Intrinsic Lymphatic Partition Rate of Mepitiostane, Epitiostanol, and Oleic Acid Absorbed from Rat Intestine

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Mepitiostane (MP), epitiostanol (EP), and oleic acid were administered to the jejunal loop of mesenteric vein- and thoracic duct-cannulated rats, and the intrinsic lymphatic partition rate (ILPR) of the absorbed compounds was directly determined. When ^{14}C-EP was administered to the jejunal loop, recovery of unchanged EP in the mesenteric blood and the lymph was 7.9 and 0.03% of the administered dose, respectively. In contrast, following administration of ^{14}C-MP, recovery of unchanged MP in the mesenteric blood and the lymph was 1.2 and 15.0%, respectively. Thus, following passage through the mucosal cell, 99.6% of the unchanged EP was partitioned into the blood and 0.4% into the lymph, while for unchanged MP, 7.6% was partitioned into the blood and 92.4% into the lymph. When ^{14}C-oleic acid was administered to the jejunal loop, most of the penetrating oleic acid was incorporated into triglycerides in epithelial cells and transferred exclusively into the lymph. However, of the unchanged oleic acid, only 37.6% was partitioned into the lymph and 62.4% into the blood. The ILPR was 92.4% for MP, 0.4% for EP, and 37.6% for oleic acid. We conclude that the ILPR values indicate the true lymphotropic property of the compounds.

KEY WORDS: mepitiostane; intrinsic lymphatic partition rate; intestinal lymphatics; portal absorption; epitiostanol; oleic acid.

INTRODUCTION

Orally administered drugs and nutrients are transferred to the systemic circulation via the portal and/or lymphatic route following passage through the mucosal cells of intestinal lumen. Usually, the portal route is considered to be the main route for compounds absorbed from the intestine (1,2), because blood flow is about 500 times greater than lymph flow in capillaries of the villus (3,4). However, the intestinal lymphatic system is known to play an important role in the absorption of some compounds such as long-chain fatty acids (5,6), cholesterol (7–9), triglycerides (10–12), lipid-soluble vitamins (13–15), and DDT (2,16). A compound absorbed via intestinal lymphatics directly enters the systemic circulation at the level of the subclavian vein. If a drug is extensively metabolized in the liver, the lymphatics may be important as the route to avoid the first-pass metabolism in the liver.

We previously found that mepitiostane (MP), an oral prodrug of epitiostanol (EP), is a typical drug which can avoid the first-pass metabolism in the liver by lymphatic absorption (17). However, the portal absorption of MP could not be defined. Usually, the apparent portal absorption of a compound is calculated by subtracting the amount transferred into lymph from the amount absorbed (18,19) or estimated from the concentration of the compound in the peripheral (20–23) or portal plasma (24,25) after administration to thoracic duct-cannulated rats. However, the true proportions of absorbed compound transferred by the portal and lymphatic routes cannot be determined from the data by these methods.

The present paper reports a direct measurement of the intrinsic lymphatic partition rate, that is, the percentage recovered in the lymph of the absorbed compound in the circulatory system of EP, MP, and oleic acid using mesenteric vein- and thoracic duct-cannulated rats.

MATERIALS AND METHODS

Materials

[4-^{14}C]Mepitiostane (^{14}C-MP, 10.6 μCi/mg) and [14-^{14}C]epitiostanol (^{14}C-EP, 57.5 μCi/mg) were synthesized at Shionogi Research Laboratories. [1-^{14}C]Oleic acid (199 μCi/mg) was purchased from Amersham International plc. The radiochemical purity of the radioactive compounds was confirmed by thin-layer chromatography to be higher than 98%. EP, 2α,3α-epithio-5α-androstan-17-one (KEP), 5α-androst-2-en-17-one (KO), and MP used as authentic samples for the thin-layer chromatography were also synthesized at Shionogi Research Laboratories. Other chemicals and reagents were of analytical or reagent grade. Oil solutions were prepared by dissolving the radioactive compounds in sesame oil at a concentration of 10 mg/ml. The test emulsions were obtained by adding the oil solution to 30 vol of fresh rat bile followed by shaking with a mixer (Microthermo-mixer, Termo Co.) for 1 min just before the administration.

