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Research Article

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Absorption of Bile Acids from the Large Bowel in Man

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ABSTRACT The absorption of bile acids from the human large bowel was studied in eight patients. All patients had cholecystitis and cholelithiasis and had to undergo cholecystectomy. Cholic acid-^{14}C was injected during surgery into the lumen of the cecum, hepatic flexure of the colon, or transverse colon in six patients, under the visual control of the surgeon. Common duct bile was collected by T tube daily for 5 days, and bile acids were extracted. Significant amounts of radioactivity appeared in T tube bile in each patient. T tube bile acids contained a total of 43.6–84.6% of the administered radioactivity; the average for the six patients was 58.9%. The majority of the tracer was excreted during the first 24 hr. In an additional patient cholic acid-^{14}C was given in the form of an enema 5 days postoperatively. In this subject 30.8% of the retained radioactivity was excreted through the T-tube in 48 hr. The labeled cholic acid was recovered as both cholic and deoxycholic acid from T tube bile. Thin-layer chromatographic analysis of the bile acid samples indicated that the fraction of radioactivity recovered as deoxycholate increased with time during the postoperative period. Gas-liquid chromatographic analysis showed that the daily total quantity of excreted bile acids increased significantly from the 1st–5th days of the experiment. The amount of cholate excreted in T tube bile increased markedly with time, that of chenodeoxycholate increased moderately, and that of deoxycholate decreased sharply during the 5 days of the experiment. In three patients, injection of radiopaque material mixed with the tracer showed no evidence of regurgitation into the small bowel by serial X-rays. In an additional patient, tube aspirate from the terminal ileum contained no radioactivity. The results indicate that cholic acid is converted to deoxycholic acid in the human colon, and both of these bile acids are absorbed from the human large bowel in significant amounts. These data establish the previously unproved concept that significant absorption of bile acids takes place from the large bowel of man.

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INTRODUCTION

According to available evidence bile acids are absorbed or reabsorbed from the lower part of the small intestine in man (2). The human liver synthesizes cholic acid and chenodeoxycholic acid (3, 4), whereas the human gall bladder contains in addition to these two compounds a variety of bacterial transformation products, mainly deoxycholic acid and minute amounts of lithocholic and

TABLE I

Pertinent Laboratory and Other Experimental Data in Eight Patients Studied

Patient	Age	Sex	Laboratory data at time of experiment: Total serum bilirubin	Alk. phosphat. Kau.*	SGOT	SGPT	Thymol turb.	Serum albumin	Serum globulin	Tracer‡	Remarks and specific procedures
			mg/100 ml					*g/100 ml*			
C. C.	60	F	0.3	9.1	—	—	1.2	5.4	1.3	Cecum	—
M. P.	50	M	0.6	20.0	38	40	—	—	—	Hepatic flexure of colon	Radiopaque material§
D. C.	34	F	0.1	6.2	25	10	2.2	3.9	3.2	Cecum	Gantrisin 4 g/day preoperatively
A. S.	31	M	0.5	7.0	—	—	1.1	—	—	Hepatic flexure of colon	Radiopaque material§
B. W.	55	F	0.7	10.0	14	12	0.1	4.0	2.6	Hepatic flexure of colon	Radiopaque material,§ 10 mg of cholic acid added‖
B. S.	72	F	14.3	36.0	440	80	1.6	3.7	2.8	Transverse colon	Chloramphenicol, 2 g/day, 20 mg of cholic acid added‖
H. A.	36	M	0.4	5.0	—	—	1.1	—	—	Hepatic flexure of colon	Contents of terminal ileum tube aspirated
L. A.	79	M	5.8	34.6	165	—	—	4.2	3.5	Enema through rectum	Procedure done 5 days postoperatively

SGOT, serum glutamic oxaloacetic transaminase; SGPT, serum glutamic puruvic transaminase.

* King–Armstrong Units.

‡ Site of administration of cholic acid-^{14}C solution.

§ 25 ml of diatrozoate sodium mixed with tracer.

‖ Nonradioactive cholic acid added to the solution of tracer.

other cholanic acids (4–6). Transformation of cholic into deoxycholic acid, and conversion of chenodeoxycholic into lithocholic acid, has been demonstrated "in vitro" by the incubation of the primary bile acids with fecal material or with bacterial cultures (7–10). Attempts to isolate and identify the various bacteria responsible for these changes have been made (8–10).

