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Influence of semisynthetic diet and type of fat on the turnover of deoxycholic acid in the rabbit*

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SUMMARY

The half-life, pool size, and daily synthesis of deoxycholic acid in ten rabbits were 7.0 days, 752 mg, and 75.8 mg, respectively, on the control diet; and 24.1 days, 1010 mg, and 30.2 mg, respectively, on a semi-synthetic diet containing hydrogenated coconut oil. Corresponding values in eight rabbits on a diet in which the hydrogenated coconut oil was replaced by corn oil were 26.9 days, 1164 mg, and 30.6 mg, respectively, compared to 8.0 days, 910 mg, and 85.0 mg during the control period. The marked drop in serum cholesterol level that occurred when the dietary coconut oil was replaced by corn oil was not accompanied by an increased bile acid synthesis or excretion. The fecal excretion of bile acids was reduced in rabbits fed the semisynthetic diets, whereas the urinary excretion was the same as on the control diet. Urinary elimination corresponded to about 24% of the daily synthesis of deoxycholic acid in rabbits fed the semisynthetic diets. The concentration of bile acids in blood was calculated to be 1.15–3.04 mg/100 ml of whole blood in four rabbits fed the hydrogenated coconut oil diet, and 0.33–2.02 mg in four rabbits fed the corn oil diet. By using a gas-liquid chromatographic technique, a preliminary survey of the fecal sterols and bile acids in rabbits fed different diets was made. The dietary effects on bile acid metabolism are discussed, and it is concluded that the turnover of bile acids in rabbits is the same whether hydrogenated coconut oil or corn oil is fed.

Several investigations have been published on the influence of dietary fat on fecal bile acid excretion. After studying the metabolism of labeled cholesterol in rats and humans, three groups of investigators have concluded that approximately equal amounts of bile acids are excreted with the feces whether the diet given is rich in unsaturated or saturated fat (1–3). However, other groups of investigators using titrimetric or colorimetric methods have found that fecal bile acid excretion is increased by unsaturated fat (4–8). Because of these conflicting results and because the investigations do not permit any conclusions regarding bile acid *turnover*, we have found it of interest to study the problem with the aid of labeled bile acids. The rabbit has been chosen as the experimental animal because of the pronounced influence of dietary fat on its serum cholesterol level. It was thought that, if the changes in serum cholesterol level were brought about by differences in bile acid turnover, it would be possible to detect these differences most easily in the rabbit.

EXPERIMENTAL METHODS

Details of the various methods used have been described in the preceding paper (9).

Diets and Animals. The control diet was the conventional low-fat commercial pellet diet used in the previous investigation. The experimental diets, made up in pellet form, were semisynthetic (containing casein, starch, sucrose, glucose, fat, salts, vitamins, and cellulose) and differed only in the type of fat included. The fat constituted 26% of the calories either in the form of hydrogenated coconut oil or corn oil. A detailed description of these diets has been published by Wigand (10). All diets were given in isocaloric amounts. The rabbits, which weighed between 2 and 3.5 kg, were usually given about 80–90 g of the control diet and about 60–70 g of the semi-synthetic diets per day. Water was given ad lib.

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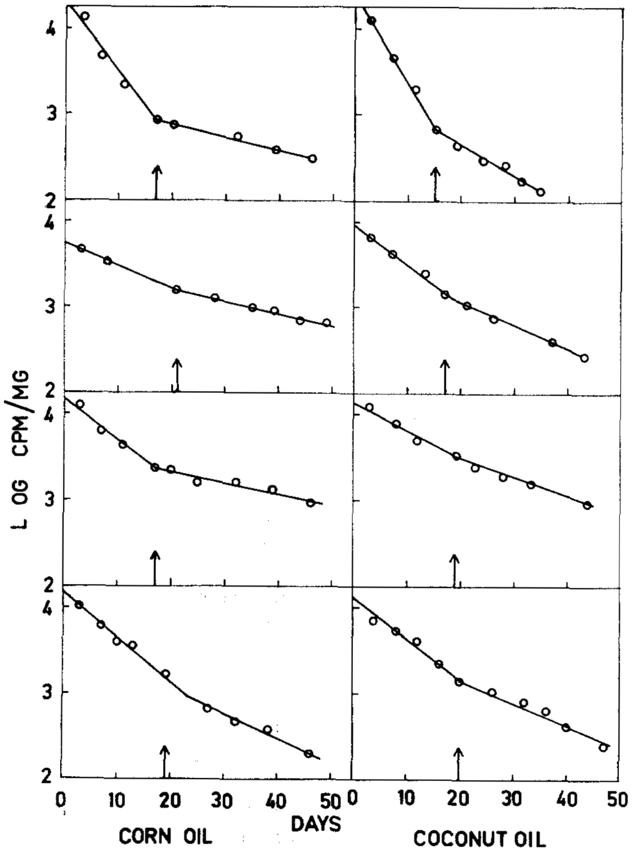