Animals

Female Sprague–Dawley rats (11–13 weeks; body wt, 240–290 g) were purchased from CLEA Japan, Inc., and maintained on commercial chow (CA-1 pellets, CLEA Japan, Inc.) and water ad libitum until surgery. The rats were anesthetized with ethyl urethane (1.4 g/kg, s.c.), and following abdominal incision, the thoracic duct was cannulated with polyethylene tubing (PE50, Clay-Adams) which had been rinsed with dilute heparin by a modification of the method of Bollman (26). The tail vein was also cannulated with heparin-filled polyethylene tubing (PE50). A closed loop of jejunum (5–8 cm) was made by ligation at both ends. After intravenous injection of 0.7 ml of dilute heparin (2000 units/ml) via the tail vein, a third cannula filled with dilute heparin was introduced into the mesenteric vein governing the test jejunal segments. Immediately, 0.5 ml of the test emulsion was instilled into this jejunal loop. The animals were secured on a warmed plate maintained at 38°C. The lymph and mesenteric venous blood were collected in the test tube via each cannula during the 2- or 3-hr period after dosing, under blood transfusion via the tail vein. A diagram of the experimental set-up is given in Fig. 1.

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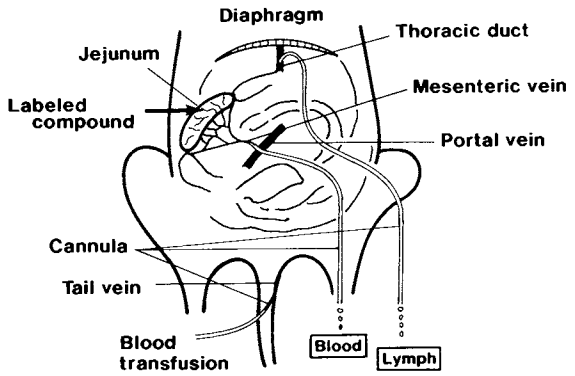

Fig. 1. Diagram of the experimental setup with cannula contraction and sampling system.

Analytical Procedures

Measurement of Radioactivity. Samples (0.05–0.5 ml) of lymph and extracts of lymph and whole blood were added to 10 ml of scintillation fluid (Monophase 40, Packard). Radioactivity was measured with a liquid scintillation counter (Aloka, Model LSC-673). Part of the blood and intestinal homogenates were taken in Combustocones (Packard) and the radioactivity was determined by the combustion method using a sample oxidizer (Packard, Tri-Carb Model 306).

Measurement of Unchanged Drug and Metabolites. Lymph (0.1–0.3 ml) was extracted twice with 5 ml of ethyl ether. Blood (2 ml) was diluted with 4 ml of distilled water and extracted twice with 6 ml of ethyl ether after the addition of 4 ml of acetone. The pooled extracts were evaporated after the total radioactivity had been measured by scintillation counting and chromatographed with the appropriate reference compounds. The radioactive compounds were identified by comparing their R_f values with those of authentic samples using thin-layer chromatography [Merck silica gel 60 plate; EP, petroleum ether (boiling range: 30–60°C)—acetone (3:1); MP, petroleum ether–ethyl ether (10:1); oleic acid, petroleum ether–ethyl ether–acetic acid (75:25:1)]. The areas known to contain specific compounds were scraped.

