ponent, was used in deriving the analytical data reported here; corrections were not required for oleate and linoleate under the outlined experimental conditions (see Analytical Methods). Studies with model mixtures of pure esters suggest that calculations based on these procedures are more accurate than those determined by relating the area of each component to the total area obtained from all components in a GLC-chromatogram.

The studies reported here revealed that the lipid depressant activities exhibited by four marine oils could be duplicated by ingestion of the whole fish products which contain these oils. In hypercholesterolemic rats, both the fish and fish oils promoted a more favorable balance between the cholesterol and phospholipid components found in blood and liver (i.e., a lower TC/TP). Observations of Mead and Gauze (17) suggest that a low TC/TP value is probably one of the best indexes of a normal distribution of lipids in the blood vascular tissues.

Although Wood and his coworkers (18) have reported that unsaponifiables from certain fish liver oils are effective cholesterol depressants in chickens, it would seem unlikely that the small amounts of marine sterols found in these supplements (Table I and Fig. 1) were responsible for the lipid depressant activities of the four species of fish. The diets contained between 5 and 20 times more cholesterol than the marine sterols; the bile acid components of the experimental diets would also be expected to mask the effects of the marine sterols found in the fish supplements. Furthermore, our other studies (3,4) and those of De Groot and Reed (7) have shown that the unsaponifiables from the whole body oils of tuna and menhaden, and the liver oil from cod, have little influence on the circulating lipids of hypercholesterolemic rats.

Results from these studies demonstrate that it is possible to bring about a significant reduction in blood and tissue lipids of hypercholesterolemic rats by very small dietary changes. The oil supplements from ocean perch represented only 5.5% of the total caloric contents of the diets (¼ of the dietary fat); yet this small alteration in the chemical composition of the diet was enough to promote nearly a 50% reduction in the combined cholesterol and phospholipid components of the blood (Table I). It is not apparent, however, why doubling the intake of a similar oil (menhaden) failed to promote a greater reduction in blood lipids. The lipid depressant activities of the marine oils may be a reflection of a combination of factors including their total unsaturation (1,2), the type of unsaturation found in their fatty acid components, and the different proportions of specific fatty acids found in the marine oils.

The apparent differences in the effects of the whole fish and their component oils on liver lipids may be related to the lipotropic activities of the fish proteins. Other investigators (8) have observed that dietary protein concentrations, and the amino acid components of proteins, can influence both liver and blood lipids under similar experimental conditions.

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Hexane and Ethanol as Peanut Oil Solvents

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Abstract

Absolute ethanol is a better solvent for extracting peanut grits than 95% ethanol, with hexane intermediate in its action. More nonlipids solids are extracted by 95% ethanol than absolute ethanol as compared with none by hexane. Ethanol-extracted oils are slightly higher in color and free fatty acids than hexane-extracted oils.

PREVIOUS WORK in this laboratory has shown that peanut flakes disintegrate upon contact with a solvent, producing powder difficult to handle in a continuous extractor. Extraction rates were therefore determined on two sizes of peanut grits using three solvents: hexane, 95% ethanol, and absolute ethanol. While ethanol, as pointed out by Rao and Arnold (3), is not practical for general extraction use in this country, it has advantages for use in countries such as India where many peanuts are grown. Since preliminary examination of early experimental results indicated that the ethanols were apparently extracting more non-glyceride material than hexane, determinations were made of the amounts of solid non-glyceride material and upon the purity of the oil extracted.

Two sizes of grits were used: through 14-mesh and retained on 20-mesh, and through 20-mesh and retained on 30-mesh. The extractions were carried out in glassware rate extraction apparatus similar to that used in previous studies in this laboratory (1) except somewhat larger. The extraction chamber was 12 inch high by 2 inch diam. Both extraction chamber and solvent were heated to the desired temperature

by water from a constant temperature source. Total oil extracted was determined at the end of 10-min. intervals but only the results for 60-min. extractions are shown (Table I). Two other series in which the grits were preheated did not produce significantly different data and are not included.

