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Received July 2, 1956. P.S.E.B.M., 1956, v93.

Inhibitory Effect of Chlorpromazine on Arterial Lipid Deposition in Cholesterol Fed Rabbits.* (22683)

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Reduced arterial lipid deposition in cholesterol fed rabbits has been achieved in a number of ways(1). Most of these inhibitory procedures cause weight loss, debilitation or other injurious effects. Chlorpromazine can be administered to animals for long periods of time without causing apparent impairment of health. Nevertheless, it is said to depress cellular activity, lower body temperature and arterial blood pressure, and, possibly, lower oxygen consumption of tissues(2). It is therefore reasonable to assume that this drug might alter cholesterol metabolism and influence the usual responses of rabbits to cholesterol feeding. The present experiment was designed to study the effect of chlorpromazine in this connection.

Method. Twenty-four albino rabbits weighing from 1810 to 2320 g initially were used. Nine received daily, 10 mg/kg intramuscular injections of chlorpromazine (Thorazine hydrochloride; Smith, Kline and French Laboratories)† and were fed rabbit food pellets containing 1% cholesterol(3). Nine rabbits were given equivalent injections of chlorpromazine but were fed the stock diet without added cholesterol. Six rabbits received only the cholesterol diet. The animals were given a daily ration of 100 g and food consumption was recorded daily. Body weights were determined every other week. Blood (9-10 cc) was drawn by ear vein initially and at the end of the 1st, 3rd, 6th, and 9th weeks. Free and esterified cholesterol, phospholipid, total lipid, proteins and sugar were determined on the serum as described previously(3). Determinations of serum bilirubin (Evelyn and Malloy) were also attempted but the levels were always too low to obtain quantitative readings. All animals survived and were sacrificed at the end of the tenth week. Complete gross and microscopic studies of the viscera were performed. The aortas were stained grossly with Sudan IV and the distribution of lipid plaques charted as previously described(3).

Results. Blood chemistry: Cholesterol levels rose progressively in all cholesterol fed animals but by the end of the third week it was apparent that the chlorpromazine group had significantly lower values (Table I). At the ninth week the total cholesterol of this group was only about one-half as high (Mean 985, Max. 1368, Min. 760 mg/%) as that of those fed the 1% cholesterol diet only (Mean 1834, Max. 2166, Min. 1535 mg/%). The ratio of free to esterified cholesterol did not change significantly in either group. Total cholesterol rose slightly and transiently in 8 of 9 chlorpromazine rabbits on stock diet reaching its peak during the third week. In these 8 rabbits the mean cholesterol was 322 mg/% (Max. 457, Min. 234). No explanation can be given for the failure of the 9th rabbit to show this change. It fell in all 8 to a mean level of 157 mg/% by the sixth week and was within normal limits at the ninth week. The free and ester fraction ratios were not altered during this temporary rise.

* This investigation was supported by Research Grant from National Heart Institute of N. I. H., Public Health Service.

† We wish to thank the Smith, Kline & French Laboratories for donating this preparation.

TABLE I. Total Serum Cholesterol in Cholesterol Fed and Chlorpromazine Treated Rabbits.

	Total serum cholesterol, mean mg %				
	Wk of exp.				
	0	1	3	6	9
Chlorpromazine only	61	68	293	157	95
Chlorpromazine and cholesterol	55	411	759	961	985
Cholesterol only	65	374	1140	1560	1834

Serum phospholipids (Table II) were also elevated in all cholesterol fed animals but to a lesser degree than serum cholesterol. The ratio of cholesterol to phospholipid was very similar in both the chlorpromazine and non-chlorpromazine treated, cholesterol fed rabbits. It varied from about 3:1 to 2:1 in most determinations. Phospholipid values were also slightly elevated in the third and sixth weeks in rabbits receiving only chlorpromazine but generally lagged behind the slightly elevated serum cholesterol. There was, however, no significant change in the ratio of these 2 lipid fractions. Neutral fat (calculated as previously described(4)) was slightly elevated in all cholesterol fed animals especially in the latter weeks of the experiment, varying from 0.48 g % to 0.82 g %. There was no significant difference between the chlorpromazine-treated and non-chlorpromazine treated cholesterol fed groups. Neutral fat levels remained low and remarkably constant throughout the experiment in the group receiving only chlorpromazine. Serum globulin tended to rise in all chlorpromazine treated rabbits but this was inconstant in degree. A few had reversed albumin globulin ratios on several occasions. Blood sugar and serum bilirubin were never significantly elevated.

