Endogenous lipid composition of the intestinal lymph of rats raised on fat-free, lard, or corn oil diets

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SUMMARY Studies are reported on the structure of triglycerides from the intestinal lymph of fasted rats grown from weaning to 4 months of age on a fat-free diet or on diets containing lard or corn oil. Experiments are also reported on the incorporation of dietary palmitic-1-C^{14}, oleic-1-C^{14}, and linolenic acids and of tripentadecanoin into the triglycerides of the intestinal lymph of animals in the above groups. These studies show that the distribution of the fatty acids among the triglycerides synthesized in the intestinal mucosa does not conform to a random pattern.

The composition of endogenous lipid (determined by the prolonged feeding period) was found to influence both the relative extent of incorporation of fatty acids into triglycerides or phospholipids, and also the resynthesis of different fatty acids into triglycerides.

Linoleic acid was predominantly in the β-position of triglycerides of which it was a constituent, and palmitic acid was predominantly in the β-position in triglycerides containing fatty acids other than linoleic acid.

KEY WORDS lipids · intestinal lymph · rat · fatty acid composition · phospholipids · triglyceride synthesis · fatty acid distribution

Concepts of the processes of digestion and adsorption of triglycerides in mammals have recently been well elaborated (1–5). Briefly, ingested triglycerides are hydrolyzed in the small intestine by pancreatic lipase, primarily to fatty acids and β-monoglycerides. Some α-monoglycerides are produced, together with some glycerol and free fatty acids, including those from the original β-position. These compounds are resynthesized in the intestinal mucosa into triglycerides which appear in the lymph.

Resynthesis is believed to take place by two major pathways, namely by reacylation of β-monoglycerides (6–10) and via the phosphatidic acid system (7, 11–15). The fate of the absorbed α-monoglycerides is not so clear, but it is believed that these compounds are split by specific enzymes in the intestinal wall. The fatty acids thus liberated are also resynthesized into triglycerides (10, 16–18).

Quantitative aspects of the process have been studied extensively by Mattson and Volpenhein (1, 2) by isotopic techniques. Their data indicated that in the rat approximately 72% of the glycerol molecules contained in dietary triglycerides are absorbed as β-monoglyceride. An additional 6% was estimated to be adsorbed as α-monoglyceride. The α-monoglycerides probably contain largely those acids from the original β-position, as a result of acyl migration. Approximately 22% of the dietary triglycerides are completely hydrolyzed. The resynthesis of the fatty acids into triglycerides via either pathway was believed to follow a random pattern, although there was some evidence for a slight preferential incorporation of palmitic acid into the β-position. The distribution of the fatty acids among the newly synthesized triglycerides appearing in the lymph has therefore been referred to as a "partially random" pattern (19).

Although Mattson and Volpenhein (2) recognized the presence of endogenous lipid in the thoracic lymph, they made no allowance for the incorporation of endogenous fatty acids into newly synthesized triglycerides. Gottenbos and Thomasson (20) considered the possibility that the amount and composition of endogenous triglycerides of thoracic lymph was independent of the dietary fat. However, in experiments on thoracic duct cannulated animals, Karmen and co-workers (21–23) found that the composition of the endogenous lipid had a profound influence on the composition of the lymph lipid; in some

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experiments it comprised approximately 40% of the total lipid. It was further shown that some fatty acids were preferentially incorporated into cholesterol esters. These workers also found a small but significant preferential incorporation of linoleic acid into the β-position of the triglycerides of thoracic lymph.

The experiments reported here indicate that there is a selectivity governing the positional arrangement of the fatty acids in the synthesis of triglycerides of intestinal lymph, so that the distribution of fatty acids among the triglycerides of intestinal lymph does not conform to a random pattern. Moreover, the composition of the endogenous fatty acids of the lymph markedly influences the distribution of newly absorbed fatty acids among the triglyceride molecules and between the phospholipids and triglycerides. The latter observation is in accordance with the findings of previous studies (21–24).

