown.

[b]Numbers in parentheses are reference numbers.

by previous workers. The fatty acid composition of *A. rohituka* seed oil seems to be similar to that of the seed oil of *Swietenia macrophylla,* as reported by Chakraborty and Chowdhury (22) based on the spectrophotometric and low temperature crystallization methods. In the seed oil of *A. rohituka,* the saturated acids constitute ca. 1/3 of the total fatty acids, with palmitic acid being present in twice the proportion of stearic acid. However, in the seed oils of *Swietenia macrophylla* and also of *Azadirachta indica,* palmitic and stearic acids are present in about equal proportions, though these two acids together make up some 30% of the total fatty acids. The seed oil of *A. rohituka* also differs considerably from that of *Azadirachta indica* in the content of C_{18} unsaturated acids. The latter is rich in oleic acid (44-62%) and low in linoleic acid (7-16%), while the former contains both the acids within a narrow range (22-29%). Moreover, the seed oil of *A. rohituka* contains 13.6% linolenic acid, and, as such, it shows a general resemblance to the oils from the seeds of citrus fruits which are already being considered of commercial importance.

The mole percents of different categories of component glycerides present in the seed oil of *A. rohituka* are similar to those calculated by Gunstone (23) for the lime seed oil with comparable fatty acid composition (24). The triglyceride compositions of the two samples are GS_3 2.2,–; GS_2U 28.6, 31; GSU_2 48.1, 49; and GU_3 21.1, 20 percents mole, respectively. However, the detailed composition of the component glycerides of *A. rohituka* seed oil shows considerable deviation from that of lime seed oil determined by Hilditch and coworkers (24) using low temperature crystallization technique: disaturated oleins 12.7, 25; disaturated linoleins 11.3, –; saturated oleolinoleins 15.4, 30; and saturated linoleolinolenins 8.8, 23 percents mole, respectively.

According to the theory of Gunstone, the 2-position of the glycerol moiety is preferentially acylated by C_{18} unsaturated acids, and there is a preference for linoleic acid over oleic and linolenic acids for the 2-position (23,25). It will be evident from Table II that the present findings agree fairly closely though not completely with the hypothesis so far as the acylation of the 2-position by C_{18} unsaturated acids is concerned. The monoglyceride of the seed oil of *A. rohituka* as obtained by lipolysis is composed of 92.3% of C_{18} unsaturated acids, but a deviation is observed regarding the selectivity of linoleic and oleic acids for the 2-position. In the present instance, at least, oleic acid shows preference over linoleic acid for the 2-position. Whether this has to do with the presence of all three C_{18} unsaturated acids in considerable proportions within a narrow margin (15-30%) needs further study on the seed oils of similar composition for confirmation.

The seed oil of *A. rohituka,* like citrus seed oils, also shows promise to be a good substitute for cottonseed oil, except for increased liability to oxidative rancidity. *A. rohituka* seed oil, after hydrogenation until linolenoyl glycerides have disappeared, would appear to be practically equivalent in composition to cottonseed oil hydrogenated to a similar iodine value and as such may be used in the soap industry. On the other hand, this oil may also be used for the preparation of drying oil by solvent segregation. The availability of the seeds and its oil content lend further possibility to commercial exploitation of this seed oil.

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