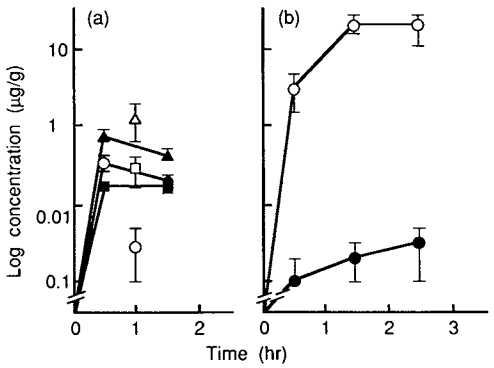

Fig. 2. EP, MP, and metabolite levels in lymph and blood. (a) Concentrations of EP (circle), KEP (triangle), and KO (square) in thoracic duct lymph (open) and mesenteric blood (filled) after intrajejunal administration of ^{14}C-EP (0.5 mg/kg) in rats. (b) Concentration MP in thoracic duct lymph (○) and mesenteric blood (●) after intrajejunal administration of ^{14}C-MP (0.6 mg/kg) in rats. Each value represents the mean ± SD of three rats.

These samples as well as the remaining portions of the plate were transferred to counting vials to measure the radioactivity after the addition of 10 ml of Monophase 40.

RESULTS

Appearance of Unchanged Drug and Metabolites in Blood and Lymph After Intrajejunal Administration of Labeled Compounds

Figure 2 shows the concentration of unchanged drug and metabolites in the mesenteric blood and the thoracic duct lymph following intrajejunal administration of ^{14}C-EP or ^{14}C-MP in sesame oil with bile. As seen in Fig. 2a, EP lymph concentration was lower than the EP blood concentration but the concentration of its metabolites KEP and KO in lymph were somewhat higher than the blood concentration of each compound. In contrast, MP lymph concentration following treatment with ^{14}C-MP was about 500 times higher than the MP blood concentration (Fig. 2b). Thus, MP seems to have a lymphotropic property, unlike EP.

Table I shows the percentage of administered radioactivity recovered in the mesenteric blood and the thoracic duct lymph at 2 hr following administration of ^{14}C-EP. At the end of the experiment, the radioactivity present in the jejunal loop (containing the content) was 49.7% of the administered dose. Therefore, about 50% of administered dose was absorbed from the jejunal loop at 2 hr. Most of this absorbed radioactivity (45.8% of the administered dose) was recovered in the mesenteric blood and unchanged EP, KEP, and KO in this radioactivity were found at 8, 17, and 5% of the administered dose, respectively. On the other hand, only a small amount (2.2%) of the administered radioactivity appeared in the lymph and 0.03% (of the administered dose) of this radioactivity was due to unchanged EP, while 1.52 and 0.34% were due to KEP and KO, respectively. As described in a previous investigation (27), KEP and KO are metabolites of EP formed in the intestinal mucosa in absorption processes.

Table II shows the percentage of the administered radioactivity recovered in the mesenteric blood and the thoracic duct lymph at 3 hr after the intrajejunal administration of ^{14}C-MP. After the administration of ^{14}C-MP, 15.6% of the administered radioactivity was recovered in the thoracic duct lymph and 7.4% in the mesenteric blood. A large portion of the blood radioactivity was due to polar metabolites, with unchanged MP accounting for only 1.2% of the administered dose. In contrast, most of the lymph radioactivity (96.2%) was due to unchanged MP, the amount which could be recovered being 15.0% of the administered dose. Total recovery for EP was 97.8% of the administered dose and 98.7% for MP. Thus, in this experimental system, almost all of the radioactivity absorbed from the jejunal loop was recovered in the mesenteric blood and the thoracic duct lymph. This means that direct measurement is possible of the true proportions of absorbed compound transferred by the portal and lymphatic routes.

Partition of EP, MP, and Metabolites Between Blood and Lymph

Figures 3 and 4 show the absorption behavior from the

Table I. EP and Metabolites in Blood and Lymph 2 hr After Intrajejunal Administration of ^{14}C-EP (0.5 mg/kg) in Thoracic Duct- and Mesenteric Vein-Cannulated Rats[a]

Sample	Blood or lymph flow (g)	% of dose			
		Radioactivity	EP	KEP	KO
Jejunal loop, 2 hr	—	49.7 ± 9.8	—	—	—
Blood					
0–1 hr	17.1 ± 2.0	27.3 ± 5.6	5.1 ± 1.6	10.6 ± 1.9	2.4 ± 0.1
0–2 hr	35.6 ± 5.0	45.8 ± 10.3	7.9 ± 2.2	17.3 ± 3.8	4.9 ± 0.6
Lymph, 0–2 hr	1.6 ± 0.5	2.20 ± 0.36	0.03 ± 0.02	1.52 ± 0.50	0.34 ± 0.07
Total, 2 hr	—	97.8 ± 1.6	—	—	—

[a] Each value represents the mean ± SD of three rats.