FIG. 1. Examples of semilogarithmic plots of the specific activity of biliary deoxycholic acid during periods when the diet was changed. The arrows indicate the first day that the semi-synthetic diet was fed in place of the conventional commercial pellets.

The method used for determining half-life, pool size, and daily production of deoxycholic acid was described in the preceding paper (9). Because of the large individual variations, each rabbit had to serve as its own control and received a dose of deoxycholic acid-24-C^{14} when fed the control diet. The decrease in the specific activity of biliary glycodeoxycholic acid was followed for a period of time long enough to allow accurate determination of the turnover of deoxycholic acid. When such data had been obtained, the diet was changed to a semi-synthetic one containing either corn oil or hydrogenated coconut oil. The specific activity of glycodeoxycholic acid was such as to permit continued measurements of the declining activity during the change of diet and also for a considerable period of time on the new diet.

When variable periods of time (27–147 days) had elapsed after the change to the semisynthetic diet, each rabbit received a new dose of labeled deoxycholic acid. A sufficient number of bile samples were taken to allow the determination of the deoxycholic acid turnover, after which a change of diet was again made. Rabbits fed coconut oil were switched to corn oil; one rabbit, fed corn oil first, was transferred to the coconut oil diet. In cases where the specific activity of glycodeoxycholic acid was high enough, continued measurements of the specific activity were made during the change of diet and for a considerable period of time on the new diet. When the animals had been fed the second semi-synthetic diet for about 1–3 months, a new dose of isotope was administered and the turnover data were again determined.

Since these studies required a very long observation time, some rabbits died during the experiments. The main causes of death were accidents during the collection of bile samples and infections. Several rabbits died after having been fed the coconut oil diet for several months. The cause of death was not established. However, all values given in the present paper were obtained with apparently healthy rabbits.

Blood samples were taken from a marginal ear vein. Total serum cholesterol was determined by the method of Abell *et al.* (11) as modified by Anderson and Keys (12). Feces and urine were collected and worked up as described in the preceding paper (9).

RESULTS

Qualitative analysis of the biliary bile acids with paper and gas-liquid chromatography gave the same results in all the rabbits studied irrespective of the diet fed. Glycodeoxycholic acid was by far the predominant bile acid (9). Mainly, free deoxycholic acid was found in the small intestine, both on the commercial diet and on the semi-synthetic diets. The distribution of radioactivity within the body was similar to that found in rabbits fed commercial pellet food. Since this distribution is influenced by the functional state of the gallbladder, it would be difficult to detect variations caused by dietary changes.

When rabbits fed the control diet were given either of the semisynthetic diets, there was an almost immediate effect on the rate of decrease of the specific activity of the biliary glycodeoxycholic acid (Fig. 1). The slope of the line obtained by plotting the logarithm of the specific activity *versus* time became less pronounced. In many instances, the new decreased slope was maintained unchanged when followed for four half-lives. In other cases, however, the slope tended to decrease progressively, giving apparently longer half-life values with time. There was no significant difference in the effect of the two types of semisynthetic diets. This was further illustrated in an experiment with rabbit K8. This rabbit was first studied during the change from control to corn oil diet and back to control diet (Fig. 2,

