attributed to the persistence of effective concentrations in blood and tissues.

Whether the presence of the methoxy groups is correlated to toxicological and chemotherapeutic characteristics of Madribon cannot yet be decided, although the influence of the methoxy group on the properties of sulfamethoxypyrimidine might allow such assumption by analogy.

Summary. A new sulfonamide, Madribon, exerted high protective activity in 12 experimental bacterial infections produced by gram-positive and gram-negative organisms. The frequency of drug administration could be reduced to a single dose in all 7 infections in which this technic was tried.

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Dietary Saponin and Plasma Cholesterol in the Chicken.*† (24371)

PAUL GRIMINGER AND HANS FISHER (Introduced by J. B. Allison)
Dept. of Poultry Science, Rutgers University, New Brunswick, N. J.

Considerable interest exists in factors influencing plasma cholesterol level. Hypercholesterolemia induced by cholesterol feeding is now a widely known phenomenon in the chicken. Recently, Newman and co-workers (1) have demonstrated a decrease in serum cholesterol (and also liver cholesterol) of chicks which were fed cholesterol and saponin simultaneously. Schoenheimer and Sperry (2) have shown that saponins have the property of forming stable complexes with cholesterol; therefore, a reduction of blood cholesterol due to feeding of saponin with a cholesterol containing diet might be expected. Blood cholesterol level of chicks however, can be significantly varied not only by dietary cholesterol, but also by other factors such as exercise (3), and a low level of protein in the diet (4,5). In the present report an attempt was made to evaluate the influence of saponin on endogenous hypercholesterolemia in chickens, induced by feeding low protein diets.

Methods. Three experiments were performed. In Exp. 1, male crossbred chicks were started on experimental diets at one day of age, and at 24 days blood was taken for plasma cholesterol analysis. In Exp. 2, newly hatched female cross-bred chicks were first kept on conventional starter mash for 1 week, and plasma analyzed for cholesterol after 16 and 23 days on the experimental diets. In Exp. 3, 1-year-old White Leghorn roosters were used, and their plasma cholesterol determined 10 and 20 days after the birds had been put on experimental diets. Composition of diets is given in Table I, and dietary additives to the different lots appear in Table II. Ten chicks or 5 adult birds were kept on each treatment in each experiment. Chicks were housed in electrically heated and thermo-

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TABLE I. Composition of Basal Diets.

Ingredient	%
Exp. 1 and 2	
Cerelose	56.05
Starch	20
Assay protein C-1	12
Salt mixture*	5.30
Non-nutritive fiber	3
Refined corn oil	3
Choline chloride	.20
Vitamin mixture*	.15
Vit. A, D and E concentrate*	.10
DL-methionine	.10
Glycine	.10
Total	100
Exp. 3	
Cerelose	84.85
Soybean oil meal (50% protein)	6.25
Refined corn oil	3
Salt mixture*	3
Dried distillers solubles	2.50
Choline chloride	.15
Vitamin mixture*	.15
Vit. A, D and E concentrate*	.10
Total	100

* Fisher and Johnson(6).

statically controlled battery brooders with raised wire floors, and mature birds in individual batteries. Feed and water were supplied *ad lib.*; weekly feed consumption was recorded for chicks and daily feed consumption for roosters. Blood was drawn for cholesterol analysis from the heart in the chick experiments and from the wing vein in the experiment involving mature birds. Plasma cholesterol was determined according to the method of Zlatkis *et al.*(7), as modified by Weiss and Fisher(8), using the stable iron reagent of Rosenthal and co-workers(9).

Results. In each of the 3 experiments the feeding of low protein diets (lot 1) was associated with increases in plasma cholesterol, when compared with lots fed conventional protein levels (lot 5) (Table II). Dietary saponin decreased blood cholesterol level in all cases. It should be pointed out that saponin decreased, as expected, not only cholesterol-induced hypercholesterolemia, but also endogenous hypercholesterolemia caused by feeding low protein diets. Considering the large standard errors, it might be argued that reductions in plasma cholesterol due to feeding of saponins are relatively small; it should be pointed out, however, that a trend was established by showing such differences in every experiment.

Feeding of saponin reduced weight gains of chicks (Table III); addition of cholesterol failed to overcome this reduction, though a tendency to diminish the depression may be present. Weight losses of roosters during the 20-day experimental period averaged 400 g for lots 3 and 4 *vs.* 280 g for lots 1 and 2. Saponin (lots 3 and 4) reduced feed intake, as compared with lots 1 and 2, by 34% in

TABLE III. Weights of Chicks in Experiments 1 and 2 at Time of Plasma Cholesterol Determination.