Table II. MP and Metabolites in Blood and Lymph 3 hr After Intrajejunal Administration of ^{14}C-MP (0.6 mg/kg) in Thoracic Duct- and Mesenteric Vein-Cannulated Rats[a]

Sample	Blood or lymph flow (g)	% of dose	
		Radioactivity	MP
Jejunal loop, 3 hr	—	75.7 ± 1.4	—
Blood			
0–1 hr	31.4 ± 4.6	1.7 ± 0.2	0.2 ± 0.1
0–2 hr	64.2 ± 9.0	4.5 ± 0.4	0.6 ± 0.1
0–3 hr	96.7 ± 14.9	7.4 ± 0.7	1.2 ± 0.4
Lymph			
0–1 hr	1.1 ± 0.4	2.6 ± 0.5	6.0 ± 5.6
0–2 hr	1.6 ± 0.4	9.1 ± 2.8	8.8 ± 2.7
0–3 hr	2.2 ± 0.4	15.6 ± 1.8	15.0 ± 1.7
Total, 3 hr	—	98.7 ± 1.9	—

[a] Each value represents the mean ± SD of three rats.

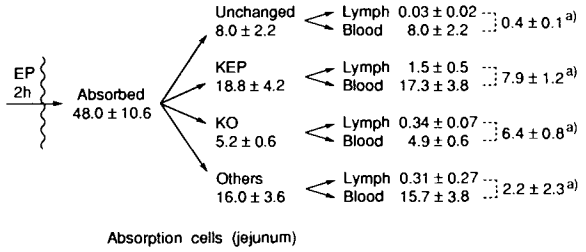

Fig. 3. Partition of EP and metabolites between thoracic duct lymph and mesenteric blood. ILPR: $[D_{lymph}/(D_{lymph} + D_{blood})] \times 100$ (%). The results are the percentage of the dose (mean ± SD, $n = 3$).

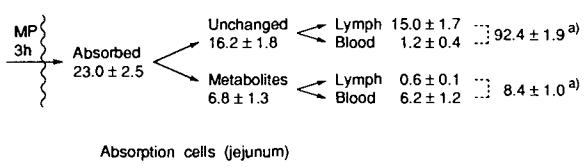

Fig. 4. Partition of MP and metabolites between thoracic duct lymph and mesenteric blood. ILPR: $[D_{lymph}/(D_{lymph} + D_{blood})] \times 100$ (%). The results are the percentage of the dose (mean ± SD, $n = 3$).

jejunum of EP and MP obtained from the data shown in Table I and Table II, respectively. When EP was administered to mesenteric vein- and thoracic duct-cannulated rat, 48.0% of the administered radioactivity was recovered in the mesenteric blood and the lymph after 2 hr, but only 8.0% of the administered dose was due to unchanged EP, 18.8% was KEP, and 5.2% KO. Unchanged EP and its metabolites, KEP and KO, were partitioned between blood and lymph as shown in Fig. 3. The percentage recovered in the lymph of the absorbed compound in the circulatory system (lymph and blood) was defined as the intrinsic lymphatic partition rate (ILPR) and the ILPRs were 0.4% for EP, 7.9% for KEP, and 6.4% for KO. On the other hand, following the administration of ^{14}C-MP, 23.0% of the administered radioactivity was absorbed from the jejunal loop at 3 hr. A part of the absorbed MP was metabolized in mucosal cells during the absorption processes and most of the unchanged MP was partitioned into the lymph. The ILPR of MP was 92.4%.