MATERIALS

Highly purified (>99%) cholesteryl oleate, triolein, oleic acid, cholesterol, linolenic acid, and tripentadecanoin and purified hydrogenated egg lecithin were obtained from The Hormel Institute, Austin, Minn.

Corn oil and lard used in the diets were commercial samples, the composition of which is reported elsewhere (25).

Palmitic acid-1-C^{14} and oleic acid-1-C^{14} were purchased from Tracerlabs, Inc., Richmond, Calif., and purified by thin-layer chromatography (TLC) on chromatoplates coated with silicic acid containing 10% calcium sulfate. The final preparations were >99% pure, chemically and radiochemically, and had specific activities of 8.5×10^3 dpm/mg and 9.5×10^3 dpm/mg respectively.

METHODS

Diets

Three groups of male rats of the Sprague-Dawley strain were raised from weaning to about 4 months of age on a fat-free diet or on the same fat-free diet supplemented with corn oil or lard. The fat-free diet consisted of vitamin test casein 16%, sucrose 74%, α-cellulose 4%, Wesson salt mixture 4%, 1% of a mixture of vitamins in casein, and 1% of casein containing choline chloride (26). Lard or corn oil was substituted isocalorically for an amount of sucrose equal to 10% (by weight) of the diet. The fat-free group received only the basal diet. The weight of the animals on the fat-free diet reached a plateau of 220–240 g at about 4 months of age. Up to this time there was little difference in the rate of growth of the animals of the three groups, and all animals selected for use in the present experiments were of similar weight.

Collection of Lymph and Plasma

The intestinal lymphatics of the rats were cannulated as described by Bollman, Cain, and Grindlay (27). While the lymph was being collected, a 0.9% (w/v) saline solution was administered at about 1 ml/hr through a small plastic tube inserted through a small incision in the abdominal wall into the fundus region of the stomach. Two or three animals from which about 10 ml of lymph could be collected per 12 hr period were selected for use in each experiment. Each animal selected for an experiment was fasted for about 24 hr prior to the operation. The lymph collected during the first 24 hr, while the animals were recuperating from the effects of the operation and adjusting to their confined environment, was discarded.

Test substances were introduced into the stomach in the form of an emulsion with a small amount of saline solution and bile salts. The lipid of the lymph (usually 15–20 ml) was recovered by three extractions with a total of about 200 ml of chloroform–methanol 2:1 (v/v). The combined extracts were filtered and evaporated to dryness, and the residue was reextracted with dry chloroform and filtered. The lipid was then recovered by evaporation of the solvent, weighed, and dissolved to give a measured volume of solution for lipid class analysis.

Animals were exsanguinated via the aorta under light ether anesthesia. A small amount of sodium heparin was added to the blood to prevent coagulation and it was centrifuged immediately. The plasma from each animal was extracted three times with 100 ml of ethanol–ether 2:1 (v/v) and the lipid was recovered by evaporation of the solvent and reextraction with chloroform as described for lymph lipids.

Lipid Analysis

Quantitative analysis of the lipid classes was carried out by TLC on silicic acid coated chromatoplates with the solvent system petroleum ether–ethyl ether–acetic acid 85:15:1 (v/v) as previously described (28).

Transmethylation of triglycerides and polar lipids was carried out on 1–10-mg samples, each of which was sealed under nitrogen in a glass ampule with 2–3 ml of dry 5% methanolic HCl and heated at 100° for 1 hr.

