

## On the Mechanism of the Intestinal Fat Absorption. II

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We have recently<sup>1</sup> investigated the composition of lymph fat collected from the ductus thoracicus of the cat after feeding C<sup>14</sup> labelled fat. The recoveries of this fat in the lymph were, however, low, *viz.* 4.3—12.8 per cent probably because the animals were anesthetized throughout the entire period of lymph collection. In the present study we have used rats having the intestinal lymph duct cannulated by the Bollman *et al.* technique<sup>2</sup> in order to get better recoveries and more reliable results.

Table 1 shows the types of fats given by stomach tube to adult male rats under a light ether anesthesia at least 18 hours after the operation. Some of the animals were used two or more times. In these cases the animals were fasted 12 hours before the fat administration. The lymph was collected in two portions 0—10 and 10—24 hours after the feeding, extracted with ethanol: ether 3:1 and then with light petroleum. The total fat from the 0—10 hours samples were separated on columns of magnesium oxide, the neutral fat was eluated with acetone, the phospholipids with metanol. The phospholipids thus eluated were the choline containing phospholipids which constituted 90—100 per cent of the lymph phospholipids<sup>3</sup>. The amounts of non-choline containing phospholipids obtained by dissolving the column with hydrochloric acid were too small to be accurately assayed, and hence the values given here only refer to the fatty acids of the choline containing phospholipids. The total fat from the 10—24 hours portions and the two fractions from the first portions were saponified and the activity of the fatty acids determined after wet combustion<sup>3</sup>.

The recoveries in the lymph of the fat fed varied widely. This is probably due to the fact that the conditions of the animals after the operation were different. In some cases the fat rapidly passed the intestine and appeared in the faeces in larger quantities than is found in the normal animal<sup>4</sup>. Diarrhea occurred in some animals especially after feeding hydrolysed corn oil. The

complete quantitative evaluation of the significance under normal conditions of the lymphatic pathway in fat absorption can not be made in these experiments as accessory intestinal lymphatics are ligated and no data are available regarding how much of the small intestine are drained by the main lymphatic which was cannulated.

In all three groups of experiments, however, the same maximum recoveries were found after 24 hours, *i. e.* 80.9, 85.0 and 87.5 per cent of the activity given. These results indicate that under good conditions most of the absorbed fat is transported via the lymphatic channels to the systemic circulation whether fed as glycerides or free fatty acids. These findings do not accord with the partition hypothesis of Frazer<sup>5, 6</sup>. They are, however, in agreement with earlier work of Munk<sup>6</sup> and Ivy<sup>7</sup> and recent work of Tidwell<sup>8</sup> with chylomicroncounting and of Bloom *et al.*<sup>9</sup> using the same technic as we used but without investigating the composition of the lymph fat and of Bollman *et al.*<sup>10</sup> investigating the intestinal lymph of unanesthetized dogs.

The proportions of neutral fat and phospholipids in the lymph were in all three cases about the same. Ninety per cent of the fatty acids were present in the neutral fat and the remaining 10 per cent in phospholipids. The neutral fat consisted chiefly of triglycerides; cholesterol and cholesterol esters representing only a minor part of this fraction. No free fatty acids or soaps appeared in the lymph<sup>3</sup>.

The specific activity of the neutral fat fatty acids were in all three types of experiments about 80—90 per cent of the activity of the fatty acid mixture fed when assayed after hydrolysis. This finding evidence a very active synthesis of glycerides in the intestinal mucosa from free fatty acids fed in A and in B as indicated by work of Munk<sup>6</sup>, Sinclair<sup>11</sup> and Ivy<sup>7</sup> and in contradistinction to Frazer<sup>5</sup>.

The specific activities of the phospholipid fatty acids also showed about the same values in the three types of experiments. This finding is somewhat different from the results in our experiments on cats<sup>1</sup> and indicates that the glycerides might be completely hydrolysed in the intestinal lumen of the rat and then resynthesized in the intestinal wall in agreement with the theories of Verzar<sup>12</sup>. If the hydrolysis was only partial in the intestinal lumen as supposed by Frazer the highly active free fatty acids given in A in a small amount together with inactive corn oil, were only partly diluted by the free fatty acids liberated on partial hydrolysis of the glycerides. The free fatty acid mixture available for glyceride synthesis should then have a specific activity higher than it would if the hydrolysis were complete as in C. In B the specific activity of the eventual free fatty acids is independent of the degree of hydrolysis. As other work in this laboratory<sup>5</sup> indicates that the synthesis of glycerides in

Table 1. The composition of rat intestinal lymph fat. Activity % = specific activity in % of administered fatty acids.