The presence of bacterial transformation products of primary bile acids in the human gall bladder implies that either significant absorption or reabsorption of bile acids occurs from the bacterium-rich large bowel in man, or that significant numbers of bile acid–transforming bacteria are present in the human small intestine. In the present study, the absorption of bile acids from the large bowel in man was investigated.

METHODS

Eight patients were studied. The age, sex, and pertinent laboratory data are included in Table I. Each participant had cholecystitis and cholelithiasis with a previous history of common duct obstruction, so that cholecystectomy and the exploration of the common hepatic duct became necessary and was surgically indicated. At the time of surgery six of the subjects had normal serum bilirubin concentrations, and two patients had clinical jaundice with an elevated serum bilirubin level (Table I). The patients were afebrile and ambulatory just before surgery. The experimental procedure was explained to them in detail, and informed consent was obtained. Two additional patients were approached but refused to participate in the study. One patient (D.C., 34 F) was given 4 g of sulfisoxazole by mouth daily for 6 days before surgery. Another (B.S., 72 F) received 1 g of chloramphenicol by mouth daily for 2 days before surgery, and the antibiotic was continued intravenously for the duration of the experiment after the removal of the gall bladder. The remaining six patients received no antibiotics.

Cholic acid-carboxyl-^{14}C [1] (56 mc/mmole) was purified by thin-layer chromatography (TLC) (11). Appropriate tracer amounts were dissolved in 20 ml of water and were sterilized in sealed vials. After sterilization the tracer was rechromatographed (TLC) and was found to be 98.5% pure on radioassay and yielded a single peak on gas-liquid chromatography.

Cholecystectomy was performed in each patient, and a T tube was placed in the common hepatic duct by the

[1] New England Nuclear Corp., Boston, Mass.

surgeon. In two patients appendectomy was performed, and 9.6–22 μc of cholic acid-^{14}C was injected into the lumen of the cecum through the appendiceal stump, under the visual control of the surgeon (Table I). In four additional patients (Table I) the tracer was injected into the lumen of the hepatic flexure of the colon through the wall of the intestine with a fine needle. A purse string suture was placed over the needle hole in this area to close it securely. In another patient (Table I) the labeled cholic acid was injected into the lumen of the transverse colon about 10 cm distal to the hepatic flexure. In three patients (Table I) 25 ml of radiopaque material (diatrizoate sodium) was mixed with the tracer and was injected into the lumen of the hepatic flexure of the colon. Serial X-rays (flat plate of the abdomen) were taken during the first 48 hr after surgery (every 6 hr during the first 18 hr, then every 10 hr). In two patients (Table I) 10 and 20 mg of nonradioactive cholic acid, respectively, was added to the tracer solution and was injected into the lumen of the large bowel together with the labeled material.

T tube bile was collected daily for 5 days in six patients. The bile was drained into sterile bottles containing 100 ml of ethanol, in 24-hr portions. In one of these subjects (B.W., 55 F), the bile output during the first 24 hr was collected in four separate 6-hr portions; the bile output from the 2nd–5th day was collected as 24-hr specimens. In another patient (H.A., 36 M) a tube was passed into the terminal ileum, facing the ileocecal valve. Ileal contents were aspirated, and T tube bile was collected for 10 hr after surgery in this patient. The ileal aspirate was processed like the T tube bile, to test for the possible presence of radioactivity as described below.

In the eighth patient (L.A., 79 M) the experiment was carried out 5 days after surgery. At this time, the patient was afebrile, out of bed, and was fed by mouth. Cholic acid-^{14}C was mixed with 250 ml of an aqueous 20% diatrozoate sodium solution, and was administered into the large bowel as an enema through a rectal tube with an inflated balloon. The progress of the fluid was monitored by X-ray fluoroscopy, and periodic flat plates of the abdomen were taken. The mixture filled the large bowel uniformly from the rectum to the hepatic flexure of the colon, and faintly outlined the cecum. There was no evidence of regurgitation of the fluid into the small bowel. The enema fluid was retained for 1½ hr, was then allowed to flow out through the rectal tube, and was collected in a plastic bag. The enema fluid was processed for quantitative radioactivity as outlined below, and the amount of radioactivity retained in the large bowel was determined. Bile was collected from a T-tube, inserted during surgery 5 days earlier. The bile collection began at the time of the start of the enema, and was carried out for 48 hr in two separate 24-hr containers as described above.