Methyl esters were subjected to gas–liquid chromatography (GLC) in an F & M flame ionization instrument equipped with a $^1/_4$ inch $\times$ 8 ft column packed with 8% (w/w) ethylene glycol succinate polymer (EGSS-X, Applied Science Laboratories, Inc., State College, Pa.) on 100–120 mesh Gas Chrom P at 175° and operated with a carrier gas flow of 60 ml/min. Percentages of fatty acids were calculated from peak areas corrected by the relative weight per cent of active carbon atoms in the molecule (29). Quantitative results with

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TABLE 1 Lipid Class Composition of Intestinal Lymph and Blood Plasma after a 24-Hr Fasting Period

	Lymph			Plasma
	Corn Oil Diet	Lard Diet	Fat-Free Diet	Fat-Free Diet
	Weight %			
Cholesterol esters	9.1	6.7	6.0	29.2
Triglycerides	52.6	65.4	61.6	14.2
Free fatty acids	tr.	1.2	1.5	1.4
Cholesterol	3.0	3.2	6.9	8.6
Polar lipids (mostly phospholipids)	35.3	23.5	24.0	46.6

standard mixtures of long-chain methyl esters (16:0, 18:0, 18:1, 18:2, and 18:3) agreed with the known composition with a relative error less than ±2%.

Triglyceride structure was determined by a combination of TLC and lipase hydrolysis (25). The triglycerides were separated on chromatoplates coated with silicic acid impregnated with silver nitrate. An aliquot of each of the recovered triglycerides was methanolyzed, and the methyl esters were subjected to gas–liquid chromatography in the presence of an internal standard. The positional arrangement of the fatty acids in the components of each fraction was determined by pancreatic lipase hydrolysis.

Lipids were assayed for radioactivity by the technique of Snyder and Stephens (30) using a Packard Tri-Carb scintillation counter.

RESULTS AND DISCUSSION

Composition of Intestinal Lymph

The analyses of the lipid classes of the intestinal lymph of all groups and of the plasma of the fat-free groups of animals are presented in Table 1.

The relative error of the method is less than ±5% (28) and all values are the average of triplicate analyses. Although a comparison was made only on the fat-free group, it is probably true to say that in all groups there was little similarity between the lipid class compositions of lymph and plasma. The lipid class composition of the lymph of the fasted rat is also unlike that of depot fat, which consists almost entirely of triglycerides. Since lymph is generally considered only in relation to its role in triglyceride metabolism, it is noteworthy that it may contain as much as 35% phospholipid (Table 1).

The fatty acid composition of the diet influenced the lipid class composition of the lymph, as is also shown in Table 1. The endogenous lymph lipid of the corn oil group contained a higher percentage of phospholipid than did that of the groups on the lard and fat-free diets. This was apparently at the expense of triglyceride, indicating a greater synthesis of phospholipids when the diet was high in polyunsaturated fatty acids.

Table 2 shows the marked influence of diet on the fatty acid composition of the intestinal lymph, which is in accordance with its well-known influence on the fatty acid composition of the depot fats of animals. The values in Table 2 are the averages of duplicate analyses.

The high amount of 20:3 and low amount of 20:4 acids in the fat-free group (Table 2) is typical of other tissues of animals with an essential fatty acid deficiency. Common to all groups and apparently characteristic of lymph, compared to other tissues, is the unusually high polyunsaturated fatty acid content of the triglycerides (on any but a fat-free diet). Since the animals were fasting when the observations were made, the fatty acid composition of the intestinal lymph in these experiments represents that of the endogenous pool of fatty acids which is normally employed in the resynthesis of dietary fatty acids into triglycerides in the intestinal mucosa.

TABLE 2 Fatty Acid Compositions of Fractions of Dietary Fat and of the Corresponding Intestinal Lymph and Plasma Lipids of Rats