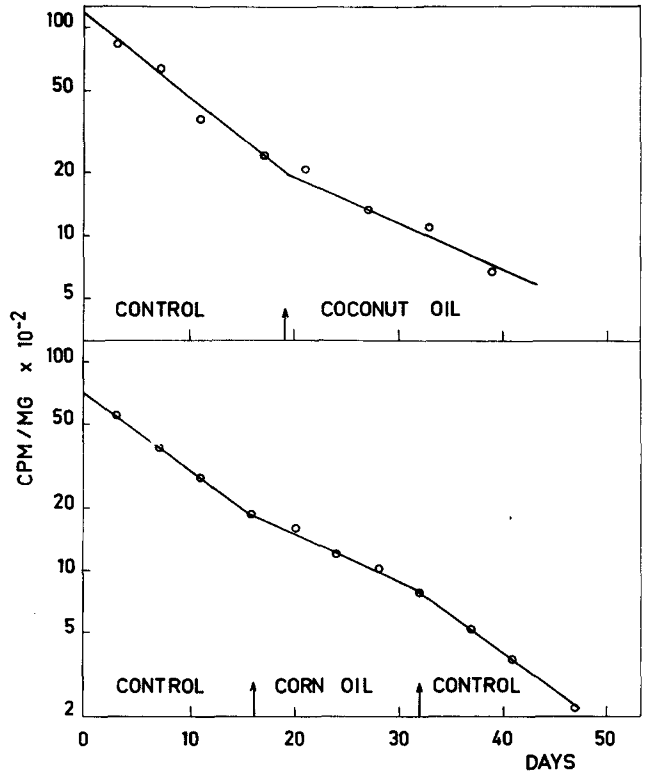

FIG. 2. Semilogarithmic plot of the specific activity of biliary deoxycholic acid during periods of dietary changes (rabbit K8).

TABLE 1. TURNOVER OF DEOXYCHOLIC ACID IN RABBITS ON THE CONTROL DIET AND THEN ON ONE OF THE SEMISYNTHETIC DIETS

Rabbit No.	Diet	No. of Days on Diet*	Half-Life	Pool Size	Production
			days	*mg*	*mg/day*
KD	Control	. . .	5.5	484	61.3
	Coconut oil	25	13.0	785	41.8
K1	Control	. . .	4.1	522	88.3
	Coconut oil	109	16.0	872	37.9
K14	Control	. . .	7.0	902	89.9
	Coconut oil	79	29.0	900	21.6
K19	Control	. . .	3.2	275	59.7
	Coconut oil	90	24.0	436	12.7
K20	Control	. . .	6.2	508	56.9
	Coconut oil	100	13.0	274	19.6
K11	Control	. . .	9.4	823	61.7
	Corn oil	79	40.0	1480	25.6
K15†	Control	. . .	7.0	978	96.8
	Corn oil	90	26.0	1115	29.8
K17	Control	. . .	3.8	654	120.0
	Corn oil	118	23.0	749	22.6
	Corn oil	240	24.0	618	17.8

* At the beginning of the turnover determination.

† This rabbit was studied with cholic acid-4-C^{14} on the corn oil diet.

TABLE 2. TURNOVER OF DEOXYCHOLIC ACID IN RABBITS ON THE CONTROL DIET AND ON BOTH TYPES OF THE SEMISYNTHETIC DIETS

Rabbit No.	Diet	No. of Days on diet*	Half-Life	Pool Size	Production	Serum Cholesterol†
			days	*mg*	*mg/day*	*mg/100 ml*
K12	Control	. . .	9.6	1020	73.8	24
	Coconut oil	44	36.0	1770	34.1	789
	Corn oil	147	32.0	2120	46.7	182
K30	Control	. . .	8.1	744	63.9	42
	Coconut oil	95	31.0	1740	39.0	301
	Corn oil	77	32.0	1245	27.0	103
K8	Control	. . .	7.7	1031	93.5	76
	Coconut oil	147	18.0	1265	48.8	699
	Corn oil	60	15.8	882	38.8	134
K10	Control	. . .	5.7	872	106.0	49
	Corn oil	75	18.0	880	34.0	95
	Coconut oil	60	27.8	782	19.5	321
KC	Control	. . .	12.5	1160	64.3	101
	Coconut oil	24	33.0	1280	26.9	263
	Corn oil	26	27.6	900	22.6	

* At the beginning of the turnover determination.

† Mean during the turnover determination.

lower half). The animal was then given the control diet for another month, and the effect of changing from control to coconut oil diet was studied (Fig. 2, upper half). As seen in Fig. 2, the rate of fall of specific activity of deoxycholic acid was equal in all control periods and more rapid than during the periods on either of the semisynthetic diet. Different amounts of labeled deoxycholic acid were given in the two experiments, which explains the absolute differences in specific activity between the upper and lower parts of Fig. 2.

When the animals had been given a semisynthetic diet for about 1–5 months, labeled deoxycholic acid was administered and the half-life, pool size, and daily production of deoxycholic acid were calculated. The results are shown in Tables 1 and 2. It is seen that in all cases the semisynthetic diets caused a prolongation of the half-life and a decrease in the daily synthesis of deoxycholic acid. The pool size decreased in only one rabbit and was increased or unchanged in the other animals. These changes were of the same order of magnitude with both types of semisynthetic diets. This is clearly illustrated in Table 2, which shows the results obtained with rabbits given all three types of diet during different periods.

