The rates of bile acid synthesis were calculated from bile acid pool size and turnover rates in the 2 groups. Daily synthesis of bile acids in normal mice was 0.56 mg; in cholestyramine-treated animals, 1.84 mg. Thus the diet supplemented with 1% cholestyramine caused a 3- to 4-fold increase in bile acid synthesis or excretion. Determination of fecal digitonide-precipitable sterols showed that normal mice excreted 5.54 ± 0.65 mg/day; cholestyramine-treated mice, 7.58 ± 1.14 mg/day.

These increases in bile acid and sterol excretion account for the increased mobilization rate of accumulated blood and liver cholesterol in the mouse. However, it is not possible to tell whether excretion of just one or of both fractions is responsible for the increase in cholesterol mobilization. Although there is evidence that accumulated liver cholesterol is eliminated only after conversion to bile acid(6,13), under certain circumstances accumulated liver cholesterol can be eliminated in the sterol fraction(6).

Summary. Dietary cholestyramine (MK 135) increased the rate of mobilization of blood and liver cholesterol in mice. The bile acid turnover rate in normal mice was 5 days, in mice treated with 1% cholestyramine, 1¼ days, or 4 times as fast. Bile acid pool size was 5.62 ± 1.09 mg in normals, 4.58 ± 0.65 in cholestyramine-treated mice. The pool was shown to consist almost exclusively of cholic acid and this spectrum was not altered by cholestyramine. The daily rate of synthesis of bile acids in normal mice was 0.56 mg, 1.84 mg in cholestyramine-treated mice. Digitonin precipitable fecal sterol excretion was 5.54 mg/day in normal mice, 7.58 mg/day in mice treated with 1% MK 135. No conclusion could be reached as to whether the increased rate of mobilization of accumulated cholesterol affected by MK 135 was due to the increased excretion of either the fecal bile acid or sterol fraction exclusively, or was attributable to the excretion of both these fractions.

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Effect of Sodium Salicylate on Magnesium Metabolism in the Rabbit.* (31279)

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Mg^{28} administered parenterally in a tracer dose is rapidly concentrated in tissues(1). Tissue uptake of this isotope is facilitated by

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the simultaneous administration of insulin and glucose(2), thyroxine(3), or 2,4-dinitrophenol(4). It has been hypothesized that the common denominator responsible for the effect of these agents on magnesium metabolism might be the uncoupling of oxidative phosphorylation.

Smith(5) has suggested that the uncoupling of oxidative phosphorylation may also be the mechanism responsible for the pharmacologic action of salicylate, one of the most commonly employed therapeutic agents. The present experiments were performed in order to study the effects of long- and short-term administration of sodium salicylate on magnesium metabolism in rabbits.

Experimental procedures. Animals. Normal domestic adult male rabbits, with initial body weights between 2 and 4 kg, were kept in individual stainless steel metabolism cages and were fed a stock diet of compressed pellets containing 0.115 meq/g of magnesium. Tap water, containing 0.2 meq/l of magnesium, was given without restriction.

Isotope. Mg^{28}, with a specific activity of more than 500 μc/meq Mg, was received as $MgCl_2$ in physiologic saline solution. After addition of 1 N NaOH to form a precipitate of $Mg(OH)_2$, the mixture was centrifuged and the supernatant fluid decanted off. The precipitate was dissolved with 1 N H_2SO_4, and a stock solution of Mg^{28} containing 0.01 to 0.02 meq/l of magnesium was made up.

Radioactivity assay. Samples of plasma and tissue were assayed for gamma-ray activity with a well-scintillation counter, a total of 10,000 counts being made on each sample. All determinations were corrected for physical decay.

Chemical determination of magnesium. Serum, urine, and tissue magnesium determinations were performed by atomic-absorption spectrophotometry(6).

Preliminary observations. The lethal intravenous dose of sodium salicylate was found to be 200 mg/kg.

Four rabbits were placed in individual plastic spirometers, and the oxygen consumption of each one was measured serially before and after the interscapular administration of sodium salicylate, 20 mg/kg, twice a day for 6 days. Each rabbit showed an increase in oxygen consumption following initiation of drug therapy, the maximum increase ranging from 21 to 58%.

