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Cholesterol absorption by jejunum and ileum

Feeding experiments in man and rat have demonstrated that cholesterol is mainly absorbed in the first part of the jejunum[1-3]. One group[4] reported the major portion of cholesterol was absorbed in the distal half of the small intestine, the site of active absorption of conjugated bile salts[5]. To clarify the intestinal site of cholesterol absorption, bile salt monoglyceride micellar solutions of sterol were perfused through isolated loops of jejunum or ileum in anesthetized hamsters.

Sodium taurodeoxycholate (Maybridge Chemical Co., Tintagel, England), cholesterol (Nutritional Biochemicals Corp., Cleveland, Ohio) 1-monoolein (The Hormel Institute, Austin, Minn.) and [4-^{14}C]cholesterol (New England Nuclear Corp., Boston, Mass.) were 98% pure or better, verified by thin-layer chromatography. Micellar solutions (2.0 mM cholesterol, 5.4 mM monoolein, 6.0 mM sodium taurodeoxycholate, Krebs–Ringer phosphate buffer, pH 6.3) were prepared with a Branson Sonifier Model S125,7.5A, 5 min. Solutions contained [4-^{14}C]cholesterol, about 50000 counts/min per ml.

Male hamsters (100–110 g) were fasted 24 h. Sodium pentobarbital (80 mg/kg) was injected intramuscularly. In 9 animals, the upper jejunum was ligated 2 cm below the entrance of the common bile duct, and 12 cm distal. Both ends were cannulated using silicone tubing (AWG-19, light weight). The proximal cannula was connected to a roller pump (Holter Model RL 175). After an initial 30-min washout with buffer, the micellar solution was infused from a syringe reservoir (12 ml) at a constant rate of 1 ml/min for 2 h. The distal cannula drained back into the reservoir. Ileal segments, in 8 other animals, were cannulated at the ileocecal junction and 12 cm proximally. All perfusions were carried out in a constant temperature room at 37°.

After perfusion, the cannulated intestinal segment was removed intact, rinsed, measured, weighed and homogenized. Lipids were extracted from portions of the original and final perfusion solutions and the gut. Lipid classes were separated by thin-layer silicic acid chromatography. The radioactivity of eluted cholesterol and cholesterol esters was assayed by liquid scintillation counting[6,7]. Calculations from radioactivity data included: percent of cholesterol removed from the original solution (uptake); μmoles cholesterol taken up from solution per unit length or weight; μmoles cholesterol recovered in the intestinal wall per unit length or weight; percent esterification of total intestinal cholesterol. Recovery of radioactive cholesterol from lipid extracts averaged 95%. Cholesterol mass of jejunum and ileum was measured by gas–liquid chromatography of thin-layer chromatographic eluates, as trimethylsilyl ethers, on 3% QF 1 in a Barber–Colman gas chromatograph with hydrogen flame detector. Cholestane was used as a standard[8]. Mass data were calculated as μmoles per unit weight.

The perfused ileum absorbed significantly less cholesterol than did jejunum (Table I). The sterol removed from perfusate per cm of ileum was significantly less than absorption per unit length of jejunum. Uptake from perfusate per 100 mg of tissue was not significantly different in jejunum and ileum ($P<0.1$). The lengths of experimental jejunal and ileal segments were the same; ileal weight per unit length was significantly less than jejunal weight per cm. Mean cholesterol mass per unit

TABLE I

CHOLESTEROL ABSORPTION BY JEJUNUM AND ILEUM

2-h perfusion, 2 mM cholesterol, 5.4 mM monoolein, 6 mM sodium taurodeoxycholate. Means ± S.E. n = number of experiments.

Segment	*Length* (*cm*)	*Weight* (*g*)	*Radioactivity data*				*Sterol Mass* (*μmoles/g*)
			Sterol uptake			*Esters* (%)	
			(%)	(*μmoles/cm*)	(*μmoles/g*)		
Jejunum (n = 9)	12.1 ± 0.47	0.649* ± 0.0666	42.4* ± 3.20	0.895** ± 0.0776	17.75 ± 02.114	10.5 ± 2.67	5.34 ± 0.405
Ileum (n = 8)	12.1 ± 1.16	0.423 ± 0.0559	24.6 ± 4.39	0.523 ± 0.0919	15.92 ± 03.060	10.7 ± 4.90	4.53 ± 0.693