Extraction of bile acids. The volume of the bile specimens was measured. All samples were brought to constant volume with hot ethanol and were stored at 4°C until extracted. Two suitable aliquots (10–50 ml) of equal volume were taken, and a known amount of cholic acid-^{14}C was added to the second sample of each pair of specimens to monitor recovery. The per cent recovery of the added cholic acid-^{14}C, extracted parallel with each of the bile acid samples, varied from 72 to 102% in the eight patients, with an average of 89.5%. The data shown in the results section have been corrected for recovery. The specimens were filtered, and 10–30 ml of a solution of 33% KOH was added to bring the pH to 11. The mixtures were put in a steam bath at 75–80°C for 1 hr under a gentle nitrogen stream. This procedure resulted in the evaporation of most of the ethanol and the hydrolysis of any cholesterol esters which might have been present. Neutral steroids were removed from the aqueous solution with equal volumes of hexane by three extractions. The hexane was backwashed with water and discarded, and the water was added back to the original solution. The remaining aqueous solution was autoclaved in a pressure cooker at 15 lb of pressure for 3 hrs. The solution was cooled in an ice bath, and methanol was added in a ratio of water : methanol of 5 : 2. The cooled solution was acidified (Congo red) with 50% sulfuric acid. Fatty acids were removed by two extractions with an equal volume of hexane; the hexane was backwashed with water and was discarded. Bile acids were extracted from the water : methanol layer by shaking five times with an equal volume of peroxide-free ethyl ether. The ether was washed with water and was evaporated at 30–40°C under a gentle nitrogen stream. The residue containing the bile acids was dissolved in 100 ml of methanol and stored at 4°C. Radioactivity was determined in a Tri-Carb liquid scintillation spectrometer, in a toluene solution containing 4 mg of 2,5-diphenyloxazole and 0.3 mg of 1,4-*bis*-2-(5-phenyloxazolyl)-benzene per ml. For each unknown amount, a parallel sample was run with known amounts of radioactivity in the vial to determine quenching (internal standard method).

The amount of radioactivity recovered by the above method from the different fractions was determined as follows: a known amount of cholic acid-^{14}C was added to a nonradioactive bile specimen, which was processed in quadruplicate. The neutral steroid fractions (hexane extraction, alkaline medium) contained an average of 1.06% of the total radioactivity present. The corresponding average figure for the fatty acid fractions (hexane extraction, acid medium) was 1.75%, and the bile acid fractions (ethyl ether extraction, acid medium) was 88.1%, respectively. The remainder (water : methanol layer) contained but trace amounts of radioactivity.

Thin-layer chromatography (TLC). TLC was carried out on 20 × 20 cm silica gel G plates. Each bile acid extract was chromatographed on a separate plate with a solvent system of isooctane : isopropyl ether : acetic acid, 50 : 25 : 25(11). Known bile acids (cholic, deoxycholic, and chenodeoxycholic) were run on each plate, together with the unknown samples. No attempt was made to account for the small percentage (less than 5%) of radioactivity which may have been present in the bile in ketonic bile acids produced by bacterial action. The plates were sprayed with a 10% solution of phosphomolybdic acid in ethanol. Each row of spots, corresponding to a

TABLE II

Excretion of Radioactivity in T Tube Bile (Bile Acid Fraction of Extracts) in Six Patients for 5 Days, after the Injection of Cholic Acid-^{14}C into the Lumen of the Large Bowel

Day	C.C.	M.P.	D.C.	A.S.	B.W.	B.S.	Average‡
				% recovered*			
1	29.4	78.9	44.2	55.9	57.4	39.3	50.9
2	5.5	1.8	3.5	4.2	3.2	5.6	4.0
3	2.5	2.6	1.4	1.0	0.3	1.8	1.6
4	2.9	1.1	4.3	0.5	0.1	1.0	1.7
5	3.3	0.2	1.1	0.3	0.1	0.4	0.9
Total§	43.6	84.6	54.5	61.9	61.1	48.1	58.9

* Per cent of total administered radioactivity, recovered from T tube bile.