Lot	Exp. 1 24 days*	Exp. 2 23 days	Exp. 2 30 days
1	106 ± 9†	158 ± 9	188 ± 14
2	133 ± 11	151 ± 6	192 ± 8
3	72 ± 6	119 ± 4	140 ± 5
4	89 ± 8	126 ± 8	150 ± 14
5	280 ± 26	258 ± 8	351 ± 16

* Age of chicks.
† Mean wt (g) ± stand. errors of mean.

TABLE II. Dietary Additives and Plasma Cholesterol Values of Chickens.

Lot	Additives to basal*	Plasma cholesterol in mg %: Exp. 1 24 days†	Exp. 2 16 days	Exp. 2 23 days	Exp. 3 10 days	Exp. 3 20 days
1	None	253 ± 15‡	209 ± 11	228 ± 10	258 ± 17	297 ± 70
2	.3% cholesterol	348 ± 26	242 ± 13	250 ± 13	495 ± 85	579 ± 94
3	.6% saponin§	219 ± 21	181 ± 10	212 ± 10	227 ± 20	221 ± 18
4	.6% saponin & .3% cholesterol	295 ± 27	219 ± 18	229 ± 21	290 ± 80	376 ± 60
5	Protein‖	154 ± 11	135 ± 10	141 ± 7	220 ± 33	201 ± 22

* As % of total diet; replacing equal amounts, by wt, of cerelose.
† Days on experimental ration.
‡ Mean wt (g) ± stand. errors of mean.
§ Tannin-free saponin, S. B. Penick and Co.
‖ Exp. 1 and 2, 18% assay protein C-1, .2% DL-methionine, and .2% glycine. Exp. 3, 22.5% soybean oil meal (50% protein) and .1% DL-methionine.

Exp. 1, 22% in Exp. 2, and by 9% in the rooster experiment.

Discussion. In theory, saponins could exert their cholesterol-complexing action either inside the intestinal lumen or in the blood after their absorption. However, absorption of significant amounts of saponin would probably cause hemolysis(2,10) which was not apparent in these experiments. Furthermore, if complexing were to take place in the blood stream, cholesterol would be released from its complex with saponin during analysis and therefore would still be measured as plasma cholesterol. Hence, the reactions by which saponins reduce plasma cholesterol must occur not in the blood, but in the intestinal lumen. Peterson(11) suggested that an insoluble sterol-saponin compound is formed in the digestive tract when cholesterol and saponins are fed simultaneously. This could explain the depressing effect of dietary saponins on blood cholesterol, when the diet contains cholesterol. In an endogenous hypercholesterolemia and with a cholesterol-free diet, the cholesterol in the intestinal tract originates from biliary and intestinal secretions. Extending Peterson's theory to endogenous cholesterol, one might explain lowering of blood cholesterol levels, as observed in lots 3 in each experiment, by failure to reabsorb part of this cholesterol due to its unavailability after complexing.

Feed consumption data suggest that age might play a role in degree of depression of feed intake with dietary saponin. Chicks put on saponin diets at the age of one week showed less depression than chicks started on diets at one day, and adult roosters seemed to be least affected.

Summary. A study with growing chicks and adult roosters indicated that dietary saponin will depress blood plasma cholesterol previously elevated by feeding low protein levels in presence and absence of dietary cholesterol. It is suggested that complexing of saponin with cholesterol secreted in the intestinal lumen makes less cholesterol available for reabsorption from the intestinal tract.

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Host Resistance in Hemorrhagic Shock XV. Isolation of Toxic Factor from Hemorrhagic Shock Plasma* (24372)

HERBERT A. RAVIN, FRITZ B. SCHWEINBURG AND JACOB FINE
Yamins and Kirstein Laboratories for Surgical Research, Beth Israel Hospital and Dept. of Surgery, Harvard Medical School, Boston, Mass.

Previous reports from this laboratory describe several properties of hemorrhagic shock plasma which distinguish it from normal plasma, *viz.*: 1. Infusion of irreversible shock plasma is lethal to a reversibly shocked animal, whereas infusion of normal plasma restores normal peripheral blood flow and leads to recovery(1). 2. Shock plasma induces the local and general Shwartzman reaction in suit-

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