Body weight and food consumption. All 24 rabbits consumed their entire daily 100 g food ration except on very few occasions when 10-20 g of food was left in the containers. None developed diarrhea. All animals gained weight consistently throughout the experiment. The mean weight gain in the group receiving both chlorpromazine and cholesterol was significantly less (58 g/wk) than in the groups receiving only chlorpromazine (103 g/wk) and cholesterol only (98 g/wk). All animals were in good health throughout the experiment and after the first 2 injections of chlorpromazine showed no obvious signs of sedation. Body temperature as measured intraperitoneally with a telethermometer during the eighth week of the experiment was not lowered significantly in chlorpromazine treated rabbits.

Necropsy findings. A striking inhibition of lipid deposition in the aortas, heart valves, and pulmonary arteries was noted in all but one of the cholesterol fed animals that received chlorpromazine (Fig. 1). In these 8 only a few thin patches of lipid were found chiefly in the aortic arch. One rabbit had fairly extensive plaques, but even in this animal the lesions were less extensive than in all but one of the group receiving cholesterol only. Such individual variation is a common finding in cholesterol feeding experiments in rabbits. Rabbits receiving chlorpromazine only had no gross vascular disease.

The livers of the rabbits receiving both chlorpromazine and cholesterol were smaller (mean wt 103 g) and less yellow than those that received cholesterol only (mean wt 125 g). They were only slightly larger and paler than the livers of animals that received only chlorpromazine (mean wt 95 g). Histologically, the liver cells of the chlorpromazine

TABLE II. Serum Phospholipid and Neutral Fat in Cholesterol Fed and Chlorpromazine Treated Rabbits.

Group		Wk of exp. 0	1	3	6	9
Chlorpromazine only	Phospholipid (mean mg %)	109	74	175	141	91
	Neutral fat (mean g %)	.35	.39	.45	.38	.42
Chlorpromazine and cholesterol	Phospholipid (mean mg %)	51	277	445	377	431
	Neutral fat (mean g %)	.38	.48	.60	.55	.55
Cholesterol only	Phospholipid (mean mg %)	67	210	496	600	771
	Neutral fat (mean g %)	.38	.43	.27	.61	.82

treated, cholesterol fed rabbits were less swollen and contained less finely dispersed lipid droplets than did those of the cholesterol fed animals that received no chlorpromazine. No evidence of liver cell damage or bile stasis was noted in any chlorpromazine treated rabbits. Enlargement and increased lipid storage was equally prominent in the adrenal cortices of both cholesterol fed groups. Histologic study of the other viscera revealed no significant changes that could be attributed to chlorpromazine administration. The mean heart weight of all 3 groups was approximately the same.

Discussion. It seems likely that the reduced amount of lipid deposited in the arterial walls in chlorpromazine treated, cholesterol fed rabbits is related to the lower levels of blood cholesterol observed in these animals as compared to the cholesterol fed rabbits that did not receive this drug. Except under special circumstances the degree of arterial lipid deposition in cholesterol fed rabbits varies directly with the degree of hypercholesterolemia in our own experience. There is no evidence that alterations in blood lipid interrelationships such as lowered cholesterol/phospholipid or cholesterol/neutral fat ratios played any role. It is also clear that malnutrition or general debilitation were not a factor in inhibiting arterial lipid deposition since the chlorpromazine treated animals remained in good health throughout the experiment. The degree of inhibition of lipid deposition was greater than that observed previously(3) in cortisone treated, cholesterol fed rabbits on a comparable regimen. If generalized depression of metabolism was involved such as occurs in hypothyroidism it might be expected that chlorpromazine would have accentuated hypercholesterolemia and enhanced arterial lipid deposition. This was obviously not the case. The failure of chlorpromazine treated, cholesterol fed rabbits to gain as much weight as the control groups, suggests the possibility that even though these animals consumed as much cholesterol, its absorption from the gut may have been reduced. However, the mean weight gain in the rabbits receiving only chlorpromazine was as great as in those fed cholesterol only.