Following passage through the mucosal cell, 99.6% of the unchanged EP was partitioned into the blood capillaries and 0.4% into the lymphatics. In contrast, for unchanged MP, 92.4% was partitioned into the lymphatics and 7.6% into the blood capillaries.

DISCUSSION

Intestinal absorption usually refers to the process of uptake of a compound from the site of absorption into the systemic circulation. This process includes the penetration through the epithelial cells, metabolism in the epithelial cells, and transfer from the epithelial cells into the blood capillaries and/or the central lacteals in the lamina propria (Fig. 5). Although the principal site of drug metabolism is the liver, some drugs and nutrients are metabolized in the epithelial cells of the gastrointestinal tract during absorption. For example, vitamin A (28) and cholesterol (8) are trans-

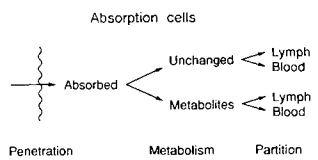

Fig. 5. Schematic representation of absorption behavior of drugs from the small intestine.

Table III. Oleic Acid and Metabolites in Blood and Lymph 3 hr After Intrajejunal Administration of ^{14}C-Oleic Acid (1.1 mg/kg) in Thoracic Duct- and Mesenteric Vein-Cannulated Rats[a]

Sample	Blood or lymph flow (g)	% of dose		
		Radioactivity	Oleic acid	Triglyceride
Jejunal loop, 3 hr	—	72.1 ± 4.0	—	—
Blood				
0–1 hr	24.3 ± 1.9	0.6 ± 0.1	0.6 ± 0.1	0.0 ± 0.0
0–2 hr	50.5 ± 6.8	1.5 ± 0.3	1.4 ± 0.3	0.0 ± 0.0
0–3 hr	76.8 ± 14.1	2.2 ± 0.5	2.1 ± 0.5	0.0 ± 0.0
Lymph				
0–1 hr	0.9 ± 0.5	3.8 ± 2.1	0.3 ± 0.2	3.6 ± 3.3
0–2 hr	1.3 ± 0.6	12.6 ± 4.5	0.7 ± 0.1	11.3 ± 4.2
0–3 hr	1.7 ± 0.6	22.7 ± 3.7	1.3 ± 0.1	20.5 ± 3.4
Total, 3 hr	—	97.1 ± 0.8	—	—

[a] Each value represents the mean ± SD of three rats.

ferred into the lymph after being esterified in the intestinal epithelial cells, and testosterone (2) and naftifine (25) enter the portal blood after being extensively metabolized. EP and MP used in this investigation were also extensively metabolized during intestinal absorption (Tables I and II). Therefore, as shown in Fig. 5, the partition ratio of unchanged drug or metabolite between the blood and the lymph, that is, the intrinsic lymphatic partition rate, must be determined by separation of the metabolic processes which occur during absorption in order to evaluate the lymphatic transfer characteristics of the compound. In this study, the method for direct measurement of the intrinsic lymphatic partition rate of compound was established using mesenteric vein- and thoracic duct-cannulated rats.

When ^{14}C-EP was administered to the jejunum loops of rats, 0.03% of the administered dose (0.06% of the absorbed dose) was recovered in 2-hr thoracic duct lymph as unchanged EP (Table I). In contrast, following the administration of ^{14}C-MP, 15.0% of the administered dose (64.7% of the absorbed dose) appeared in the lymph over the following 3 hr as unchanged MP (Table II). This indicates that MP is selectively absorbed via the lymphatics, but EP is not. However, as described previously, these values do not indicate the partition ratio of unchanged drug between the blood and the lymph because of the participation of the metabolic process during absorption. The ILPR obtained by separation of the metabolic process was 0.4% for EP and 92.4% for MP (Figs. 3 and 4). It should be emphasized that this ILPR, not the percentage of the administered dose (or of the absorbed dose), indicates the extent of the true lymphotropic property of EP and MP.