When rabbits given the coconut oil for several months were transferred to the corn oil diet, there was a sharp drop in the serum cholesterol level. As shown in Fig. 3, this change was not accompanied by any change in the decay curve of deoxycholic acid.

Excretion of Bile Acids in Feces and Urine. Approximately 20–40 g of feces was excreted daily, the weight of which was 10–20% higher when the control diet was fed than when the semisynthetic diet was given. This difference is explained by a higher water content of the

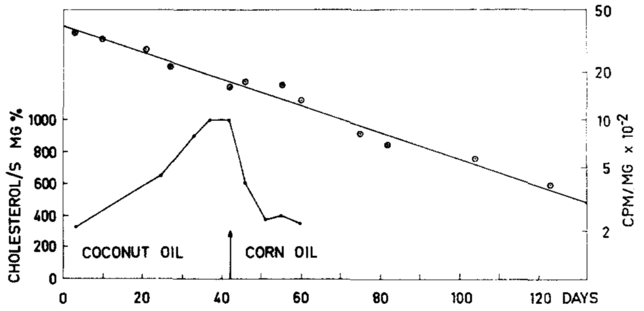

FIG. 3. Semilogarithmic plot of the specific activity of biliary deoxycholic acid (O—O), and changes in serum cholesterol level (●—●) during a period when the diet was changed from the saturated to the unsaturated type (rabbit K12).

feces during the control period and by the larger amount of cellulose in the control diet.

When rabbits on the control diet were given any of the semisynthetic diets, there was an immediate decrease in fecal excretion of the isotope administered (Fig. 4 and 5). This decrease approximately corresponded to the changed decline of the specific activity curves. When rabbits were transferred from the coconut oil diet to the corn oil diet, no change in the fecal isotope excretion was found. Thus, in rabbit K12 (see Fig. 3), the fecal radioactivity corresponded to a mean of 17 and 20 mg of deoxycholic acid, respectively, in samples collected before and after the change of diet.

Fecal excretion did not account for all of the expected daily isotope elimination. In animals fed the semisynthetic diets, the urinary excretion of radioactivity amounted to about 24% of the total activity recovered. This was equivalent to 5.7 mg and 8.0 mg of labeled bile acids per day in the rabbits fed the coconut oil and corn oil diets, respectively.

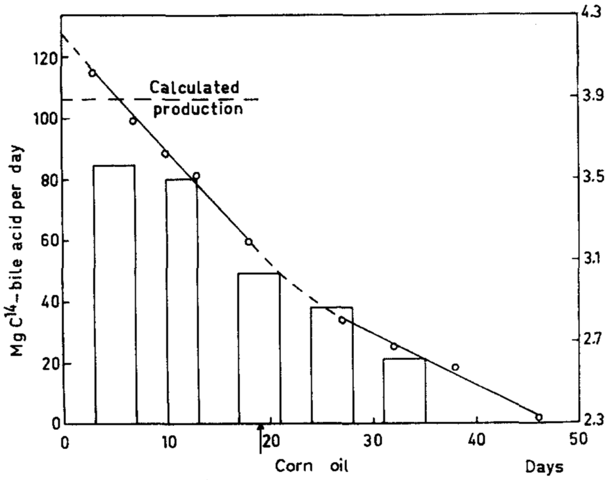

FIG. 4. Semilogarithmic plot of the specific activity of biliary deoxycholic acid (O—O), and fecal excretion of isotope during a period of dietary change. The arrow indicates the first day that the semisynthetic diet was given. The fecal excretion is expressed as milligrams of labeled bile acids per day (rabbit K10).

TABLE 3. COMPARISON BETWEEN THE DAILY PRODUCTION OF DEOXYCHOLIC ACID CALCULATED FROM HALF-LIFE AND POOL SIZE, AND THE DAILY EXCRETION OF LABELED BILE ACIDS IN FECES AND URINE

Rabbit No.	Diet	Calculated Production*	Calculated Fecal Excretion†	Calculated Urinary Excretion†
		mg/day	*mg/day*	*mg/day*
KC	Coconut oil	26.9	11.1	8.4
KD	" "	41.8	29.3	5.1
K10	" "	19.5	12.0	5.1
K14	" "	21.6	11.4	4.3
K8	Corn oil	38.8	23.1	11.8
K11	" "	25.6	14.0	8.5
K12	" "	46.7	30.5	6.9
K15‡	" "	29.8	13.4	7.6
K30	" "	27.0	15.7	5.3

* Based on measurements of pool size and half-life.
† Total daily radioactivity in fecal or urinary bile acid fraction divided by the specific radioactivity of biliary deoxycholic acid.
‡ This rabbit was studied with cholic acid-4-C^{14}.

