more research is needed to (a) find why zinc deficiency causes an "arthritic" type syndrome in chicks fed soy protein, and how histamine protects against it, and (b) determine if histamine will counteract other "arthritic-like" or "perosis-like" defects.

Summary. Histidine at 1.0% and 2.0% of the diet, or histamine at 0.2% of the diet prevented the "arthritic-like" or "perosis-like" syndrome in zinc-deficient chicks fed soy protein diets, while having little or no effect on other symptoms of zinc deficiency. Other histidine metabolites and related compounds such as urocanic acid, β-imidazole acetic acid, imidazole and thiolhistidine had little or no effect on the zinc-deficient chick. A direct histidine-histamine relationship is postulated.

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Effect of Mercuhydrin Alone and in Conjunction with Ammonium Chloride On Radiostrontium Excretion in Man.* (31938)

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As the time after ingestion or inhalation of radiostrontium increases, an increasing fraction of radiostrontium that entered the body is deposited in the skeleton, where it becomes less accessible for removal. Orally administered ammonium chloride which produces both acidosis and diuresis has been shown to increase urinary excretion of calcium and radiostrontium at time of injection of the tracer(1-3) and as late as 2 weeks thereafter(2). To determine the effect of enhanced diuresis on radiostrontium excretion, the diuretic agent mercuhydrin, which potentiates the diuretic action of ammonium chloride(4) but does not cause acidosis, was used alone and in conjunction with ammonium chloride in studies performed under constant conditions in man.

Materials and methods. Six patients were studied on the Metabolic Research Ward where they received a constant, analyzed low calcium diet containing an average of 200 mg calcium and 750 mg phosphorus per day. Four of the patients received tracer doses of Sr^{85} intravenously (Patients 1-4) and 2 patients orally (Patients 5 and 6). Nine studies were performed on the effect of mercuhydrin on Sr^{85} excretion and 5 on the effect of mercuhydrin used in conjunction with ammonium chloride (Table I). Mercuhydrin, 2 ml, was injected intramuscularly on 1-3 days at time intervals ranging from 1 to 21 days following Sr^{85} administration. Ammonium chloride and mercuhydrin were used together at time intervals ranging from 4 to 27 days following Sr^{85} administration; 9 g ammonium chloride were given orally in divided doses per day

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for 4 or 5 days and mercuhydrin (2 ml) was injected intramuscularly on the last day of ammonium chloride administration.

A single dose of $Sr^{85}Cl_2$ (0.1-0.4 μc/kg) was given intravenously or orally on the first day of each study as previously described(1). Plasma levels of Sr^{85} were determined at 1, 4, 8 and 24 hours following injection or ingestion of Sr^{85}, daily for 6 days, and 3 times per week thereafter. Urinary Sr^{85} excretions were determined daily on aliquots of 24-hour urine collections and each stool specimen was radioassayed for Sr^{85}. Urinary calcium was determined daily, fecal calcium on aliquots of 6-day stool pools by a modification of the method of Shohl and Pedley(5).

Results. The plasma levels of Sr^{85} decreased in a similar manner in all patients following intravenous injection of Sr^{85}, the plasma curve of Patient 3 being typical of that of the other patients (Fig. 1), except for Patient 2 in whom the Sr^{85} plasma level remained considerably higher throughout the study.

The effect of intramuscular injections of mercuhydrin given on the day of intravenous injection of Sr^{85} and on 2 days thereafter is shown in Fig. 2 (Patient 1). Urinary Sr^{85} excretion was higher on the first day of mercuhydrin injection than on the first day of the control study, 15.0% *versus* 11.4% of the administered dose, respectively, while Sr^{85} excretion was only slightly higher on the second day of mercuhydrin injection than in the control study. On the third day of mercuhydrin administration this excretion became even lower. Urinary calcium excretion increased markedly on the first day of mercuhydrin injection but decreased on the subsequent 2 days of administration of this diuretic. Following discontinuation of mercuhydrin, urinary excretions of Sr^{85} and of calcium decreased to levels below those in the control study. Sr^{85} plasma levels on the days of mercuhydrin injections were similar to those of the control study (Fig. 3).