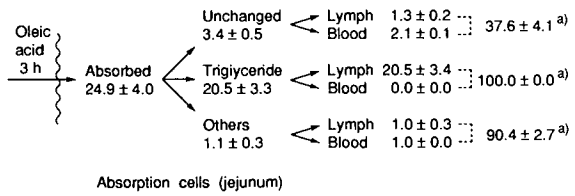

Fig. 6. Partition of oleic acid and metabolites between thoracic duct lymph and mesenteric blood. ILPR: $[D_{lymph}/(D_{lymph} + D_{blood})] \times 100$ (%). The results are the percentage of the dose (mean ± SD, $n = 3$).

This problem can be examined further using oleic acid which was transferred into the lymph following resynthesis to triglyceride in the epithelial cells. Table III shows the percentage of unchanged oleic acid and metabolites of the administered dose recovered in the mesenteric blood and the thoracic duct lymph at 3 hr following administration of ^{14}C-oleic acid to the jejunum loops. Figure 6 also shows the absorption behavior from the jejunum of oleic acid obtained from the data shown in Table III. As shown in Fig. 6, 25% of the administered dose of ^{14}C-oleic acid had been absorbed at 3 hr after the administration. Most of this penetrated oleic acid was incorporated into triglycerides in epithelial cells and transferred almost exclusively into the lymph. This result agreed very closely with a general view of the absorption behavior of long-chain fatty acids (5,6). However, it should be noted that if oleic acid is not incorporated into triglycerides in epithelial cells, not all the oleic acid is transferred to the lymphatics. The ILPR of oleic acid is 37.6% and thus, one-third of the absorbed oleic acid is partitioned to the lymphatics and two-thirds to the blood capillaries. The portal system is the main route of unchanged oleic acid absorbed from the jejunum. On the other hand, the ILPR of triglyceride is 100.0%, i.e., all triglycerides are partitioned into the lymphatics.

The present experiments clarified the intrinsic lymphatic partition rate of EP, MP, and oleic acid from the rat jejunum. The transfer of drugs by the lymphatics is important when trying to avoid the first-pass effect of the liver and to prevent or treat lymphatic metastasis in cancer. The absorption data of many compounds examined in detail, as in the present experiments, will be required to delineate the factors regulating the partition of drugs and nutrients between the lymph and the blood in the gastrointestinal tract.