* $P < 0.01$.
** $P < 0.02$ (Student's t test) comparing jejunum and ileum.

weight intestine was similar in jejunum and ileum. Mean water contents of jejunum and ileum were similar (dry weights 22.2 % and 22.4 %, respectively). Mean volumes of solution absorbed from perfusate during experiments were 1.7 ml (jejunum) and 1.3 ml (ileum). Equivalent specific activities of cholesterol were found in the sterol esters of both the jejunum and ileum. More radioactive cholesterol per unit weight at the end of perfusion was recovered in the ileal tissue than in the jejunal tissue (2.062 ± 0.5366 *vs.* 1.766 ± 0.2404 μmoles) (mean ± S.E.). These differences were not statistically significant. Jejunal and ileal absorption of sterol was similar in the first hour. Net absorption leveled off in the ileum in the second hour (Fig. 1). Jejunal cholesterol absorption continued at the same linear rate in the second hour.

The perfused isolated loop of jejunum in an anesthetized hamster is an experi-

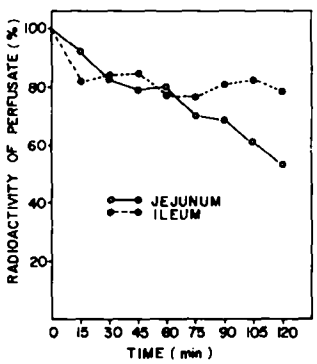

Fig. 1. Disappearance of radioactive cholesterol from loops of small intestine perfused *in vivo* with micellar solutions of 2.0 mM cholesterol, 5.4 mM monoolein and 6.0 mM sodium taurodeoxycholate in Krebs–Ringer phosphate buffer, pH 6.3. Initial radioactivity is expressed as 100%. Percent of initial radioactivity ([^{14}C]cholesterol) remaining in the recirculated perfusate is shown at varying time intervals for 2 h. The perfusate was sampled (0.1 ml) at 15-min intervals. Samples were transferred directly into scintillation fluid (2,5-diphenyloxazole, naphthalene, dioxane) and radioactivity counted in a liquid scintillation spectrometer. Regression equation for disappearance of radioactive cholesterol in jejunal loops over 2 h is $y = -0.3481\,x + 97.1969$.

mental preparation useful for studying absorption. Electron micrographs of preparations perfused with micellar solutions of cholesterol appear normal[9]. The linear time-course of disappearance of radioactivity from perfusates indicates normal absorptive function[10].

Radioactive cholesterol disappeared at similar rates in the first hour from perfused segments of jejunum and ileum suggesting that the mechanism of entry of the sterol is the same in both tissues. Initial processes of absorption to the plasma membrane, binding to membrane components, and diffusion into the cell are followed by intracellular modifications of esterification with fatty acid, and chylomicron formation. Significant exchange of free cholesterol between intestinal tissue and perfusing medium has been demonstrated by both radioactive[6,11] and mass[12] measurements of sterol. Since cholesterol mass of jejunum and ileum is the same per unit weight, while ileal weight per unit length was significantly less than jejunum, the exchangeable pool of cholesterol in the ileal segment was considerably less than that of the jejunal segment. The quantitative role of exchange in cholesterol absorption has not been ascertained, but the plateau in the second hour in ileum may be due to depletion of exchangeable free cholesterol. Cholesterol from the intestinal tissue pool is also secreted (or excreted) into the intestinal lumen[13]. Such a process may be greater in ileum than in jejunum, but was not quantified. The reduced rate of cholesterol absorption by the ileum during the second hour may have been due to several factors. A preferential absorption, by active transport, of conjugated bile salts by the ileal segment would result in less stable micelles of cholesterol. Such an explanation is analogous to that proposed by SIMMONDS *et al.*[13] in studies of upper jejunal absorption of cholesterol from a micellar solution. Rapid absorption of monoglyceride in jejunum resulted in less stable micelles, which would be expected to reduce absorption of cholesterol by the jejunum. Thus, selective absorption of different micelle solubilizers could lead to less efficient absorption of cholesterol by either jejunal or ileal segments in the recirculating system used in these experiments. Further experiments will be required to establish the major parameters regulating and controlling the rate of cholesterol absorption by both jejunum and ileum.

Data did not support any differences between jejunum and ileum in sterol esterification or chylomicron formation under the experimental conditions.

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