The results of the studies of fecal and urinary elimination are summarized in Table 3. The values in this table were obtained by dividing the total radioactivity of fecal and urinary extracts by the specific activity of the biliary deoxycholic acid at the time of sampling. The results are mean values of samples collected intermittently during several months. Samples taken during the first week were not included. The calculated amounts of bile acids excreted per day were constant during the study (see preceeding paper [9]).

Nature of Fecal Sterols and Bile Acids. Preliminary investigations of the nature of fecal sterols and bile acids have been carried out by using gas chromatog-

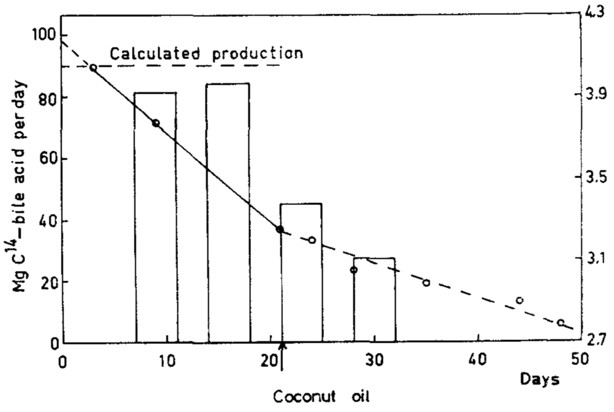

FIG. 5. Same as in Fig. 4 except for the type of semisynthetic diet fed (rabbit K14).

TABLE 4. Retention Times of Some Sterols Relative to Cholesterol on the Same Column and Under the Same Conditions Used for the Chromatograms Shown in Fig. 6

Sterol	Relative Retention Time	Sterol	Relative Retention Time
Coprostane-3β-ol	0.87	7-Dehydrocholesterol	1.14
Coprostane-3α-ol	1.00	Ergosterol	1.26
Cholesterol	1.00	Stigmasterol	1.40
epi-Cholesterol	1.00	Cholestanone	2.17
Cholestane-3β-ol	1.10	7-Ketocholesterol	5.72

raphy (13, 14). Purification of fecal extracts was done as described in the preceeding paper (9). Since the extraction procedures were not primarily designed for the analysis of neutral sterols, minor changes might have occurred in some sterols during the progress of purification. Large differences between rabbits were found in the sterol chromatograms. Examples of gas chromatograms of neutral sterols are shown in Fig. 6 (rabbit K8). This figure shows major peaks with the retention times of 6.6, 7.6, 8.3, 8.6, 10.9, and 12.5 min. The two last-mentioned peaks can probably be ascribed to metabolites of dietary sterols. The three first-mentioned peaks have the retention times of coprostanol (6.6 min); cholesterol, epicholesterol, and 3α-coprostanol (7.6 min); and cholestanol (8.3 min), respectively. The relative retention times of some sterols under the same conditions are shown in Table 4. The proportions between the first three peaks were different in samples from different animals. The "coprostanol" peak was exceptionally large in rabbit K8 on the control diet (Fig. 6). This peak usually diminished markedly when semisynthetic diets were given so that the "cholesterol" and "cholestanol" peaks became predominant.

Gas chromatograms of fecal bile acids were qualitatively similar but differed markedly in the relative sizes of the peaks. Since the nature of the compounds is partially unknown, quantitative studies cannot yet be carried out. Gas chromatograms of samples from rabbits fed different diets are shown in Fig. 7 and 8. Peaks with the following retention times (in minutes) deserve mention (tentative structure of acid in parentheses): 6.3(3β-hydroxycholanic), 7.1 (lithocholic), 12.0 (3β,12α-dihydroxycholanic), 14.2 (deoxycholic), 16.4 (probably not a bile acid), 18.7 (3β-hydroxy-12-ketocholanic), 22.6 (12-ketolithocholic), 25.8 (unknown), and 40.0 (3,12-diketocholanic). Although differences are seen in the proportions between the peaks in samples from the same rabbit on different diets, these differences have not been consistent in the animals studied.