Fatty Acid	Dietary Fat		Intestinal Lymph						Plasma	
	Corn Oil	Lard	Corn Oil Diet		Lard Diet		Fat-Free Diet		Fat-Free Diet	
	TG	TG	PL	TG	PL	TG	PL	TG	PL	TG
	Weight %									
14:0	tr.	1.4	0.4	1.0	0.3	0.7	0.6	1.5	0.5	2.0
15:0	—	—	0.3	0.7	0.2	0.5	0.4	0.7	0.2	0.6
16:0	11.6	23.4	22.5	21.6	23.7	26.7	22.4	26.2	23.7	27.9
16:1	tr.	3.2	2.4	4.4	2.2	3.8	8.7	11.4	7.6	15.2
17:0	—	—	0.5	0.5	tr.	0.4	0.6	0.8	0.4	1.7
18:0	2.3	11.7	16.6	9.4	20.6	9.5	14.7	7.3	16.3	2.2
18:1	28.7	48.7	11.2	22.3	13.5	33.1	29.9	43.8	28.3	49.1
18:2	57.4	11.6	21.8	27.4	16.2	12.4	2.9	1.5	1.5	1.3
20:1	—	—	1.1	3.5	1.0	5.4	1.3	1.4	0.6	—
20:3	—	—	tr.	tr.	tr.	tr.	13.3	5.4	19.2	—
20:4	—	—	23.2	9.2	22.3	7.5	5.2	tr.	1.7	—

TG = Triglycerides. PL = Polar lipids (mostly phospholipids).

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Triglyceride composition analyses of the lymph of the animals in each group are summarized in Table 3. The same classes of triglycerides occurred in all groups and the main influence of the different dietary fats was on the relative amounts of each class. The disparity between the found and calculated values indicates that the distribution of the fatty acids among the triglyceride classes does not follow a random pattern. Examination of the distribution of the fatty acids in the β-position of the triglyceride molecules of selected classes of triglycerides (Table 4) showed more clearly the divergence from a random distribution pattern and that there appeared to be a preference of some fatty acids for the β- or α-positions in the molecule. If a random pattern prevailed, the value for a particular fatty acid in the total triglyceride would be the same as that in the β-monoglyceride. How-

TABLE 3 TRIGLYCERIDE STRUCTURE OF INTESTINAL LYMPH AND PLASMA

Triglyceride Type ($\alpha\beta\alpha$)	Lymph						Plasma	
	Corn Oil Diet		Lard Diet		Fat-Free Diet		Fat-Free Diet	
	Found	Random	Found	Random	Found	Random	Found	Random
	Weight %							
SSS*	5.7	5.4	5.2	6.9	5.7	4.4	1.7	3.8
SSM	7.9	6.8	14.6	13.4	15.5	14.3 }	17.9	21.6
SMS	2.2	3.4	4.6	6.6	9.6	7.1 }		
SMM	3.4	4.3	12.4	12.8	24.9	23.3 }	53.9	41.6
MSM	3.3	2.2	8.9	6.4	9.5	11.7 }		
SSD	7.0	8.9	2.7	4.2	0.6	0.4 }	2.1	0.4
SDS	5.8	4.4	3.1	2.1	1.4	0.2 }		
MMM	2.0	1.4	6.4	6.2	16.8	19.1	23.1	26.6
SMD	3.8	5.6	3.1	4.0 }				
SDM	9.4	5.6	7.3	4.0 }	2.3	1.9	1.3	1.7
DSM	3.6	5.6	3.2	4.0 }				
MMD	1.9	3.5	2.1	3.9 }	0.8 }	1.6 }	tr. }	
MDM	3.8	1.8	3.8	1.9 }				4.3
Others	40.2†	41.1	22.6	23.5	12.9	16.0		

* S = saturated, M = monoenoic, D = dienoic and T = tetraenoic fatty acids.
† This 40.2% was analyzed for component triglycerides in a separate analysis as follows:

	Found	Random
SD_2	11.3	10.8
MD_2	6.4	6.8
S_2T	4.6	3.2
D_3	2.5	2.9
SMT	4.3	4.0
M_2T	1.4	1.3
SDT	6.6	5.2
Others	3.1	6.9

TABLE 4 FATTY ACID COMPOSITION OF LYMPH TRIGLYCERIDES AND OF CORRESPONDING β-MONOGLYCERIDES IN ANIMALS OF THE CORN OIL GROUP