‡ Averages of the six patients.

§ Total 5-day excretion.

specific bile acid, was scraped quantitatively from the plates into a liquid scintillation counter vial. 4 ml of methanol was added to the gel, and the mixture was heated to 50°C for 30 min. After cooling, toluene scintillation solution (see above) was added to the tubes, and radioactivity was determined. Quenching was monitored automatically with an external standard source of radioactivity. The total radioactivity disintegrations per minute recovered from each plate was calculated, and the results were expressed in per cent of radioactivity distributed among the various bile acids.

Gas-liquid chromatography (GLC). Bile acids were esterified with methanol by standing overnight at room temperature in 2% sulfuric acid in methanol. After methylation, the solution was diluted with an equal volume of water. The methyl esters of the bile acids were extracted three times with equal volumes of ethyl ether: benzene (1:1). The extracts were washed with aqueous saturated sodium bicarbonate solution, and then with water, until neutral. The organic solvent was evaporated at 60°C under a gentle air flow. The methyl esters of the bile acids were dissolved in ethanol, the solution was brought to constant volume, and an aliquot was taken for radioactivity count. The radioactivity of this sample was compared to the activity of the original methanol solution of the bile acids, to correct for recovery (85–95%). A measured aliquot of the solution containing the bile acid methyl esters was used for GLC on a 6 ft × 4 mm column packed with 3% QF-1 (methylfluoroalkyl silicone) on 100/120 mesh gaschrom Q.[2] The instrument used was a Barber-Coleman, model 5000, with flame ionization detectors. Known quantitative standards of methyl cholate, deoxycholate, and chenodeoxycholate were chromatographed under the same conditions, and the amount of the individual bile acids in each unknown was calculated by comparison with the standards by a disc integrator. Corrections for recovery were done as indicated above.

[2] Applied Science Laboratories Inc., State College, Pa.

RESULTS

The procedure was well tolerated by each patient, and no ill effects were encountered at any time because of the experiment. On the 2nd day after surgery the patients were out of bed. On the 3rd postoperative day they were given clear fluids to drink, and on the 4th or 5th day they were fed solid food. On the 3rd postoperative day the patients were given an enema. Spontaneous bowel movements, if any, began on the 4th or 5th postoperative days. The temperature of the patients did not exceed 100.5°F at any time during the postoperative period.

Absorption of bile acids from the large bowel. Significant amounts of radioactive bile acids appeared in T tube bile in each patient. Table II shows the results of the excretion of radioactivity through the T tube in six patients, who were studied for the full 5 day duration of the experiment. The total amount of labeled material excreted during the 5 days varied from 43.6 to 84.6% of the administered dose. The average excretion for the group was 58.9% of the radioactive dose given. The major part, 29.9–78.9%, was excreted through the T tube during the first 24 hr. The average excretion for the 1st day was 50.9% of the administered dose of label in the six patients. Smaller amounts were excreted from the 2nd–5th

TABLE III

Excretion of Radioactivity in T Tube Bile, and Results of the Analysis of Bile Acids in Patient B.W., 55 Yr Female, during the First 24 Hr of the Experiment

Procedure	Unit	Hours after injection of tracer 0–6	6–12	12–18	18–24
Total radioactivity recovered	*dpm*, % of total given	15.1	24.1	10.4	7.8
TLC; distribution of radioactivity; % of total *dpm* (per plate)	Origin	1.6	2.5	0.7	1.1
	Cholic acid	34.3	15.8	5.1	4.3
	Chenodeoxycholic	1.7	1.7	2.0	2.1
	Deoxycholic	62.0	79.8	92.0	92.1
	Solvent front	0.3	0.2	0.3	0.4
GLC; mass of bile acid methyl esters, *mg* (total per sample)	Cholic	83.9	85.8	44.9	35.9
	Chenodeoxycholic	56.7	40.6	24.2	11.4
	Deoxycholic	46.4	35.0	22.7	16.8

TLC, thin-layer chromatography; GLC, gas-liquid chromatography.