Thirteen days following oral administration of Sr^{85}, mercuhydrin increased urinary Sr^{85} excretion; however, on several days thereafter the urinary Sr^{85} excretion had distincly decreased (Fig. 4, Patient 5). The changes in Sr^{85} excretion during and after mercuhydrin administration were associated with corresponding changes in urinary calcium excretion. However, urinary calcium excretion increased 10-fold while urinary Sr^{85} excretion was only doubled as compared to the excretion values in the control study. These results were re-

TABLE I. List of Patients Studied.

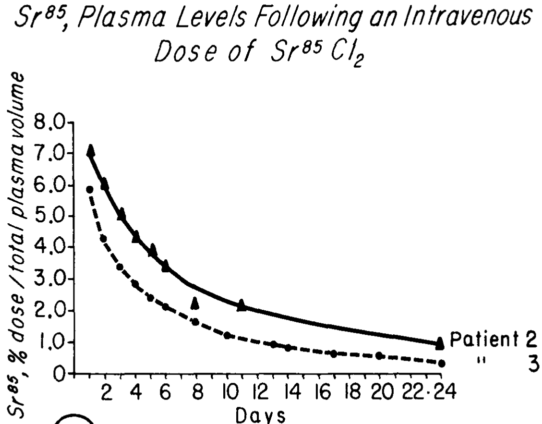
Patient	Age, sex	Diagnosis	Sr^{85} administration: Route	No. of tracers	Agents used*	No. of studies	Study days	Days after Sr^{85} tracer
1	64, ♀	Osteoporosis	Intrav.	2	Mercuhydrin, i.m.†	1	3	1–3
2	60, ♂	Klinefelter's syndrome	"	1	Mercuhydrin, i.m. NH_4Cl, p.o.† and mercuhydrin, i.m.	1 1	1 5	16 27–31
3	77, ♀	Osteoporosis	"	1 1	Mercuhydrin, i.m. NH_4Cl, p.o. and mercuhydrin, i.m.	3 1	1, 1, 1 4	11, 16, 21 22–25
4	51, ♂	Peripheral neuropathy	"	1	NH_4Cl, p.o. and mercuhydrin, i.m.	1	5	4–8
5	72, ♂	Congestive heart failure	Oral	2 2	Mercuhydrin, i.m. NH_4Cl, p.o. and mercuhydrin, i.m.	3 2	1, 1, 1 4, 4	9, 13; 13 12–15, 15–18
6	70, ♀	Myxedema	"	1	Mercuhydrin, i.m.	1	1	14

* Mercuhydrin (2 ml) was given by intramuscular injection. Ammonium chloride (9 g/day) was given orally for 5 days and mercuhydrin (2 ml) was injected intramuscularly on last (5th) day of NH_4Cl administration. Patient 3 received ammonium chloride for 4 days.

† i.m. = intramuscularly; p.o. = orally.

produced in the same patient in a second study.

Table II shows the results obtained with intramuscular injections of mercuhydrin in the remaining patients. Mercuhydrin led to a considerable increase in urinary Sr^{85} excretion only in Patient 2, from 0.45% to 1.19%, while the increase in Sr^{85} excretion was only slight in the remaining 7 studies (Patients 3, 5 and 6), the difference in urinary Sr^{85} excretion between the day prior to mercuhydrin injection and the day of injection of the



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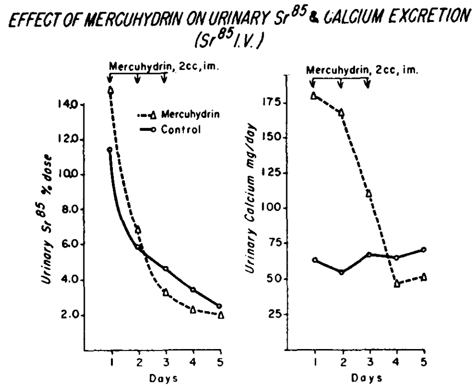