Number of rats	Form of fat administered	Lymph in ml 0-10 h	Total fat in lymph mg 0-10 h	Total cholesterol in lymph mg 0-10 h	Per cent of administered activity recovered in lymph		Fatty acids recovered in lymph as:			
					0-10 h	0-24 h	Phospholipid fatty acids		Neutral fat fatty acids	
							Weight %	Activity %	Weight %	Activity %
5	0.5 ml corn oil + 2.5 mg active palmitic acid -1-C <sup>14</sup> * A	10.4 (16-7)	351 (482-230)	6.5 (9.9-3.2)	52.6 (78.8-35.8)	57.0 (80.9-38.4)	10.9 (14.0-8.3)	50.3 (55.5-43.6)	89.1 (91.7-86.0)	88.0 (99.1-78.8)
6	0.5 ml corn oil trans-esterified with 2.5 mg active palmitic acid -1-C <sup>14</sup> * B	13 (16.5-8.5)	366 (644-196)	7.5 (12.3-4.9)	49.3 (83.4-29.1)	61.7 (85.0-30.2)	11.3 (12.8-9.4)	40.2 (49.7-29.4)	88.7 (90.6-29.4)	79.4 (95.7-64.0)
5	0.5 ml hydrolysed corn oil + 2.5 mg esterified -1-C <sup>14</sup> * C	14.4 (18-12)	260 (426-124)	6.4 (10.6-4.1)	39.5 (72.0-14.5)	62.3 (87.5-45.6)	11.3 (13.3-9.4)	37.2 (50.6-26.6)	88.7 (90.6-86.7)	85.1 (96.8-78.8)

\* Specific activity about 2 · 10<sup>6</sup> counts per minute when assayed as BaCO<sub>3</sub>.

the intestinal wall is in some way related to the phospholipids of the intestinal mucosa, it seems probable that a partial hydrolysis in experiment A would have been reflected in the specific activity of the phospholipids given off from the intestinal wall into the lymph.

The results of this investigation show that in the rat under the conditions of these experiments about 3 per cent of the active palmitic acid fed is transported in the lymph to the blood as phospholipid fatty acids. Thus the intestinal wall supplies a quantitatively important part of the blood phospholipids during fat absorption in the rat as has previously been indicated to be the case in the cat<sup>1</sup>.

#### SUMMARY

The mechanism of the intestinal fat absorption has been studied with C<sup>14</sup> labeled fat in rats with the intestinal lymph duct cannulated.

It has been found that:

1. Absorbed fat is mainly transported via lymphatic channels to the systemic circulation whether fed as glycerides or as free fatty acids.

2. Free fatty acids administered alone or together with glycerides appear in the lymph in glycerides and phospholipids. No free fatty acids or soaps appear in the lymph.

3. The intestinal wall supplies a quantitatively important part of phospholipids to the blood during fat absorption.

4. A complete hydrolysis of the fat in the intestinal lumen might occur in the rat.

#### REFERENCES

1. Bergström, S., Borgström, B., Carlsten, A., and Rottenberg, M. *Acta Chem. Scand.* **4** (1950) 1142.
2. Bollman, J. L., Cain, J. C., and Grindlay, J. H. *J. Lab. Clin. Med.* **33** (1948) 1349.
3. Borgström, B. *Acta Chem. Scand.* To be published.
4. Bergström, S., Borgström, B., and Rottenberg, M. *Acta Chem. Scand.* To be published.
5. Frazer, A. C. *Physiol. Revs.* **20** (1940) 561, *Arch. Sci. physiol.* **2** (1948) 15.
6. Munk, I., and Rosenstein, A. *Arch. path. Anat. Physiol. Virchow's* **123** (1891) 230.
7. Freeman, Smith and Ivy, A. C. *Am. J. physiol.* **114** (1935) 132.
8. Tidwell, H. C. *J. Biol. Chem.* **182** (1950) 405.
9. Bloom, B., Chaikoff, I. L., Reinhardt, W. O., Entenman, C., and Dauben, W. G. *J. Biol. Chem.* **184** (1950) 1.
10. Bollman, J. L., Flock, E. V., Cain, J. C., and Grindlay, J. H. *Am. J. Physiol.* **163** (1950) 41.
11. Sinclair, R. G. *J. Biol. Chem.* **82** (1929) 117; *Ibid.* **121** (1937) 361.
12. Verzar, F., and McDougall, E. J. *Absorption from the intestine.* London (1936); *Arch. Sci. physiol.* **2** (1948) 43.

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