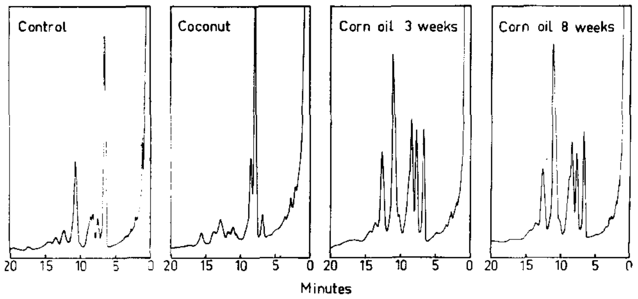

Fig. 6. Gas chromatography of sterols in fecal samples collected during different dietary periods (rabbit K8). Conditions: 6-ft x 5-mm glass column, 0.5% QF-1 on 100–140 mesh Gas-Chrom P, 198°, 1.0 kg/cm².

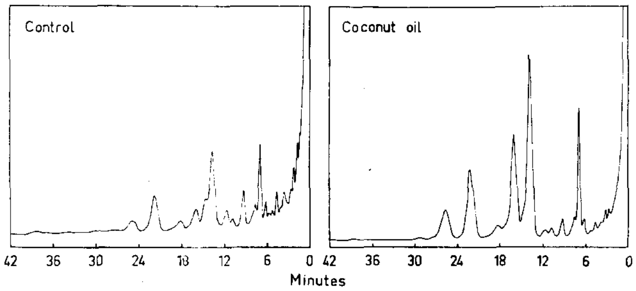

Fig. 7. Gas chromatography of methylated bile acids in fecal samples collected during different dietary periods (rabbit KD). Same column as in Fig. 6. Temperature, 219°.

Blood Bile Acids were calculated by dividing the total radioactivity in blood by the specific radioactivity of deoxycholic acid in bile. The results are summarized in Table 5. It was calculated that a mean of 2.05 mg and 1.23 mg of deoxycholic acid per 100 ml of whole blood was present in rabbits fed the coconut and corn oil diet, respectively. There was a large variation even between blood samples from the same animal, possibly due to technical difficulties.

DISCUSSION

The present investigation has shown that the fecal excretion of deoxycholic acid decreases pronouncedly

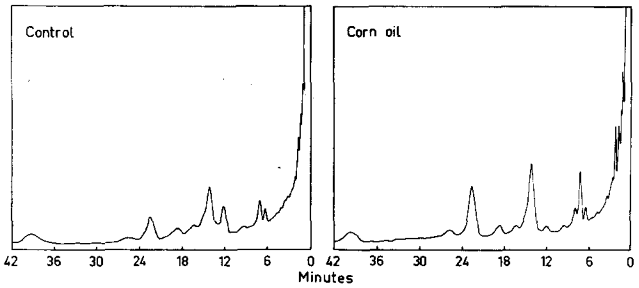

Fig. 8. Gas chromatography of methylated bile acids in fecal samples collected during different dietary periods (rabbit K10). Column conditions as in Fig. 7.

TABLE 5. CONCENTRATION OF BILE ACIDS IN BLOOD EXPRESSED AS MG OF DEOXYCHOLIC ACID PER 100 ML OF WHOLE BLOOD

Rabbit No.	Diet	No. of Samples Analyzed	Bile Acids, mg/100 ml Mean	Bile Acids, mg/100 ml Range	Days Between First and Last Sample
KD	Coconut oil	1	2.79	.....	..
K10	" "	4	1.68	1.20–2.41	90
K21	" "	3	1.49	1.15–2.13	4
K22	" "	2	2.23	1.42–3.04	2
Mean	" "	...	2.05	1.15–3.04	..
K8	Corn oil	3	1.42	0.78–2.02	15
K30	" "	3	1.39	1.21–1.49	18
KC	" "	5	1.33	0.65–1.85	86
K17	" "	6	0.79	0.33–1.67	37
Mean	" "	...	1.23	0.33–2.02	..
Mean*	Control	...	1.21	0.26–3.10	..