TABLE IV

Average Per Cent Distribution of Radioactivity in T Tube Bile (Bile Acid Extracts) by Thin-Layer Chromatographic Analysis in Six Patients during the 1st and 2nd Days of the Experiment

	Day 1		Day 2	
	Average	SE	Average	SE
	%		%	
Origin	1.8	0.3	3.7	0.9
Cholic acid	34.4	9.1	11.5	2.3
Chenodeoxycholic acid	5.0	1.8	7.8	1.3
Deoxycholic acid	58.0	33.1	76.7	3.5
Solvent front	0.1	0.04	0.4	0.28

From the 3rd–6th days of the study the number of counts in most specimens (see text) was insufficient under the experimental conditions given (less than 3000 counts/100 min per TLC plate.)

days. The peak excretion period was between the 6th and 12th hr of the 1st day after the administration of the labeled material in one patient (Table III).

In four patients (C.C. 60 F, D.C. 34 F, A.S. 31 M, and B.S. 72 F) serial blood samples were drawn during the experiment every 4 hr during the 1st day, and then daily thereafter. 1 ml of serum of each sample was checked for the presence of radioactivity. There was no demonstrable radioactivity present in the serum of the patients.

Analysis of bile acids. The results of TLC analysis of the bile acid extracts of T tube bile are shown in Table IV in the six patients studied for 5 days. In the bile acid extracts of the first 24 hr of the experiments an average of 34.4% of the radioactivity migrated to the spot corresponding to cholic acid, and an average of 58% of the label was recovered from the spot corresponding to deoxycholic acid (Table IV). The corresponding figures for the 2nd day averaged 11.5 and 76.7% respectively. During the 3 following days the amount of radioactivity recovered from the TLC plates was too low to be measured, except in one patient. In this subject only 1.1% of the radioactivity migrated to the spot corresponding to cholic acid, and 88.7% of the label was recovered from the spot corresponding to deoxycholic acid on the 5th day of the study. There were only small amounts of radioactive material recovered from the origin, solvent front, and the spots corresponding to chenodeoxycholic acid (Table IV). Since it may be presumed that labeled cholic acid was not transformed to chenodeoxycholic acid, the 5 and 7.8% of radioactivity recovered from the spots corresponding to chenodeoxycholic acid may represent overlaps due to the imperfection of the method or unidentified ketonic acids referred to in Methods. While it has been suggested that pure cultures of certain fecal anaerobes can dehydroxylate cholic acid in the 12-position to form chenodeoxycholic acid (12), there presentely exists no evidence that chenodeoxycholic or lithocholic acids can be formed from cholic acid in man (13). Table III includes the distribution of radioactivity in the bile extracts of the fractions of the first 24 hr of the experiment in one patient.

The average daily volume of bile excreted through the T tube was 320, 281, 263, 372, and 470 ml/24 hr, respectively, from the 1st–5th postoperative days in six patients. The results of the GLC analysis of T tube bile are shown in Table V. The daily excretion of total bile acids decreased sharply during the 2nd and 3rd days after surgery. During the 4th and 5th postoperative days, the amount of total bile acids excreted increased several fold (Table V). Although there was an increase in the volume of bile flow as well, the concentration of bile acids in the T tube bile increased two to threefold from the 1st to the 5th day after surgery. The amount of cholic acid excreted through the T tube increased from an average of 382 mg on the 1st day, to an average of 2391 mg on the 5th day. Chenodeoxycholic acid excretion showed less of a change (Table V). The excretion of deoxycholic acid decreased from an average of 152 mg on the 1st postoperative day to 22 mg on the 5th day. Table III includes the excretion of bile acids during the fractions of the first 24 hr in one subject.

Site of absorption. In three patients radiopaque material was injected into the lumen of the hepatic flexure of the colon together with the tracer (Table I). Serial X-rays of the abdomen indicated that the radiopaque material filled the hepatic flexure, the transverse colon, and faintly outlined the cecum during the 48 hr of monitoring. There was no evidence of regurgitation of the radiopaque material through the ileocecal valve into the small bowel on any of the X-rays.