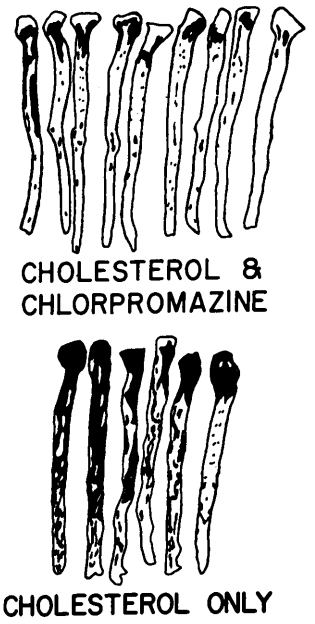

FIG. 1. Extent of aortic lipid deposition in rabbits fed cholesterol and inj. with chlorpromazine (upper row) compared with that in rabbits fed cholesterol only (lower row). Black areas represent portions of the intima covered by sudanophilic lipid deposits.

The inhibitory effect of chlorpromazine on hypercholesterolemia in these rabbits may have been mediated through the liver. Blood cholesterol levels are believed to be closely regulated by the metabolism of this lipid in the liver(5). To support this interpretation is the finding that the livers of the chlorpromazine treated animals failed to enlarge and to store as much lipid material as did those of rabbits that received only cholesterol. Chlorpromazine is known to produce jaundice occasionally and presumably some sort of liver injury in man although the exact mechanism has not been elucidated. The transient rise in serum lipids and globulin during the 3rd and 5th weeks in rabbits receiving only chlorpromazine was greater than ordinarily occurs in untreated rabbits and was therefore presumably due to the drug. These changes suggest that a brief period of metabolic readjustment may have occurred. This period corresponds somewhat to the time at which jaundice appears most frequently in man after chlorpromazine therapy is instituted. In some instances these attacks of jaundice in

man have been associated with moderate elevations of serum cholesterol(6). This has usually been explained on the basis of biliary stasis. There appears to be little evidence that chlorpromazine has any striking effect on the usual blood lipid levels found in man. Hollister and Kanter(7), however, report that it may be effective in lowering blood lipid levels of patients with essential hyperlipemia.

Summary. The blood cholesterol levels of cholesterol-fed rabbits given daily intramuscular injections of chlorpromazine fail to rise as high as in rabbits fed cholesterol only. Deposition of lipid in arteries and in the liver is also greatly reduced. Repeated injections of chlorpromazine in rabbits on stock diets may cause transient periods of mild hypercholesterolemia.

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Received July 18, 1956. P.S.E.B.M., 1956, v93.

Studies with 5-Bromouracil in Rodents and Dogs.* (22684)

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The growth inhibitory activity of 5-bromouracil was first observed in *Lactobacillus casei*(1). Later the substance was shown to be extensively incorporated in lieu of thymine in the deoxyribonucleic acids of bacteria(2, 3,4) and of a T_2 phage of *Escherichia coli*(4). Such findings suggest that the agent may be a thymine antagonist—a supposition supported by the near identity in size of the bromine constituent and of the methyl group of thymine (2). Anti-thymine actions may also be involved in the potentiation by 5-bromouracil of the inhibitory effects of folic acid antagonists in *L. casei*(5). Because of these findings, it has become of interest to ascertain whether 5-bromouracil might potentiate the action of amethopterin in the treatment of human leukemia.† With this in mind the work presented here was initiated as a preclinical study of the pharmacological actions of 5-bromouracil. During the study special interest arose in its catabolic fate *in vivo* because of the presence of crystalline deposits in kidneys or bladders of intoxicated animals. It is the primary purpose of this report to document this finding and to present information concerning the chemical nature of these deposits.

Procedure. Mice, rats and dogs were the subjects of the present investigation. The

* This investigation was supported in part by research grants (C 415 and C 471) from the National Cancer Institute of N. I. H., U. S. Public Health Service, (AT (C 30-1) - 910) from the Atomic Energy Commission, and funds from Amer. Cancer Soc. and the Elsa U. Pardee Foundation.

The authors wish to express their appreciation to Dr. F. S. Philips for his advice in preparation of this manuscript, and to acknowledge the technical assistance of B. B. Adams, B. Wheelock and O. Intrieri.

The 5-Bromouracil was purchased from the Dougherty Chemical Co., New York City.

† Personal communication from Dr. J. H. Burchenal.