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REFERENCES

1. T. J. De Marco and R. R. Levine. Role of the lymphatics in the

intestinal absorption and distribution of drugs. *J. Pharmacol. Exp. Ther.* 169:142–151 (1969).
2. S. M. Sieber, V. H. Cohn, and W. T. Wynn. The entry of foreign compounds into the thoracic duct lymph of the rat. *Xenobiotica* 4:265–284 (1974).
3. B. M. Hendrix and J. E. Sweet. A study of amino nitrogen and glucose in lymph and blood before and after the injection of nutrient solutions in the intestine. *J. Biol. Chem.* 32:299–307 (1917).
4. E. J. Reininger and L. A. Spirstein. Effect of digestion on distribution of blood flow in the rat. *Science* 126:1176 (1957).
5. B. Bloom, I. L. Chaikoff, W. O. Reinhardt, C. Entenman, and W. G. Dauben. The quantitative significance of the lymphatic pathway in transport of absorbed fatty acids. *J. Biol. Chem.* 184:1–8 (1950).
6. R. Blomstrand. The intestinal absorption of linolic-1-^{14}C acid. *Acta Physiol. Scand.* 32:99–105 (1954).
7. L. Hellman, E. L. Frazell, and R. S. Rosenfeld. Direct measurement of cholesterol absorption via the thoracic duct in man. *J. Clin. Invest.* 39:1288–1294 (1960).
8. C. R. Treadwell and G. V. Vahouny. Alimentary canal. 3. Intestinal absorption. In C. F. Code (ed.), *Handbook of Physiology, Section 6*, American Physiology Society, 1968, pp. 1407–1438.
9. C. Sylven and B. Borgström. Absorption and lymphatic transport of cholesterol in the rat. *J. Lipid. Res.* 9:596–601 (1968).
10. B. Bloom, I. L. Chaikoff, and W. O. Reinhardt. Intestinal lymph as pathway for transport of absorbed fatty acids of different chain lengths. *Am. J. Physiol.* 166:451–455 (1951).
11. C. E. Rubin. Electron microscopic studies of triglyceride absorption in man. *Gastroenterology* 50:65–77 (1966).
12. A. Nilsson. Intestinal absorption of lecithin and lysolecithin by lymph fistula rats. *Biochim. Biophys. Acta* 152:379–390 (1968).
13. R. Blomstrand and L. Forsgren. Intestinal absorption and esterification of vitamin D_3-1,2-^{3}H in man. *Acta Chem. Scand.* 21:1662–1663 (1967).
14. R. Blomstrand and L. Forsgren. Vitamin K_1-^{3}H in man. Its intestinal absorption and transport in the thoracic duct lymph. *Int. Z. Vit. Forsch.* 38:45–64 (1968).
15. D. L. Yeung and M. J. Veen-Baigent. Absorption of retinal and retinyl esters via lymph and the portal vein in the rat. *Can. J. Physiol. Pharmacol.* 50:753–760 (1972).
16. D. M.-E. Pocock and A. Vost. DDT absorption and chylomicron transport in rat. *Lipids* 9:374–381 (1974).
17. T. Ichihashi, H. Kinoshita, and H. Yamada. Absorption and disposition of epithiosteroids in rats [2]: Avoidance of first-pass metabolism of mepitiostane by lymphatic absorption. *Xenobiotica* (in press).
18. S. A. Hyun, G. V. Vahouny, and C. R. Treadwell. Portal absorption of fatty acids in lymph- and portal vein-cannulated rats. *Biochim. Biophys. Acta* 137:296–305 (1967).
19. G. B. McDonald, D. R. Saunders, M. Weidman, and L. Fisher. Portal venous transport of long-chain fatty acids absorbed from rat intestine. *Am. J. Physiol.* 239G:G141–G150 (1980).
20. A. L. Warshaw, W. A. Walker, and K. J. Isselbacher. Protein uptake by the intestine: Evidence for absorption of intact macromolecules. *Gastroenterology* 66:987–992 (1974).
21. H. Yoshikawa, K. Takada, and S. Muranishi. Molecular weight dependence of permselectivity to rat small intestine blood-lymph barrier for exogenous macromolecules absorbed from lumen. *J. Pharmacobio.-Dyn.* 7:1–6 (1984).
22. H. Yoshikawa, S. Muranishi, C. Kato, and H. Sezaki. Bifunctional delivery system for selective transfer of bleomycin into lymphatics via enteral route. *Int. J. Pharm.* 8:291–302 (1981).
23. K. Katayama and T. Fujita. Studies on biotransformation of elastase. II. Intestinal absorption of ^{131}I-labeled elastase in vivo. *Biochim. Biophys. Acta* 288:181–189 (1972).
24. T. Noguchi, W. N. A. Charman, and V. J. Stella. The effect of drug lipophilicity and lipid vehicles on the lymphatic absorption of various testosterone esters. *Int. J. Pharm.* 24:173–184 (1985).
25. R. C. Grimus and I. Schuster. The role of the lymphatic transport in the enteral absorption of naftifine by the rat. *Xenobiotica* 14:287–294 (1984).
26. J. L. Bollman, J. C. Cain, and J. H. Grindlay. Techniques for the collection of lymph from liver, small intestine or thoracic duct of the rat. *J. Lab. Clin. Med.* 33:1349–1352 (1948).
27. T. Ichihashi, H. Kinoshita, K. Shimamura, and H. Yamada. Absorption and disposition of epithiosteroids in rats [1]: Route of administration and plasma levels of epitiostanol. *Xenobiotica* (in press).
28. J. Ganguly. Absorption of vitamin A. *Am. J. Clin. Nutr.* 22:923–933 (1969).