As would be expected greater extraction was secured with a decrease in grit size and an increase in temperature. Absolute ethanol was a better solvent than 95% ethanol. This slower extraction probably resulted in part from the relatively low solubility of the oil. Rao, *et al.* (4), have shown that while peanut oil is completely miscible at 72C in absolute alcohol its solubility is less than 20% in 95% ethanol at this temperature. Solubility in 95% ethanol at 72C, and absolute alcohol at 43C, are not greatly different. At the higher temperature the coagulating effect on the protein of the absolute alcohol may also be a factor.

TABLE I
Residual Extractables in Solvent Extracted Peanut Grits
(Extraction time: 60 min)

Solvent	Temperature °C	Residual extractables, % of total	
		Grit size	
		+14 −20	+20 −30
Hexane	26	35.2	16.3
	43	31.0	14.6
95% Ethanol	43	77.5	63.8
	72	34.4	13.0
Absolute ethanol	43	31.0	10.8
	72	16.5	1.3

TABLE II
Peanut Oil Quality *

Sample No.	Mesh size	Extraction Solvent temp °C	Neutral oil, %	Phospholipids, %	Free fatty acid, %	Iodine value	Color	Sap. value
1	−14 +20	Hexane at 44C	91.8	6.8	0.6	84.9	1.58	192.0
2	−20 +30	Hexane at 44C	92.7	5.9	0.8	84.7	1.67	194.0
3	−14 +20	95% Alcohol at 72C	92.4	6.5	1.7	82.5	2.92	192.0
4	−20 +30	95% Alcohol at 72C	94.0	5.5	1.3	87.8	2.53	190.5
5	−14 +20	Abs. alcohol at 72C	94.9	4.9	1.4	83.9	2.41	193.8
6	−20 +30	Abs. alcohol at 72C	94.5	4.8	1.4	84.9	3.41	191.8
7	−14 +20	Hydraulic pressed	92.4	6.5	1.0	86.3	0.33	193.1
8	−14 +20	Abs. alcohol at 72C followed by ether	92.0	6.9	1.2	84.8	2.50	192.1

* Neutral oil and phospholipids were determined by the method of Choudhury and Arnold (2). Other determinations by standard A.O.C.S. Methods. + Extraction time 2 hr. Rate, 10 ml per min.

It was noted that part of the extracted material was a solid. This was separated by filtering and extracting with ethyl ether to remove the lipids. It was found that the solid non-lipids extracted by the 95% ethanol at 72C amounted on the average to 17.4% of the total extracted material and 20.9% of the lipids extracted. The corresponding values for the non-lipids extracted by absolute alcohol were 7.2% and 7.7%, respectively. The protein content of the solids extracted by the 95% ethanol averaged 17.6% and those extracted by the absolute ethanol 6.9%. Coagulation of more protein by the absolute alcohol probably reduced the amount extracted.

The oil, filtered free of solids, was evaluated for quality. See Table II. The hexane oils are lighter in color and have a lower FFA content. The oil extracted with absolute alcohol has a slightly lower phospholipid content than the other oils.

In summary, these studies indicate the following general conclusions: A) absolute ethanol is definitely a better solvent for extracting peanut oil from grits than 95% ethanol; B) at 43C hexane is superior to 95% ethanol and slightly inferior to absolute ethanol as a solvent; C) more non-lipid solids are extracted at 73C by 95% ethanol than by absolute ethanol; and D) ethanol-extracted oils are slightly higher in color and FFA than hexane-extracted oils.

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Analyses of Lipids and Oxidation Products by Partition Chromatography: Hydroxy Fatty Acids and Esters[1]

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Abstract

A liquid-partition chromatographic procedure was used to separate hydroxy fatty acids, their methyl esters, and reduced fatty ester hydroperoxides. Mixtures of methyl stearate, mono- and dihydroxystearate, and mixtures of the corresponding free fatty acids were easily separated. Chromatographic determinations for ricinoleate in castor oils compared favorably with the chemical and infrared analyses.

The chromatographic procedure was used to separate hydroxy fatty acids in *Dimorphotheca* and *Strophanthus* seed oils. The methyl ester of dimorphecolic acid, the principal hydroxy fatty ester of *Dimorphotheca* oil, behaved like reduced methyl linoleate hydroperoxide and showed a polarity intermediate between methyl 12-hydroxystearate and methyl 9,10-dihydroxystearate. The 9-hydroxy-12-octadecenoic ester of *Strophanthus* oil had a larger retention volume than methyl

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