Plan of experiments. Exp. 1. In 14 rabbits, external balance studies for magnesium were performed for 8 days. Each morning before the rabbits were fed, the body weight, total food consumption, and 24-hour urine volume of each animal were measured. Blood from the marginal ear vein was collected into heparinized test tubes. Findings obtained during the first 2 days were considered as control data. On the third day and for 5 days thereafter, each animal received 2 interscapular doses of sodium salicylate diluted in distilled water to a concentration of 40 mg/ml; each injection contained 20 mg of sodium salicylate per kg of body weight. On the eighth day, immediately after the first dose of sodium salicylate, 0.06 meq Mg tagged with Mg^{28} was injected intravenously into each animal. The animals were killed by exsanguination exactly 4 hours after the injection of Mg^{28}, and from each animal 2 aliquots of the tissues listed in Table I were prepared for radioactivity assay. Plasma was obtained at the same time.

Exp. 2. Six rabbits, averaging 2.1 kg in weight, were bled on day 1. On the next day each animal received sodium salicylate, diluted in distilled water to a concentration of 150 mg/ml and given intravenously in a dosage of 150 mg/kg; this was followed immediately by an intravenous injection containing 0.08 meq Mg tagged with Mg^{28}. Two hours later sodium salicylate was again given intravenously in the same dose as before. The animals were killed 4 hours after the injection of Mg^{28}, and their tissues were analyzed in the same manner as in Exp. 1.

The specific activities and relative specific activity of the tissues and plasma were determined as follows:

$$\text{Specific activity of tissue} = \frac{\text{cpm/g wet weight of tissue}}{\text{meq/kg wet weight}}$$

Specific activity of plasma =

TABLE I. Effect of Sodium Salicylate on Relative Specific Activity of Magnesium in Various Tissues 4 Hours After Intravenous Injection of a Tracer Dose of Mg^{28}.

	Relative specific activity (mean ± standard error)		
	6 control animals receiving no salicylate	14 animals given salicylate for 6 days, 40 mg/kg I.S. daily	6 animals given salicylate, 300 mg/kg I.V. within 2 hr
Bone cortex	.06 ± .004†	.09 ± .01	.09 ± .01
Bone marrow	.60 ± .15	1.25 ± .22	1.82 ± .24*
Brain	.11 ± .01	.19 ± .01*	.16 ± .01
Adrenal	.94 ± .17	1.13 ± .03	1.43 ± .26
Appendix	.69 ± .03	1.03 ± .10*	.85 ± .07
Heart	1.15 ± .05	2.05 ± .15*	1.30 ± .07
Kidney	1.16 ± .04	2.27 ± .15*	1.58 ± .13*
Liver	.65 ± .09	1.32 ± .17	1.05 ± .08*
Lung	.68 ± .04	1.36 ± .13*	.94 ± .08
Muscle	.06 ± .01	.11 ± .01	.08 ± .01
Pituitary	1.11 ± .25	1.19 ± .16	
Skin	.68 ± .02	1.22 ± .16	.92 ± .08
Spleen	.94 ± .15	1.07 ± .12	.96 ± .13
Stomach	.89 ± .04	1.59 ± .10*	.91 ± .11
Testis	.86 ± .12	1.07 ± .06	.83 ± .13
Serum (meq/l)	1.58 ± .095	1.81 ± .084	2.21 ± .163*

* Statistically significant when compared with control values. † Standard error.

$$\text{Relative specific activity} = \frac{\frac{\text{cpm/ml plasma}}{\text{meq/l plasma}}}{\frac{\text{specific activity of tissue}}{\text{specific activity of plasma}}}$$

Statistical methods. In the analysis of the balance data, each animal served as its own control. In the analysis of the tissue data, the mean value for each tissue was determined for both test and control groups. In both cases, the significance of the difference between the test and control values was determined by the "t" test, a "P" of less than 0.01 being considered significant(6).

Results. Exp. 1. During the 8 experimental days, the animals gained in weight from a mean value of 2.077 kg to 2.243 kg. According to the results of the external balance study, the daily intake of magnesium was equal to the amount excreted in urine and feces.