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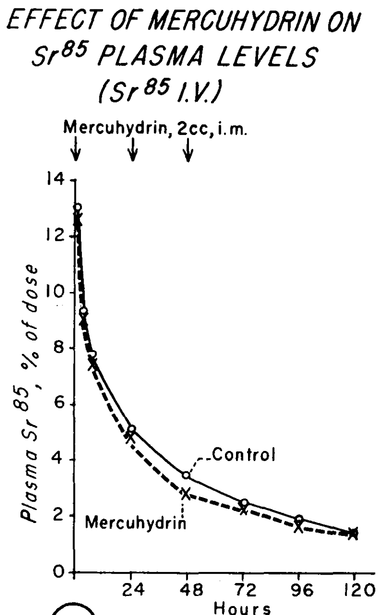

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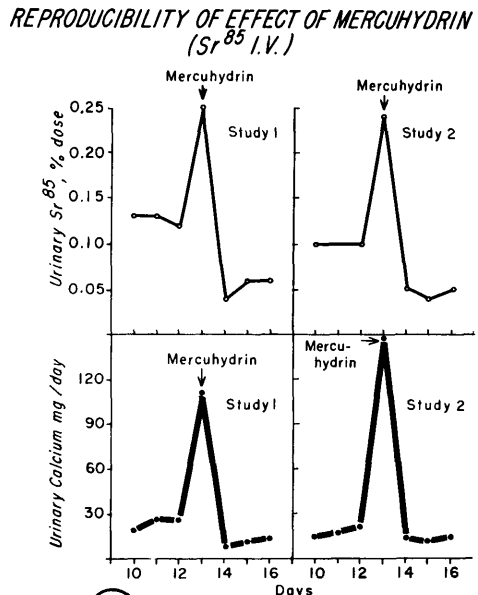

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FIG. 1. A single tracer dose of Sr^{85} was injected intravenously on 1st day of study (Patients 2 and 3).

FIG. 2. Sr^{85} was injected intravenously on 1st day of control and experimental studies. Mercuhydrin, 2 cc, was given by intramuscular route on first 3 days of experimental study (Patient 1).

FIG. 3. Sr^{85} was injected intravenously on 1st day of control and experimental studies. Mercuhydrin, 2 cc, was given by intramuscular route on first 3 days of experimental study (Patient 1).

FIG. 4. A single tracer dose of Sr^{85} was given orally on 1st day of each of the 2 studies. An intramuscular injection of 2 cc mercuhydrin was given on 13th day after oral administration of Sr^{85} in each study (Patient 5).

TABLE II. Effect of Mercuhydrin on Sr^{85} Excretion.

Patient	Day of mercuhydrin injection*	Sr^{85} (% dose retained)†	Urinary Sr^{85} (% dose): Pre-mercuhydrin days 1	2	3	Day of mercuhydrin injection	Post-mercuhydrin days 1	2	3
2‡	16	44.6	.44	.51	.45	1.19	.20	.26	.25
3‡	11	29.4	1.84	1.49	1.47	1.57	.73	.89	.74
	16	22.8	.89	.74	.84	.82	.49	.55	.45
	21	17.0	.39	.34	.40	.52	.25	.27	—
5§	13	5.0	.13	.13	.12	.25	.04	.06	.07
	13	5.8	.10	.10	.10	.24	.05	.04	.05
	9	33.9	.15	.13	.10	.22	.05	.04	.06
6§	16	3.7	.21	.20	.19	.28	.05	.08	.09

* Days following administration of tracer dose of Sr^{85}.
† Before injection of mercuhydrin.
‡ A single tracer dose of Sr^{85} was given intravenously on 1st day of study.
§ A single tracer dose of Sr^{85} was given orally on 1st day of study.

diuretic in these patients being significant, $P<0.01$. In all patients, however, urinary Sr^{85} excretion decreased sharply on the day after the mercuhydrin injection and remained at lower levels for several days than prior to administration of this diuretic.