In one patient, the contents of the terminal ileum

TABLE V

Gas-Liquid Chromatographic Analysis of the Daily Excretion of Bile Acids (mg/24 hr) in T Tube Bile (Bile Acid Extracts) in Six Patients during 5 Days of the Experiment

Patient	Day...1	2	3	4	5
			mg/24 hr		
Methyl cholate					
C. C.	227	68	76	152	229
M. P.	26	578	449	1593	3213
D. C.	165	22	332	2411	1528
A. S.	842	389	315	2983	7327
B. W.	251	105	72	776	1234
B. S.	783	199	398	1043	817
Average	382	227	274	1493	2391
Methyl chenodeoxycholate					
C. C.	178	50	31	44	59
M. P.	41	78	138	372	568
D. C.	94	18	68	582	417
A. S.	556	181	67	384	1027
B. W.	133	59	43	128	152
B. S.	236	111	73	189	134
Average	206	83	70	283	393
Methyl deoxycholate					
C. C.	240	40	13	26	55
M. P.	13	9	17	25	0
D. C.	147	11	107	79	31
A. S.	310	18	0	20	41
B. W.	121	18	8	0	0
B. S.	79	28	6	12	6
Average	152	21	25	27	22
Daily total bile acid methyl esters					
C. C.	645	158	120	222	343
M. P.	81	665	604	1990	3781
D. C.	406	51	507	3072	1976
A. S.	1708	588	382	3387	8395
B. W.	505	182	123	904	1386
B. S.	1098	338	477	1244	957
Average	741	330	369	1803	2806
Average *mg/ml* bile*	2.3	1.2	1.4	4.8	5.9

* Average amount of bile acids (mg) per 1 ml of T tube bile excreted.

were tube aspirated for 10 hr after the injection of cholic acid-^{14}C into the lumen of the hepatic flexure of the colon (Table I). During this period 34.7% of the administered dose of radioactivity was excreted through T tube bile. There was no radioactivity present in the tube aspirate from the terminal ileum.

The effect of anesthesia. In one patient labeled cholic acid was administered in the form of an enema 5 days postoperatively. The enema fluid was retained in the bowel for 1½ hr. The outflowing enema fluid, collected in a plastic bag (see Methods), contained 40.2% of the total radioactivity added. During the first 24 hr after the administration of labeled cholic acid, 29.7% of the radioactivity retained in the large bowel was recovered from T tube bile. During the 2nd day 1.1% of the label appeared in the bile. Thus a total of 30.8% of the retained dose was absorbed during the initial 2 days of the experiment. The

amount of radioactivity recovered from T tube bile during the 2 days was 18.5% of the total amount of label initially added to the enema fluid.

Other results. The addition of 10 mg and 20 mg, respectively, of nonradioactive cholic acid to the tracer in two patients (Table I) had no visible influence on the rate of absorption or on other results of the study.

The preoperative administration of sulfisoxazole or the pre- and postoperative administration of chloramphenicol had no detectable influence on the results of this investigation.

DISCUSSION

The data of the present study support the previously unproved concept that significant amounts of bile acids are absorbed from the large bowel in man. About one-half of the labeled bile acids injected into the lumen of the large bowel was absorbed and recovered from T tube bile. The absorption seemed to be relatively rapid, most of the material being absorbed during the first 24 hr. Cholic acid is relatively rapidly converted to deoxycholic acid in the human large bowel (Table III). Both of these bile acids are absorbed or reabsorbed from the human colon in significant quantity. The colonic site or sites of absorption cannot be ascertained with certainty from the present study. The cecum, ascending colon, hepatic flexure, and transverse colon may be sites of reabsorption, in whole or in part. There is no information on the remaining parts of the large bowel from the present data.

After the insertion of a T tube into the common hepatic bile duct the total mass of excreted bile acids and the concentration of bile acids in the bile decreased during the 2nd and 3rd postoperative days. By the end of the 5th day, however, the total mass of excreted bile acids increased about fourfold, and the concentration of biliary bile acids increased about threefold (Table V). The reasons for these phenomena are not apparent. Whether local edema of the tissues, the operative procedure itself, the absence of oral feeding during the first 48 hr, or the absence or decrease of feedback inhibition of hepatic bile acid biosynthesis during the latter part of the experiment are in part responsible for this phenomenon is not quite clear from these data. However, it is known that in patients with choledochostomy drainage (3) and in the bile fistula rat (14, 15) interruption of the enterohepatic circulation of the bile produces a marked increase in bile acid excretion. These data suggest that a feedback mechanism of bile acid production is operative in man as well as in the rat. It has already been shown that in bile fistula rats not only cholesterol biosynthesis from acetate (15, 16), but also the 7α-hydroxylation of cholesterol, the initial step in bile acid formation, is markedly increased (17, 18).

