## On the Intestinal Absorption of Phospholipids in the Rat ROLFBLOMSTRAND

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Recently Artom and Swanson 1, after feeding 32P-labelled phospholipids to rats and then studying the distribution of radioactivity and phosphorus in the plasma and liver, obtained results, which suggested that phospholipids may be absorbed at various stages of hydrolysis. They suggested that even a small part might be absorbed as intact molecules. Bloom et al 2. have recently made similar experiments with phospholipids labelled with 14C-palmitic acid.

The purpose of the present investigation was to further elucidate the mechanisms concerned with the absorption of phospholipids by studying the composition of lymphlipids after feeding phospholipids labelled in the fatty acid, glycerol or phosphate

portions of the molecule.

Experimental. Phospholipids containing labelled fatty acids were biosynthesized by feeding rats palmitic acid-1-14C. After 5 hours the rats were killed and the total fat from the liver was extracted with alcohol-ether and subjected to chromatography on silicic acid<sup>3</sup>. The labelled phospholipids were dissolved in olive oil (Mixture A, Table 1).

Phospholipids containing <sup>14</sup>C-labelled glycerol were biosynthesized by incubating rat liver slices with glycerol-1-<sup>14</sup>C in a medium of Krebs-Ringer phosphate buffer, pH 7.4, for 2 hours. Free radioactive glycerol was removed from the total fat by dilution with inactive glycerol and washing several times with water. The labelled phospholipids were isolated by precipitating with ice-cold acetone and then dissolved in appropriate amounts of olive oil (Mix-

ture B, Table 1). After hydrolysis almost all of the activity was found in the glycerol portion of the phospholipids and only negligible amounts in the fatty acids. In order to obtain triglycerides labelled in the glycerol portion the neutral fat from these experiments was further purified from phospholipids by chromatography on silicic acid <sup>3</sup> and then dissolved in olive oil (Mixture C, Table 1).

In the same way phospholipids containing <sup>32</sup>P were isolated from samples of blood from a patient suffering from polycythemia vera and treated with <sup>32</sup>P (Mixture D, Table 1).

These mixtures were then fed in an amount of 0.5 ml to rats with a cannulated thoracic duct and the lymph collected for the following 24 hours. The lymph fat was then subjected to chromatography on silicic acid <sup>3</sup> and the distribution of the activity between the neutral fat and phospholipids determined.

Results. In the experiments with phospholipids containing <sup>14</sup>C-labelled fatty acids 52–98 % of the administered activity was absorbed. An average of more than 65 % of the absorbed activity was recovered in the lymph lipids. The major part of this activity was obtained in the neutral fat and about 16 % in the phospholipids. If palmitic-1-<sup>14</sup>C acid is fed in the form of glyceride or as the free acid only about 4 % of the lymph activity is obtained in the phospholipids <sup>2,4,5</sup>.

In the experiments with phospholipids labelled in the glycerol portion more than 60 % of the fed activity was recovered in the lymph lipids. From 57 to 72 % of the lymph activity was found in the neutral fat and 28—43 % in the phospholipids. In similar experiments with triglycerides containing <sup>14</sup>C-labelled glycerol about 52 % of the administered activity was recovered in the lymph lipids. From 76 to 84 % of the lymph activity was found in the neutral fat and the remainder of the activity in the phospholipids.

Table 1. Recovery of activity in lymph lipids after feeding phospholipids containing A. <sup>14</sup>C fatty acids, B. <sup>14</sup>C-glycerol, C. triglycerides containing <sup>14</sup>C-glycerol and D. phospholipids containing <sup>32</sup>P, to rats with lymph fistula. Each rat received 0.5 ml of olive oil containing about 100 mg of labelled phospholipids.

Diet		Number	% absorbed activity	% fed activity	% of lymph activity recovered	
admi	inistered	of rats	recovered in lymph lipids	recovered in lymph lipids	Neutral fat	as Phospholipids
,	<b>A</b>	5	50—81		75—88	12—25 (m = 16)
*	${f B}$	4		<b>55—73</b>	57—72	28—43
	$\mathbf{c}$	2		4559	<b>76—84</b>	16—2 <b>4</b>
	D	2		5—10		100

After feeding phospholipids containing <sup>38</sup>P only about 7 % of the fed activity was

recovered in the lymph.

Discussion. The results of the first experiments show that fatty acids fed in the form of phospholipids are well absorbed mainly via the lymph. After absorption most of the labelled acid in the fed phospholipids was recovered in the lymph glycerides, but a significantly greater amount of activity was obtained in the phospholipids than after feeding palmitic-1-14C acid as glyceride or as the free acid 2,4,5. That these findings mean that a small part of the phospholipids is absorbed completely unchanged seems unlikely but final conclusions must await further experiments.

The results of the experiments with phospholipids labelled in the glycerol por-

the suggestion that most of the ester linkages in the phospholipid molecules between the glycerol and the phosphorus are split during the absorption process. The remaining part of the phospholipid molecule, i. c. a mono- or diglyceride, can then be reutilized for the formation of phospholipids or triglycerides or else degraded further to free fatty acids and glycerol.

The experiments in this investigation with triglycerides labelled in the glycerol portion suggest that a significant part of the glyceride glycerol must have been utilized for synthesis of lymph phospholipids. It is probable that this has been done from a partial glyceride. From data published by Reiser et al. it appears that they also recovered some of the labelled glycerol from fed triglyceride in the lymph phospholipids (cf. Table 2).

Table 2. Comparison of recovery and distribution in lymph lipids after feeding free labelled glycerol. triglycerides containing labelled glycerol or phospholipids containing labelled glycerol or palmitic acid.

Labelled substances dissolved in olive oil and fed to rats with lymph fistula	% fed isotope recovered in lymph lipids	, a		References
Free <sup>14</sup> C-glycerol Glyceride <sup>14</sup> C-glycerol Glyceride <sup>14</sup> C-glycerol Phospholipide <sup>14</sup> C-glycerol PL- <sup>14</sup> C palmitic acid	<2 26—35 45—59 55—73	94—95 76—84 57—72 80 75—88	5—6 16—24 28—43 20	Bernhard et al. <sup>4</sup> Reiser et al. <sup>6</sup> Blomstrand 1954 Blomstrand 1954 Bloom et al. <sup>2</sup> Blomstrand 1954

tion also suggest that the phospholipids are hydrolyzed to a considerable extent although somewhat less than the triglycerides. This suggestion is supported by experiments with free-labelled glycerol4,7 and with glycerol-phosphate 1 which have shown that these substances are only utilized to a very small extent for synthesis of triglycerides or phospholipids in the intestinal contents or cells when free fatty acids are absorbed and transported from the intestine as triglycerides to the lymph.

Since certain amount of activity was found in the lymph after feeding phospholi-pids containing <sup>22</sup>P, these results also seem to indicate the possibility that some phospholipids were absorbed without splitting of the glycerol phosphate bond. However, as the phospholipids in the lastmentioned experiments were of human origin, these experiments are not quite comparable. The preliminary experiments with \*\*P containing phospholipids support

The technical assistance of Miss M. B. Tegman is gratefully acknowledged. This work is part of investigations supported by Knut och Alice Wallenbergs Stiftelse and the Medical Faculty of the University of Lund.

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Received October 16, 1954.

Acta Chem. Scand. 8 (1954) No. 10