Ammonium chloride administered orally from the 15th-18th day after oral ingestion of Sr^{85} resulted in a gradual increase in urinary Sr^{85} and calcium excretion on the first 3 days of this treatment. Excretion of both Sr^{85} and of calcium was highest on the 4th day of ammonium chloride administration when an injection of mercuhydrin was also given intramuscularly (Fig. 5, Patient 5). The increase in urinary calcium excretion was particularly striking on that day, the calciuria having gradually increased from 10 mg/day before treatment to 100 mg on the third

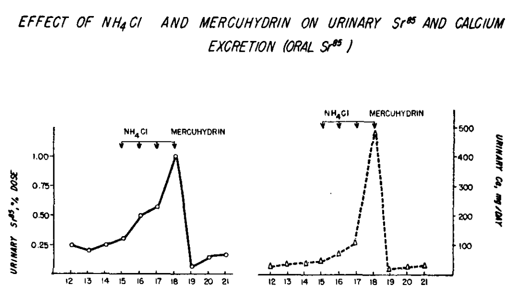

FIG. 5. Ammonium chloride (9 g/day) was given orally on 4 consecutive days starting on 15th day after oral administration of Sr^{85}. A single intramuscular injection of mercuhydrin (2 cc) was given on last (4th) day of ammonium chloride administration (Patient 5).

day of ammonium chloride administration, and rising sharply to 480 mg/day on the 4th day, when both ammonium chloride and mercuhydrin were administered.

The effect of the combined use of ammonium chloride and mercuhydrin in studies performed from the 4th-27th day after Sr^{85} administration is shown in Table III. During ammonium chloride administration, urinary Sr^{85} excretion increased gradually in 2 of the 4 patients (Patients 2 and 5), while this increase was only minimal in Patients 3 and 4, and then appeared to reach a plateau. Intramuscular injections of mercuhydrin given on the last day of ammonium chloride administration increased urinary Sr^{85} excretion further in 3 of 5 studies (Patients 2 and 5). In all cases, however, urinary Sr^{85} excretion decreased to very low levels following discontinuation of the combined use of ammonium chloride and mercuhydrin. The total Sr^{85} excretion on the days of this treatment ranged from 0.98% to 11.5% of the administered dose, corresponding to 16.0%-85.5% of the dose of Sr^{85} retained in the body (Patients 2-4 and first study of Patient 5). In the second study of Patient 5, the cumulative urinary Sr^{85} excretion was not expressed as percent of the retained dose because of very low Sr^{85} retention following oral administration of Sr^{85} (3.9% of the administered dose).

Mercuhydrin alone considerably increased the urine volume; when mercuhydrin was

TABLE III. Effect of Mercuhydrin and NH_4Cl on Sr^{85} Excretion.

Patient*	1st day of NH_4Cl admin.†	Sr^{85}, % dose retained‡	Urinary Sr^{85} (% dose): Days before treatment 1	2	3	Days of NH_4Cl and mercuhydrin§ 1	2	3	4	5	Post-treatment days 1	2	3	Total Sr^{85} excretion: % Admin. dose	% Retained dose
2	27	33.9	.20	.17	.15	.24	.38	.49	.73	.99	.11	.33	.32	2.83	8.3
3	22	16.0	.45	.50	.45	.46	.58	.60	.59	—	.29	.34	.38	2.23	13.8
4	4	85.5	4.90	3.30	2.20	2.45	2.66	2.52	2.07	1.84	1.05	.85	.79	11.54	12.2
5	12	31.1	.04	.06	.09	.05	.12	.17	.64	—	.06	.04	.07	.98	3.2
	15	3.9	.25	.24	.24	.29	.46	.56	1.00	—	.06	.15	—	2.31	—

* Patients 2, 3 and 4 received a single tracer dose of Sr^{85} intravenously at start of study, and Patient 5 received a tracer dose of Sr^{85} orally at start of both studies.
† Days following injection or ingestion of tracer dose of Sr^{85}.
‡ Before administration of NH_4Cl.
§ NH_4Cl (9 g/day) was given on each day; a single intramuscular injection of mercuhydrin (2 ml) was given on last day of NH_4Cl administration.

given on the last day of ammonium chloride administration the increase in urine volume was particularly marked (Tables IV and V). For instance, the combined use of the two drugs produced a diuresis of more than 7 liters in 2 separate studies in Patient 5 and of about 4 liters in Patient 2 (Table V), compared to a diuresis of 3 to 3½ liters induced by mercuhydrin alone (Table IV).