Acid	Triglyceride Class											
	S_3		S_2M		SM_2		$S_2D + M_3$		SMD		M_2D	
	Total*	β-MG†	Total	β-MG	Total	β-MG	Total	β-MG	Total	β-MG	Total	β-MG
	Weight %											
14:0	2.0	2.7	1.6	2.2	0.8	1.3	1.1	1.2	0.5	0.5	—	—
16:0	75.3	92.1	51.7	72.4	26.3	45.8	43.4	46.6	23.2	20.4	—	—
16:1	—	—	3.2	3.7	4.1	5.5	1.3	1.8	2.8	2.4	4.4	3.1
18:0	22.7	5.2	13.3	3.6	6.8	1.9	13.8	1.8	8.0	0.3	—	—
18:1	—	—	26.8	17.2	53.9	45.5	9.3	6.6	27.6	20.4	52.3	29.5
18:2	—	—	—	—	—	—	29.9	42.0	34.3	56.0	36.7	67.4
20:1	—	—	3.4	0.9	8.1	tr.	1.2	—	3.6	—	6.6	—

* Total fatty acids of the triglyceride.
† Fatty acids of the β-monoglyceride (after lipase splitting of the α-acid).
S = Saturated fatty acids. M = Monoenoic fatty acids. D = Dienoic fatty acids.

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ever, Table 4 shows that palmitic acid is concentrated in the β-position in the fully saturated triglycerides and those containing monoenes as their only unsaturated constituent, and linoleic acid, when present, is preferentially in the β-position. Conversely, oleic acid and especially stearic acid are distributed predominantly in the α-position of the triglycerides of intestinal lymph. Determination of the distribution of the fatty acids in the polyunsaturated triglycerides of the lymph of the corn oil group ("Others", Table 3) in a separate analysis by lipase showed that 46.7 and 49.9% of the eicosatrienoic and arachidonic acids, respectively, were esterified in the β-position (the values for random distribution would be 33.3%).

Incorporation of Dietary Fatty Acids into Lymph Triglycerides

Experiments were carried out on additional animals of the fat-free and corn oil groups in which the lymph was collected for 24 hr after the administration of radioactive fatty acids. In these experiments the lymph triglycerides were isolated from each of these animals and the results were averaged. The specific activities of the fatty acids of the triglyceride fraction and of the β-monoglyceride fraction derived therefrom by the action of pancreatic lipase were determined. By comparison of these specific activities the relative amount of administered (as opposed to endogenous) radioactive fatty acid in the β-position of the lymph triglycerides may be calculated, as demonstrated by Mattson and Volpenhein (1, 2).

In the first of these experiments 100 mg of oleic acid-1-C^{14} was administered to animals of the fat-free group. Analysis of the lymph triglycerides showed that 32.9% of the oleic acid incorporated was in the β-position. Administration of the same amount of this acid to animals of the corn oil group resulted in the esterification of 29.6% in the β-position of the intestinal lymph triglycerides. Since the proportion of triglycerides containing linoleic acid is higher in the animals raised on the corn oil diet than in those raised on the fat-free diet, linoleic acid apparently takes precedence over oleic acid for the β-position in the biosynthesis of triglycerides.

In another experiment 100 mg of palmitic acid-1-C^{14} was administered to animals of the fat-free group. In this experiment 39.6% of the palmitic acid was found in the β-position of the lymph triglycerides of these animals, indicating a preference of palmitic over oleic acid for the β-position, because the endogenous pool of fatty acids of the animals in this group contained considerable oleic acid (but little linoleic). Thus, these experiments, as well as those on lymph composition on fasting rats, indicated there was a preferential esterification in the β-position in the order: linoleic > palmitic > oleic acids.

This type of experiment may be used with a wide variety of fatty acids and animals raised on various diets to obtain more general information. The ratio of fatty acids of dietary origin to endogenous fatty acids in the lymph in these experiments was relatively low, about 1:9, as determined by the decrease in specific activity of the recovered samples. In order to obtain meaningful results on the influence of endogenous fat on the mode of the incorporation of dietary fatty acids into triglycerides, the amount of dietary fatty acid fed should be low, especially when a single fatty acid is studied. Otherwise, for instance when a single fatty acid is administered in one large dose, it may overwhelm the endogenous fatty acids appearing in lymph and the lymph triglycerides may appear to be of random structure.