Data in Table V indicate a marked increase of the amount of excreted cholate in T tube bile from the 1st–5th day of the experiment. The corresponding amounts of deoxycholate showed a sharp decrease. This finding suggests that most of the bile acids were excreted to the outside through the T tube, and thus only very small quantities of cholic acid were transported to the sites of dehydroxylation. This conclusion, however, must be qualified, since the rates of reabsorption of cholic and deoxycholic acids from the human large bowel are not known, and the figures obtained may be influenced by this factor. Ekdahl and Sjovall (3) analyzed T tube bile acids in patients with choledochostomy drainage and reported that deoxycholic acid was regularly found during the 1st postoperative day but then decreased or disappeared. The results of the present study are in agreement with this finding.

The distribution of radioactivity between different bile acids is included in Table IV. It must be pointed out that the actual distribution of the label in T tube bile may not represent the degree of conversion of cholic to deoxycholic acid within the lumen of the large bowel. There is no information available on the comparative rates of absorption of cholic and deoxycholic acids from the human large bowel. Since differences in this respect may influence the distribution of radioactivity in the excreted bile, the data in Table IV do not necessarily reflect the rate of conversion, but show only the relative percentage of respective labeled materials in the excreted bile.

It should be pointed out that the administration of sulfisoxazole and chloramphenicol (Table I) failed to alter the per cent recovery of radioactivity in deoxycholic acid. The dehydroxylase activity presumably resides in anaerobic organisms (9, 10, 12), and these organisms are resistant to most of the commonly used antibiotics.

It was clearly shown that in the present studies the tracer was absorbed from the large bowel, and that there was no regurgitation of the radioactive material through the ileocecal valve. The experiment carried out 5 days after surgery, showing a significant absorption of the tracer from enema fluid, ruled out the possibility that the process of anesthesia, the initial postoperative absence of oral feeding, or the operative process per se were responsible for the absorption of the labeled bile acids.

In the rat, it has been shown that significant amounts of bile acids are absorbed from the cecum. Lindstedt and Samuelsson estimated that in these animals 51% of the total amount of cholic acid is converted into deoxycholic acid per day (19). They further estimated that of the total amount of cholic acid which is transported to the large intestine per day, 70% is absorbed and transported back to the liver (19). Norman and Sjovall injected labeled bile acids into the lumen of the cecum of bile duct–cannulated rats (20). The bile of the animals contained 42–79% of the labeled material. The order of magnitude of these figures is in agreement with the data in man observed in the present study. The absorption of bile salts from the large bowel of the rat was confirmed by Holt (21) and by Sullivan (22).

During the enterohepatic circulation of bile, most of the bile acids excreted by the liver are reabsorbed during transit in the small bowel, are carried back to the liver by the portal vein, and are eventually reexcreted into the gall bladder and used again. The pool of bile acids is circulated 6–10 times daily through the enterohepatic cycle. At each cycle a small fraction of transiting bile acids will escape reabsorption in the small bowel, will find its way into the large intestine, and will appear in the feces. According to published data, daily fecal excretion of bile acids varies from 87 mg (cholic acid metabolites only) to 2114 mg, depending on the methods of measurement used, and on the regimen given to the patient under study (23). If the average daily excretion of bile acids is assumed to be of 500 mg in an average adult, of this amount about 200 mg is deoxycholic acid and 300 mg is nonabsorbable lithocholic acid (24). Then, in the context of the present study. an approximately equal amount of deoxycholic acid should be reabsorbed from the large intestine during daily transit. This implies that about 700 mg of bile acids escape absorption from the small bowel daily and reach the human large intestine. Of this, 200 mg are reabsorbed during transit in the large bowel. Considering that the total pool size of bile acids is about 1.2–4 g in man (25, 26), these data suggest that about 5–20% of the pool is exposed daily to the effects of bacteria in the large bowel before reabsorption takes place. The presence of significant amounts of bacterial transformation products of the primary bile acids in the human gall bladder can be explained on the basis of these data.

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