Fecal Sr^{85} excretion did not increase when mercuhydrin was used alone or in conjunction with ammonium chloride.

Discussion. Mercuhydrin, although effective as a diuretic, and resulting in an increase in urinary calcium excretion, is of little value in enhancing the excretion of radioactive strontium in man. The increase in urinary Sr^{85} excretion was variable and small in most studies although the difference between urinary Sr^{85} excretion on day prior to and on the day of mercuhydrin administration was significant, $P<0.01$. This increase was followed by a marked rebound decrease in urinary Sr^{85} excretion on the day after mercuhydrin injection which may be due to an attempt of the body to compensate for the loss of calcium induced by the diuretic, the decrease in calciuria being associated with a decrease in urinary Sr^{85} excretion. Enhancement of Sr^{85} excretion did not appear to depend on the amount of Sr^{85} retained in the body but may depend on the availability of radiostrontium for removal. For instance, the increase in Sr^{85} excretion induced by mercuhydrin in Patient 2 after 2 weeks was probably a result of the high Sr^{85} level in plasma (Fig. 1) which may have been associated with a relatively high Sr^{85} concentration in soft tissue.

Although the increase in Sr^{85} excretion during ammonium chloride administration was only slight, this excretion represented a greater increase than it seemed, since the urinary Sr^{85} excretion was very low in most cases prior to ammonium chloride administration and was decreasing steadily depending on the urinary calcium excretion of the individual as previously reported(1-3). Also, when treatment was started as late as 3-4 weeks after intravenous injection of Sr^{85}, the major portion of the administered Sr^{85} had

TABLE IV. Urine Volumes Before and During Administration of Mercuhydrin.

	Urine volume (ml/day)						
	Pre-days			Mercuhydrin	Post-days		
Patient	1	2	3	(2 ml/day)*	1	2	3
2	1250	1710	1860	2890	1940	1820	1840
5	1920	2040	1860	2950	2750	2465	1280
	2250	2165	1770	3280	1740	2080	1800
	1840	1950	1740	3660	1560	1800	1760

* A single intramuscular injection of mercuhydrin was given on this day.

TABLE V. Effect of Ammonium Chloride and Mercuhydrin on Urine Volumes.

	Urine volume (ml/day)										
	Control days			NH_4Cl and mercuhydrin days					Post-days		
Patient	1	2	3	1	2	3	4	5	1	2	3
2	1920	1840	1760	1960	1720	1920	2145	3880*	1180	1360	1480
5	2000	2060	1940	2080	2480	2500	7400*	—	1680	2100	1970
	1910	2060	1840	1280	2230	2700	7310*	—	2160	1900	2085

* Last day of ammonium chloride administration (9 g/day). Mercuhydrin (2 ml) was injected intramuscularly on that day. Patient 2 received ammonium chloride on 5 consecutive days, Patient 5 on 4 consecutive days.

been excreted. The very marked diuresis induced by the combined use of ammonium chloride and of mercuhydrin resulted in a striking and reproducible increase in urinary calcium excretion in two separate studies in Patient 5 but in a relatively small increase in urinary Sr^{85} excretion. In general, the use of both ammonium chloride and mercuhydrin did not lead to greater excretion of radiostrontium than ammonium chloride alone(1). In addition, mercuhydrin diminished the effect of ammonium chloride by inhibiting urinary excretion of Sr^{85} and of calcium following discontinuation of this combined therapy. In contrast, following discontinuation of ammonium chloride alone the urinary excretions of both calcium and Sr^{85} remained high for several days probably because of the slow recovery from metabolic acidosis induced by ammonium chloride(1,2).