Incorporation of Dietary Fatty Acids into Triglycerides and Phospholipids

The intestinal lymphatics of several additional animals of the fat-free and corn oil groups of animals were cannulated. After an observation period of 24 hr, two animals of each group that provided a satisfactory flow of lymph were given by intubation 216 mg each of a 1:2 molar mixture of palmitic acid-1-C^{14} and unlabeled linolenic acid. Linolenic acid was selected for these experiments because it is not a normal constituent of the tissues of the animals raised on the diets used here, and therefore can be simply detected by GLC. The distribution of the 16:0 between the triglycerides and phospholipids was determined by the radioassay technique of Snyder and Stephens (30). The distribution of 18:3 between the triglycerides and phospholipids was determined by GLC of the methyl esters of these fractions separated by TLC. In this analysis the separated fractions were scraped directly from the chromatoplates into vials for methanolysis.

The palmitic and linolenic acids were esterified predominantly in the triglycerides in the animals from both the fat-free and the corn oil groups. In the two animals of

TABLE 5 Incorporation of Palmitic Acid-1-C^{14} and Linolenic Acid into the Triglycerides and Phospholipids of Intestinal Lymph

	Exogenous Fatty Acids	Fat-Free		Corn Oil	
		Rat 1	Rat 2	Rat 1	Rat 2
Triglyceride	18:3/16:0	3.45	2.72	4.56	5.24
Phospholipid	18:3/16:0	9.10	5.90	4.75	5.53
$\frac{18:3}{16:0}$ PL / $\frac{18:3}{16:0}$ TG		2.64*	2.18	1.05	1.06

* A ratio of 1 would indicate an equal degree of incorporation of palmitic and linolenic acid into triglycerides and phospholipids.

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the fat-free group the percentages of palmitic and linolenic acids in the triglycerides averaged 86 and 82% respectively and in the two animals of the corn oil group, 80 and 83% respectively.

In the corn oil group, the ratio of the amount of incorporated 18:3 to 16:0 was about the same in the phospholipids as in the triglycerides (Table 5), but in the fat-free group relatively more 18:3 was incorporated into phospholipids. That the composition of the endogenous lipid influences biosynthesis of triglycerides and phospholipids is thus further confirmed.

Relation of Exogenous and Endogenous Fatty Acids in the Synthesis of Lymph Triglycerides

In this experiment 200 mg of tripentadecanoin was administered to other cannulated animals of the corn oil group. Analyses of the lipids were carried out mainly to demonstrate a mixing of fatty acids of exogenous origin with those of the endogenous pool in the synthesis of triglycerides. For this purpose the triglycerides were fractionated by silver nitrate TLC (23) into two groups, those consisting of fully saturated fatty acids, and those containing at least one unsaturated fatty acid. These groups consisted of 25.5 and 74.5%, respectively, of the total fraction. The fatty acid composition of these groups and the β-fatty acids are shown in Table 6.

