

Dietary Bile Acids and Lipid Metabolism. V. Reversibility of the Effects of Lithocholic Acid in Chicks. (28973)

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Previous reports have conclusively shown that dietary lithocholic acid will elevate plasma lipids, increase liver size and induce a proliferation of bile ductules in the livers of chicks(1-5). We have also indicated that the ductular proliferation induced by dietary lithocholic acid can be reversed by feeding a diet free of this bile acid(3). The data presented here demonstrate that the changes induced in chicks by lithocholic acid are completely reversed by feeding a diet devoid of the bile acid.

Experimental. Male Hy-line White Leghorn chicks were used in 2 experiments. The chicks were fed a stock diet for one week prior to initiation of Experiment 1 and for 3 weeks prior to Experiment 2. The chicks were allocated to the treatment groups in such a way as to have essentially identical initial mean body weights for all groups. The chicks were housed in heated cages having raised wire floors. Food and water were supplied *ad libitum*. Food consumption and body weight were determined at weekly intervals. The basal diet employed was identical to that described previously(2). Lithocholic acid when fed was substituted for glucose at a level of 0.2%. In Exp. 1, one group of chicks received the basal diet and a second group the basal diet supplemented with lithocholic acid for 3 weeks. At this time, 8 chicks were sacrificed from each group; the remaining 8 chicks in each group received the basal diet for 3 weeks. In Exp. 2, chicks were fed the basal or lithocholic acid-supplemented diet for 3 weeks; 5 chicks from each group were sacrificed at the end of this period. Ten chicks remaining in each group were then fed the basal diet for the remainder of the experiment. Five chicks from each group were sacrificed 6 and 9 weeks after initiation of the study.

At time of sacrifice, blood samples were obtained by cardiac puncture with a heparinized syringe. After sacrifice, the livers were excised, blotted to remove excess blood and weighed. A thin slice of liver was fixed in 10% neutral buffered formalin for microscopic examination; the remainder of the liver was frozen and stored at -20°C until analyzed. Plasma and liver lipids were determined as previously described(6). Liver sections were prepared for microscopic examination as described(3).

Results. The results of both experiments are presented in Table I. Feeding lithocholic acid for 3 weeks resulted in depressed weight gain, elevated plasma cholesterol and lipid phosphorus levels, and increased liver size in both experiments. Microscopic examination of the livers revealed a proliferation of bile ductules in all chicks fed lithocholic acid for 3 weeks. This is in agreement with previous reports(3).

After being returned to a bile acid-free diet for 3 weeks in Exp. 1 and for 3 or 6 weeks in Exp. 2, all biochemical alterations induced by dietary lithocholic acid had been reversed. In Exp. 1, the liver size of chicks originally fed lithocholic acid was still significantly higher than that of the control chicks, but had regressed markedly. In Exp. 2, three or six weeks of feeding the basal diet completely overcame the difference in liver size which had been obvious after feeding lithocholic acid. Although body weight remained significantly below the control group for the lithocholic acid-fed chicks, the weight gain after removal of lithocholic acid from the diet was virtually identical to that of the control group.

In both experiments, removal of lithocholic acid from the diet of chicks reversed the profound morphological changes which had taken place in the liver. No lesions were observed in the livers of chicks which had been fed the

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TABLE I. Body Weight, Liver Size, Plasma and Liver Lipids of Chicks Fed Diets Supplemented with 0.2% Lithocholic Acid and a Lithocholic Acid-Free Diet.

Experimental group*	Body wt	Plasma		Liver†			
		Cholesterol	Lipid phosphorus	Size	Fat	Cholesterol	Lipid phosphorus
	(g)	(mg/100 ml)	(mg/100 ml)	(% body wt)	(%)	(mg/g)	(mg/g)
Exp 1‡							
3 Week:							
Control	256 ± 29§	131 ± 13	12.4 ± 1.8	2.2 ± .2	5.8 ± 1.9	4.3 ± 1.0	.80 ± .09
Treatment	178 ± 27	560 ± 159	45.6 ± 13.1	8.5 ± 2.0	5.0 ± .4	4.9 ± .1	1.02 ± .07
6 Week:							
Control	535 ± 48	115 ± 22	8.3 ± 1.0	2.6 ± .4	3.5 ± .7	4.6 ± 1.0	.86 ± .14
Treatment	469 ± 17	109 ± 13	8.7 ± 1.2	3.2 ± .3	3.7 ± .5	3.8 ± 1.1	.78 ± .12
Exp 2‡							
3 Week:							
Control	462 ± 66	146 ± 13	7.6 ± 1.4	2.6 ± .1	5.1 ± .2	5.0 ± .3	1.08 ± .07
Treatment	292 ± 46	548 ± 244	32.9 ± 15.9	8.5 ± 2.9	4.5 ± .2	5.9 ± .8	.98 ± .17
6 Week:							
Control	832 ± 51	174 ± 27	11.8 ± 2.2	2.6 ± .2	5.5 ± .1	3.8 ± .1	.80 ± .04
Treatment	662 ± 56	132 ± 18	9.2 ± 2.7	2.6 ± .2	5.8 ± 1.2	4.0 ± .2	.79 ± .06
9 Week:							
Control	1010 ± 96	140 ± 18	6.7 ± 2.8	1.9 ± .3	3.8 ± .3	5.4 ± .1	.79 ± .08
Treatment	853 ± 46	118 ± 17	6.9 ± 3.0	2.2 ± .2	4.2 ± .6	4.8 ± .7	.83 ± .15

* Treatment group fed 0.2% lithocholic acid-supplemented diet for 3 weeks followed by a lithocholic acid-free diet for 3 weeks (Exp 1) or 3 and 6 weeks (Exp. 2).

† All liver values expressed on wet weight basis.

‡ Values for Exp 1 are means for 8 chicks and for 5 chicks in Exp 2.

§ Mean ± standard deviation.

lithocholic acid-free diet throughout the study.

The mechanisms involved in production of the biochemical and morphological changes by lithocholic acid ingestion are not known. However, the data presented here demonstrate that these changes are dependent upon the continued presence of lithocholic acid in the diet.

Summary. Chicks fed lithocholic acid for 3 weeks had elevated plasma lipid levels, increased liver size and depressed body weights in comparison to chicks fed a bile acid-free diet. Feeding a lithocholic acid-free diet to chicks previously fed the bile acid completely reversed all of the biochemical changes. Lithocholic acid also induced a marked proliferation of bile ductules in the livers of chicks

which was reversed by feeding a lithocholic acid-free diet.

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