The differences in effect of mercuhydrin and of ammonium chloride on Sr^{85} excretion are probably due to differences in the mechanism of action of the two diuretics(6-9). Mercurials lead to a decrease in renal tubular reabsorption of water; the latter has been shown to be independent of the tubular reabsorption of strontium in stop-flow studies in dogs(10). The effect of ammonium chloride on radiostrontium excretion is probably due to its production of metabolic acidosis (1,2), which leads to gradual dissolution of the calcium phosphate of bone(11) which is basic and acts as a buffer. Also, acidosis may result in removal of some calcium from soft tissues where it may be loosely bound to protein. During removal of calcium from bone and/or soft tissue, the Sr^{85} which has been deposited in these areas is returned to the general circulation, from which it is removed by the diuretic action of either ammonium chloride or mercuhydrin.

Summary. The diuresis induced by intramuscular injections of mercuhydrin at varying time intervals following intravenous or oral administration of Sr^{85} was associated with an inconsistent and only slight increase in urinary Sr^{85} excretion despite a concomitant increase in urinary calcium excretion. Following the discontinuation of mercuhydrin there was marked inhibition of the urinary excretion of Sr^{85} and calcium. Further increase in urine volume induced by mercuhydrin used in conjunction with ammonium chloride resulted in a marked increase in urinary calcium but not in further enhancement of Sr^{85} excretion. Mercuhydrin actually diminished the effect of ammonium chloride by inhibiting radiostrontium excretion following discontinuation of this combined therapy. This re-

bound inhibition of Sr^{85} excretion contrasts with the sustained increase in Sr^{85} and calcium excretion following the discontinuation of ammonium chloride.

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Epinephrine Induced Dermal Necrosis in Endotoxemia: Attempts to Define Response on an Immunological Basis.* (31939)

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Rabbits given endotoxin intravenously develop hemorrhagic necrosis at the site of a subsequent intradermal injection of epinephrine. Resistance to the local effect of epinephrine has followed an 8-day course of endotoxin administered intravenously(1). Because the nature of acquired resistance to endotoxin is poorly understood, the present study involved attempts to explain along immunological lines the hemorrhagic dermal necrosis resulting from endotoxin and epinephrine. Since active immunization of rabbits with endotoxin prevents dermal necrosis by epinephrine it appeared feasible to attempt a similar resistant state in susceptible rabbits by the transfer of whole serum or serum fractions from endotoxin-resistant rabbits. It has been reported that the pyrogenic response to endotoxin could be prevented in susceptible rabbits by the transfer of serum fractions from resistant animals(2). In addition, it was desirable to reexamine the reproducibility of the hemorrhagic response to epinephrine in rabbits given endotoxin, and the duration of resistance to epinephrine in animals given repeated doses of endotoxin.

Materials and methods. One kg female New Zealand white rabbits, 6-8 weeks old, were used throughout. A single lot of *Escherichia coli* endotoxin prepared in our laboratory, as previously described, was used in all experiments(3). For challenge, the desired quantity of endotoxin was injected into a marginal ear vein. Immediately thereafter, 0.1 ml epinephrine 1:1000 (aqueous) was injected intradermally into the rabbit's shaved flank. The site of epinephrine injection was examined at 24 hours for hemorrhagic necrosis. Negative reactions consisted of a 3 $\times$ 3 mm white or pink spot. Positive (+) reactions appeared as larger patches of purple discoloration; reactions measuring 6 cm^2 or greater were considered strongly positive (++). *Active desensitization.* Rabbits were given daily injections of endotoxin into the marginal ear vein as previously described(1): 1 μg days 1-4, 10 μg days 5-8. All animals were challenged 24 hours following the final injection. Serum was obtained by heart puncture just prior to challenge and frozen. Control animals were given either no injections prior to challenge or injections containing equivalent volumes of sterile, non-pyrogenic

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