Cholic Acid: Adequate Stimulus for Hyperlipemia in Normal Fasting Rat.* (20135)

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Recent communications from this laboratory(1,2) have reported that experimental increase in the plasma content of cholic acid of the rat caused by either biliary obstruction or by intravenous injection of sodium cholate into normal fasting rats quickly leads to a hypercholesteremic state. Since cholesterol is only one of several lipids known to be increased in concentration in the plasma after obstruction of the bile duct(3,4), it was of interest to determine whether, and to what extent, various plasma lipid fractions of the normal intact rat changed after continuous intravenous injection of sodium cholate.

I. *Methods.* A. *Physiological.* Normal, male, Long-Evans rats were employed in this study. Both the 17 experimental and 9 control rats were operated upon under ether anesthesia and the right kidney removed. The unilateral nephrectomy was done in order to retard the renal excretion of the injected cholate. Then one end of a flexible polyethylene cannula (diameter: 0.011 inch) was inserted into the inferior vena cava of each animal via a lumbar tributary. The other end of the cannula was attached to a previously described apparatus(2) which allowed the continuous intravenous injection of 0.3 ml of fluid per hour. The abdominal incision then was closed and the rats were placed in individual Bollman cages(5). In this manner, the continuous intravenous injection of sodium cholate in Tyrode's solution (equivalent to 75 mg of cholic acid per 1 cc) was given to the experimental rats for 24 hours at a rate equivalent to 23 mg of cholic acid per hour. The control rats received an equal volume of Tyrode's solution. The rats were starved for 12 hours before operation and also during the experiment. Blood samples taken before, and 24 hours after, the beginning of the injection were analyzed for cholesterol, total lipid, phospholipid, neutral fat and cholic acid.

B. *Chemical.* Cholesterol in plasma was determined according to the method of Saifer and Kammerer(6) modified as reported previously(7). Phospholipid was determined according to the method of Fiske and SubbaRow (8) using the modifications of Stewart and Hendry(9). Plasma lipid phosphorus was converted to phospholipid by multiplying by the factor 25. Total plasma lipid was determined as described by Bragdon(10), and neutral fat was determined by difference. Plasma cholic acid (bile acids) was determined by absorption spectrophotometry, as reported in the modification by Wilken(11) of the bile acid method of Minibeck(12).

II. *Results.* The data are presented in Table I. It can be seen that, in the experimental group, each of the plasma lipids approximately doubled in concentration during the 24 hours in which sodium cholate was administered. In this sense, the hypercholesteremic effect of cholic acid is not a specific one, but produces a general and comparable increase in all plasma lipids.

III. *Discussion.* The data show that, under the influence of cholate, the animals become not only hypercholesteremic but also hyperlipemic without any significant change in the ratio of cholesterol to phospholipid in the plasma. In normal humans(4) the ratio of cholesterol to lipid phosphorus is constant despite normal variations in the concentrations of lipid fractions. At pathological levels of cholesterol, this ratio increases rapidly. It seems probable, therefore, that the cholate administered did not derange the lipid metabolism of the rat to such an extent that normal lipid interrelationships could no longer be maintained.

The parallel increase in each lipid class is consistent with a change in the binding properties of the plasma proteins for lipids, brought about by the injection of cholate. The result-

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TABLE I. Cholate Induced Lipemia.

	No. of rats	Avg wt, g	Total lipid, mg/100 cc	Total cholesterol, mg/100 cc	Phospholipid, mg/100 cc	Neut. fat, mg/100 cc	Cholate, mg/100 cc
			Before inj. of cholate				
A. Rats to be given cholate							
	17	247	175	49	47	79	2.1
Range:		211–302	100–276	37–60	13–90	14–164	1.8–2.2
S.E. mean			16	2	7	16	0.03
B. Control rats							
	9	239	145	45	—	—	—
Range:		213–263	114–228	33–65			
S.E. mean			12	4			
			24 hr after inj. of cholate				
A. Rats given cholate							
	17	247	353	106	108	142	19
Range:		211–302	214–650	78–151	34–286	25–285	7–35
S.E. mean			28	5	20	27	3
B. Control rats							
	9	239	154	62	—	—	—
Range:		213–263	124–210	54–72			
S.E. mean			9	3			

ing lipemia is not specific for one type of lipid, as might be the case if a specific chemical bond were established, but is indiscriminate, suggesting a nonspecific form of lipid adsorption. Such an alteration of the lipoproteins of blood so that they are capable of carrying and retaining more lipids has been offered as a hypothesis to explain the hypercholesteremic effect of both Triton WR 1339 (13), and of cholic acid(14).

IV. *Summary.* Experimental hyperlipemia was produced by intravenous injection of sodium cholate into normal fasting rats. The lipemia resulted from a comparable percentage increase in the concentration of each plasma lipid fraction, and not from preferential increase of any one particular plasma lipid.

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