* See preceding paper (9).

when the diet is changed from a conventional low-fat commercial pellet diet to a semisynthetic diet containing casein, starch, sucrose, glucose, fat, salts, vitamins, and cellulose. At the same time, there is a rapid increase in the apparent half-life of the deoxycholic acid. The last-mentioned finding does not necessarily mean that deoxycholic acid synthesis is diminished since the deoxycholic acid pool may increase during the period of dietary change. Later determinations of half-life and pool size, when the animals had been given the semisynthetic diet for several months, indeed showed that 10 out of 13 animals had an increased deoxycholic acid pool. The mean increase in all rabbits was about 30%. There was no correlation between the increase in pool size and the length of time that the rabbits had been fed the semisynthetic diet. Unless there is a rapid and transitory increase of the pool size at the time of the change of diet, the values in most rabbits indicate that the increase of the apparent half life is primarily due to a diminished synthesis of deoxycholic acid in connection with the decreased fecal excretion.

When the turnover data were determined in animals fed a semisynthetic diet for 1-5 months, it was found that the deoxycholic acid synthesis was reduced by approximately 60% as compared with the value in the control period. The half-life of deoxycholic acid was 2.5–6 times longer and, as mentioned above, the pool size was usually increased. These findings are similar to those of Portman and Murphy in studies of the turnover of cholic acid in rats (15). They found, however, that the cholic acid pool was decreased in rats fed diets of the semisynthetic type.

Most of the isotope given was recovered in the feces and urine. As pointed out in the preceding paper (9), rabbits on the control diet have a remarkably high urinary excretion of bile acids, corresponding to about 10% of the daily deoxycholic acid synthesis. In contrast to the fecal excretion, the urinary excretion is essentially the same in rabbits fed semisynthetic diets. Because of the decreased fecal excretion, the urinary elimination corresponds to about 24% of the daily deoxycholic acid synthesis. This route for bile acid excretion has never been considered important in previous studies of dietary effects on bile acid turnover in healthy animals.

Rabbits on a control diet have a remarkably high concentration of bile acids in blood compared with rats and monkeys (9, 16, 17). The mean values found here for labeled bile acids in blood were 2.05 and 1.23 mg/100 ml of whole blood in rabbits fed the coconut oil and corn oil diets, respectively. The large individual differences noted make it difficult to evaluate the significanec of the differences between the values in rabbits fed the two types of diet. It should be mentioned that the corresponding value on the control diet was 1.21 mg/100 ml of whole blood (9).

Several mechanisms for the reduced synthesis of deoxycholic acid in rabbits on the semisynthetic diets can be visualized. Normally, cholic acid is the main primary bile acid formed from cholesterol in the liver. The major part of this acid is converted by intestinal microorganisms into deoxycholic acid, which becomes the main bile acid in rabbit bile (18). In the preceding paper, the possibility of a formation of chenodeoxycholic acid from cholesterol in the liver was discussed (9). Although chenodeoxycholic acid cannot be detected in the bile, gas chromatograms of methyl esters of fecal bile acids show the presence of a peak with the retention time of methyl lithocholate. Lithocholic acid is known to be a metabolite of chenodeoxycholic acid in the rabbit and it has also been isolated from rabbit feces (19). It is theoretically possible that the reduced synthesis of deoxycholic acid could be compensated by an increased synthesis of chenodeoxycholic acid in the liver. However, gas chromatograms of fecal bile acids did not show any consistent differences in the relative amounts of lithocholic acid in animals fed the various diets. Roughly estimated, lithocholic acid accounted for 10–20% of the total fecal bile acids.

Another explanation for the decreased deoxycholic acid production could be the formation of cholic acid metabolites other than deoxycholic acid in the intestine. One of the major metabolites of this kind in the rat is 7-ketodeoxycholic acid. This acid, however, has been found to be extensively transformed into deoxycholic acid in the rabbit (20), and its formation therefore would be largely included in the deoxycholic acid production.

Furthermore, gas chromatography of fecal bile acids has failed to detect any 7-ketodeoxycholic acid in the three groups of animals. One rabbit fed the corn oil diet was given cholic acid-4-C^{14}, and the turnover of deoxycholic acid was calculated in the usual way — i.e., by assuming that all of the activity had entered the deoxycholic acid pool. The equilibration of isotope and the fall of specific activity of the deoxycholic acid was similar to that obtained when labeled deoxycholic acid was given to other rabbits. The calculated pool size and daily production of deoxycholic acid did not differ from those found in other rabbits of the same group. These findings, and the results of the paper and gas chromatographic analyses of bile and feces in all groups of rabbits, indicate that only a small part of the cholic acid formed in the liver may bypass the deoxycholic acid pool when excreted with the feces.