Mattson and Volpenhein (1, 2) did not consider the incorporation of endogenous fatty acids with those of exogenous origin in the synthesis of triglycerides, but it is instructive to calculate whether their quantitative conclusions as to the degree of hydrolysis of triglycerides during absorption are in agreement with our findings in these experiments. They concluded that 72% of the dietary triglycerides undergo a partial hydrolysis to β-monoglycerides and that the remaining 28% are completely hydrolyzed. Therefore, 24% (72/3) of the fatty acids are absorbed in the form of β-monoglycerides and 76% of the fatty acids are absorbed in the free form. Since the fatty acid composition of the lymph triglycerides in the present experiment consisted of 31.3% pentadecanoic acid (Table 6) and 68.7% of other fatty acids of endogenous origin, it may be calculated that of every 100 mmoles of triglyceride fatty acids, 7.5 mmoles (31.3% of 24) was absorbed in the form of β-monopentadecanoin and 23.8 mmoles (31.3% of 76) as free pentadecanoic acid. Resynthesis of triglycerides occurs in two ways, by reacylation of the β-monopentadecanoin and by esterification of glycerol via the phosphatidic acid system. It may be assumed that the endogenous fatty acids compete in these processes because of the experimental findings above, and for the present, it is also assumed that this competition is nonselective, that is, esterification is random. Accordingly, the amount of triglyceride formed from reesterification of 7.5 mmoles of β-monopentadecanoin will require 15 mmoles of fatty acid. Since we have assumed that the reacylation is random, the required 15 mmoles should consist of 3.8 mmoles [(23.8/92.5) × 15] of pentadecanoic acid and 11.2 mmoles [(68.7/92.5) × 15] of endogenous fatty acids. Remaining are 20 mmoles (23.8 − 3.8) of pentadecanoic acid and 57.5 mmoles (68.7 − 11.2) of endogenous fatty acids for esterification with glycerol. These fatty acids will give 25.8 mmoles [(20 + 57.5)/3] of triglyceride. If it is assumed again that the fatty acids are distributed randomly among the triglycerides the number of mmoles with pentadecanoic acid in the β-position is 6.6 [(20/77.5) × 25.8] and the number of mmoles of triglyceride having endogenous fatty acids in the β-position is 19.2 [(57.5/77.5) × 25.8]. Therefore, the total number of mmoles of triglyceride having pentadecanoic acid in the β-position is 6.6 + 7.5 = 14.1 or 42.3% [(14.1/6.6 + 7.5 + 19.2) × 100] of the total triglycerides. This value agrees very closely with the experimentally determined value (via lipase hydrolysis and analysis of the β-monoglycerides), which was 42.0%.

The above calculations were made on the basis of a random distribution. In order to determine how much the distribution might deviate from randomness, another experiment was carried out in which methyl pentadecanoate was administered to other cannulated animals of the corn oil group. The fatty acids at the β-position contained 31.7% of pentadecanoic acid as compared to 33.3% required for a random distribution. This deviation would make no great difference in the above calculation, especially since no allowance was made for the difference between the molecular weight of pentadecanoic and of the endogenous fatty acids. It is concluded that these results support the value of 72% for the amount of hydrolysis of triglycerides to β-monoglycerides during

TABLE 6 FATTY ACID COMPOSITION OF LYMPH TRIGLYCERIDES AND OF CORRESPONDING β-MONOGLYCERIDES AFTER TRIPENTADECANOIN ADMINISTRATION

	Total Triglyceride	Total S_3*	Total Others†	β-Position S_3	β-Position Others
	Weight %				
14:0	0.5	tr.	tr.	0.5	0.6
15:0	31.3	75.9	17.2	80.4	25.4
16:0	19.0	18.6	20.4	16.7	21.5
16:1	2.0	—	2.4	—	1.8
17:0	0.7	0.8	0.5	tr.	tr.
18.0	5.7	4.7	6.3	2.4	1.0
18:1	11.7	—	16.6	—	10.5
18:2	20.4	—	27.5	—	31.9
20:1	2.0	—	2.7	—	0.3
20:4	6.7	—	6.4	—	7.0

* S_3 = fully saturated triglycerides.
† Others = triglycerides containing at least one unsaturated fatty acid.

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the absorption of fat proposed by Mattson and Volpenhein.

That the 15:0 did mix with the endogenous fatty acids is shown by its occurrence in the unsaturated triglyceride fraction. Lipase hydrolysis of the saturated fraction showed that it mixed with fatty acids of the endogenous pool in this fraction also. The results of the lipase analysis showed in trisaturated glycerides, 80.4% of it was distributed in the β-position and in the unsaturated triglycerides, 25.4% was in the β-position; of the total 15:0 incorporated into triglycerides, 42% resided in the β-position.

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