In the rabbits receiving salicylate, the following tissues showed a significant increase in relative specific activity (Table I); appendix, brain, heart, kidney, lung, and stomach. The magnesium content was significantly decreased in all of the tissues named above, as well as in the adrenal gland, bone, cortex, liver, muscle, skin, and testis (Table II).

Exp. 2. In the animals receiving 2 large doses of salicylate within 2 hours, there was a significant increase in the relative specific activity of the bone marrow, kidney, and liver. The magnesium content of the following tissues was significantly decreased: appendix, bone cortex, brain, heart, kidney, liver, and lung.

Comment. Administration of 300 mg of sodium salicylate within 2 hours resulted in a significant decrease in the magnesium content of many of the soft tissues as well as the bone cortex. Administration of smaller doses over a longer period results in even greater reduction in the tissue magnesium content. This facile depletion of tissue magnesium by salicylate is surprising, since animals maintained on a magnesium-deficient diet for a month still maintained a normal concentration of magnesium in the soft tissues(7). The increased relative specific activity in both of the present experiments is indicative of increased turnover of magnesium in many tissues, probably because of the depletion of the tissue store.

The efflux and increased turnover of magnesium were associated with increased oxidation. Recent studies on intermediary metabolism have revealed that salicylates, besides uncoupling oxidative phosphorylation, also affect transamination and dehydrogenation(8).

TABLE II. Effect of Sodium Salicylate on Tissue Mg Contents (meq/kg Wet Wt).

	6 control animals receiving no salicylate	14 animals given salicylate for 6 days, 40 mg/kg I.S. daily	6 animals given salicylate, 300 mg/kg I.V. within 2 hr
Bone cortex	319.9 ±10.00†	261.5 ±10.15*	271.3 ± 8.65*
Bone marrow	10.8 ± 1.46	6.2 ± 1.01	6.9 ± .90
Brain	12.6 ± .35	9.2 ± .19*	9.9 ± .38*
Adrenal	12.5 ± 1.32	8.4 ± .37*	13.3 ± 1.91
Appendix	20.3 ± .59	14.2 ± .73*	16.2 ± .59*
Heart	15.4 ± .24	11.3 ± .56*	13.4 ± .31*
Kidney	16.5 ± .42	10.7 ± .43*	11.9 ± .48*
Liver	16.8 ± .60	9.5 ± 1.01*	14.0 ± .59*
Lung	11.5 ± .43	7.5 ± .39*	9.2 ± .29*
Muscle	21.6 ± .84	15.0 ± 1.01*	19.9 ± .22
Pituitary	15.3 ± .85	15.0 ± 1.37	
Skin	5.2 ± .37	2.8 ± .16*	4.9 ± .30
Spleen	17.5 ± 1.70	13.7 ± .65	17.0 ± 2.55
Stomach	20.3 ± .62	13.7 ± .81*	17.1 ± 1.48
Testis	11.8 ± .34	9.4 ± .44*	10.3 ± .50

* Statistically significant when compared with control values. † Standard error.

The exact enzymatic mechanisms affecting magnesium metabolism remain to be established.

Summary. Intravenous administration of sodium salicylate, 150 mg/kg twice within 2 hours to normal rabbits, resulted in a significant decrease in the tissue magnesium content of bone cortex, brain, appendix, heart, kidney, liver, and lung. The turnover of magnesium, as measured by Mg^{28}, was increased in the bone marrow, kidney, and liver.

In animals given sodium salicylate in daily doses of 40 mg/kg for 6 days, tissue magnesium content was significantly decreased in bone cortex, brain, appendix, adrenal, heart, kidney, liver, lung, muscle, skin, stomach, and testis. The relative specific activity was increased in the brain, appendix, heart, kidney, lung, and stomach. In rabbits, sodium salicylate depletes the tissues of magnesium and increases the turnover of this element in the body.

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Evidence for the Pentose Cycle in *Nocardia corallina*.* (31280)

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Nocardia corallina is a typical actinomycete which undergoes cyclic morphological changes during normal growth; the mechanisms behind these changes are unknown. is to search for answers at the metabolic level. One approach toward eventual explanations

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