It appears therefore that the deoxycholic acid production represents the main part of the bile acids synthesized in the rabbit liver. Furthermore, variations in the deoxycholic acid production seem to be representative of the total bile acid production in the liver, at least when the rabbits are fed the diets used in this investigation.

The effect of the semisynthetic diets on bile acid turnover might be brought about either by a direct action on the liver or by a change in the absorption of bile acids from the intestine, which in turn might be due to an increased coprophagy. The very rapid decrease of bile acid excretion that occurs when the diet is changed to the semisynthetic type suggests that the primary action of these diets is on the intestinal absorption. An increased absorption of bile acids probably leads to a depression of the bile acid synthesis in the liver (21), and it might also explain the increase in pool size usually observed in rabbits on the semisynthetic diets. If the action were primarily on the synthesis in the liver, one would expect less immediate effects on the fecal excretion.

It is known that alterations in the intestinal flora can influence the bile acid turnover (22). If this is due to a change in the composition of the bile acids formed by the action of micro-organisms, one would expect a more drastic change in the fecal and/or intestinal bile acids than the relatively minor differences observed by gas and paper chromatographic analyses. Another possible mechanism for the action of micro-organisms has been suggested by Norman (23). He found that different bacteria bind various bile acid metabolites more or less firmly when these metabolites have been formed. In this way, the intestinal flora might control the amounts of bile acids accessible for absorption. Our results do not contradict such a theory.

The control diet used in this investigation differs in many respects from the semisynthetic diets. The amount of cellulose is higher in the control diet, which will increase the intestinal residue. This might be of importance for the absorption of bile acids. The differences in fecal weights were much smaller than the differences in fecal bile acid excretion in the three groups of animals.

It is well known that rabbits eat a large amount of the feces directly from the anus (24). The extent of coprophagy might possibly be greater in rabbits fed semisynthetic diets leading to an increase in the amount of bile acids available for absorption. In a preliminary experiment, a rabbit fed the control diet was equipped with a large plastic collar that prevented coprophagy. The half-life of deoxycholic acid was found to be the same whether or not the rabbit had a collar. Further investigations are needed to clarify this point.

Two semisynthetic diets were used in this work, one containing hydrogenated coconut oil and the other corn oil as the fat source. The mean values for half-life, pool size, and daily synthesis of deoxycholic acid in rabbits fed the coconut oil diet were 24.1 days, 1010 mg, and 30.2 mg, respectively, compared with 7.0 days, 752 mg, and 75.8 mg during the control period. The values for rabbits fed the corn oil diet were 26.9 days, 1164 mg, and 30.6 mg, respectively, and 8.0 days, 910 mg, and 85.0 mg in the same rabbits during the control period. It is therefore obvious that no difference in the deoxycholic acid turnover exists between rabbits fed the two types of fat. Furthermore, no change in the excretion of bile acids was observed in three rabbits studied during the change from a coconut oil diet to a corn oil diet. During the coconut oil periods, the serum cholesterol level was high and often rising, whereas during the corn oil periods serum cholesterol levels were lower and often falling. In contrast to the variations in serum cholesterol levels, the semilogarithmic plot of the specific activity of deoxycholic acid was linear. Furthermore, the ratio between total daily isotope excretion and the specific activity of biliary deoxycholic acid was constant throughout the experimental periods.

Thus, our investigation has clearly shown that the differences in serum cholesterol levels between rabbits fed hydrogenated coconut oil or corn oil diets are not correlated with differences in the bile acid production.

Preliminary quantitative analyses of fecal sterols with gas chromatography indicate that the drop in serum cholesterol level occurring when rabbits fed coconut oil are transferred to a corn oil diet is not accompanied by an increased excretion of sterols derived from cholesterol. It seems possible that the rapid drop of serum cholesterol caused by feeding corn oil to rabbits is primarily due to a transfer of serum cholesterol to the tissues. Studies of the cholesterol distribution in rats fed different fats have shown that, in a steady state, animals fed corn oil have a higher concentration of cholesterol in the liver than those fed coconut oil (25). It has also been shown that, when a low-fat diet is supplemented with 2% or 10% of corn oil, there is an increase in the cholesterol concentration of the tissues and a decrease of the serum cholesterol level (26).

The conflicting results obtained regarding the effect of dietary fat on bile acid excretion both in man and in rats (1-8) are probably due to differences in the techniques used to determine the bile acids. In view of the variable and complex nature of the fecal bile acids, it appears to us that the tracer techniques are subject to fewer errors than titrimetric or colorimetric methods of doubtful specificity.

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