Chronic Toxicity of Thiomerin Compared to Other Mercurial Diuretics.* (17956)

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Mercurial diuretics when administered intravenously offer the definite clinical hazard of producing acute cardiac effects. Attempts to find compounds to minimize or eliminate such reactions led to the clinical introduction of N-(gamma-carboxymethyl-mercapto-mercuri-beta methoxy)propyl camphoramic acid (Thiomerin)(1-6). The initial study of its toxicity was based on acute cardiac effects in which a ratio of 1:160 was expressed in comparison to Mercuhydrin(7). Some of the recent clinical reports concerning Thiomerin indicate that this ratio may be considered as the chief criterion of toxicity(1-3), although other workers acknowledge that it is but one indication of Thiomerin toxicity, *viz.*, its acute cardiac effect(4-6). Other criteria of toxicity obviously should be determined and considered for mercurial diuretics, compounds which are characterized by their rather prolonged action on the kidney. In this respect an increased incidence of systemic reactions to Thiomerin has been mentioned in one clinical report(1). Further, in mice, the intravenous lethal dose of this compound over a 4 day period was found to be of the same order as other diuretics(8). Handley *et al.*(9) demonstrated greater toxicity in the dog with a test of actual renal function, glomerular filtration as determined by creatinine clearances, and by the fact that Thiomerin caused a greater number of deaths. Inasmuch as Thiomerin when administered parenterally causes less local reaction than certain other diuretics(10), the question arose as to whether or not the simple addition of a monothiol compound, *e.g.*, Sodium Thioglycollate, to Mercurophylline or Mercuhydrin would decrease the local toxic reaction of these mercurials. Such a mixture would be similar in composition to Thiomerin since the latter is essentially Mercurophylline Injection U.S.P., in which the theophylline has been replaced by sodium mercapto-acetate(4). In referring to the work of Long and Farah(11), Lehman suggested that mixing a thiol with a mercurial previous to injection might reduce the toxicity of the latter.

The present study was initiated when it was found that mixing Mercurophylline with Sodium Thioglycollate caused a marked increase in the LD_{50} of the mercurial after subcutaneous administration. This report deals with the toxicity of Thiomerin in comparison with other mercurial diuretics and the mixture of Sodium Thioglycollate with Mercurophylline, after subcutaneous or intravenous administration in rats, and with the distribution and excretion of the mercury in the animal.

Methods. Sprague-Dawley albino rats were used as the experimental animal. The drugs

* The animals and supplies used in this study were made available by the Kremers-Urban Co. of Milwaukee.

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TABLE I.
Toxicity in Rats of Mercurial Diuretics Following a *Single* Subcutaneous Administration.

Dosage, total mercury, mg	Mercuhydrin	Mercurophylline			Thiomerin
		Plain	With benzyl alcohol	+ Na thioglycollate	
8.	5/5	10/10	0/10	5/5	12/12
4.	14/20	0/20	0/20	20/20	20/20
2.	0/10			5/20	10/10
1.5					5/15
1.					0/10

The ratio is expressed as: No. of rats killed/No. of rats injected.

tested were Meralluride (Mercuhydrin), Mercurophylline,† Mercurophylline with 2% benzyl alcohol,† Thiomerin, and the combination of equal parts of Mercurophylline† and a 6% aqueous solution of Sodium Thioglycollate. All of the drugs were used on the basis of a mercury content of 40 mg/cc. Inasmuch as Thiomerin is recommended for subcutaneous administration, the other compounds also were given by this route. Similarly, Thiomerin was tested after intravenous injection in order to compare its toxicity with that of mercurials commonly administered in this manner. The compounds were given as *single injections* in the range of doses indicated in Tables I and II. Subcutaneous injections were made into the flank and, with the exception of the doses of Thiomerin containing 1.5 mg and 1.0 mg of mercury, were not diluted before injection. For the intravenous injections the drugs were diluted to a total volume of 2 cc. Acute deaths were considered to be those that occurred during or within the first five minutes after completion of the injection. Delayed deaths were those that occurred after this initial period but in 7 days or less.

Thiomerin and Mercurophylline were used for studies of excretion and distribution of mercury in rats after subcutaneous administration. Dilutions of the 2 drugs were prepared so that 0.5 cc should contain 1.5 mg of mercury. This amount was injected and the rats placed in metabolism cages arranged for collection of urine and feces, as described by Harned, Cunningham and Gill (12). No food was available to the animals but water was permitted *ad lib.* Urine was collected during the survival of the animals which in no instance exceeded 4 days. The mercury content of the kidneys, the urine, the feces, and the carcass of the rat was determined by the dithizone method described by Kozelka(13).

Results. The results of single subcutaneous injections are presented in Table I. While no attempt was made to adjust the subcutaneous dosages to an exact mg/kg basis, the animals tested fell into two weight groups (125 to 175 g or 300 to 350 g) and representative numbers of each group were treated with each drug. It is evident that Mercurophylline with benzyl alcohol was the least toxic of the substances administered subcutaneously since there were no deaths in the group which received the dose of 8 mg of mercury. Thiomerin was the most toxic since the single injection which contained only 2 mg of mercury killed all the animals of that group. The sites of injection were examined for evidence of irritation and it was noted that Thiomerin and the combination of Mercurophylline and Sodium Thioglycollate did not produce local irritation in any dose used. The results following the intravenous administration of a single dose of the compounds are presented in Table II. In this series Mercurophylline was the least toxic, while Thiomerin again was the most toxic. Acute deaths occurred with injections of Mercurophylline and Mercurophylline with benzyl alcohol. In fact, the greatest propor-

† Differs from Mercurophylline Injection U.S.P. in that each cc contains an excess of 10 mg of theophylline.

12. Harned, B. K., Cunningham, R. W., and Gill, Edna R., *Science*, 1949, v109, 489.

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TABLE II.
Toxicity in Rats of Mercurial Diuretics Following a *Single* Intravenous Injection.

Dosage, total mercury, mg/kg	Mercuhydrin		Mercurophylline							
			Plain		With benzyl alcohol		+ Na thioglycollate		Thiomerin	
	Acute*	Delayed	Acute	Delayed	Acute	Delayed	Acute	Delayed	Acute	Delayed
36			10	1						
			11							
32			3	1						
			4							
24	0	6	7	5	42	19	0	10	0	10
	6		22		73		10		10	
16			2	1	12	1				
			12		20					
12	0	10			9	1	0	10	0	10
	10				20		10		10	
10	0	9								
	10									
8	0	5					0	8	0	10
	10						10		10	
4	0	0					0	0	0	0
	10						10		10	

The ratio is expressed as: No. of rats killed/No. of rats injected.
* Acute = Deaths within 5 min. after inj.
Delayed = Deaths within 7 days after inj.

tion of deaths at any of the intravenous dosage levels with these two compounds was due to this cardiac effect. The rats died during the injection or within 5 minutes after its completion, usually with convulsions and respiratory distress, even though the drug was given at the rate of 1.0 cc/min. This is in accord with the work of DeGraff and Lehman(14) in which they found that sudden death following the intravenous administration of these drugs cannot be avoided merely by slowing the rate at which they are administered. The production of acute deaths was not experienced with intravenous injections of Mercuhydrin, Thiomerin, or Mercurophylline when combined with Sodium Thioglycollate, and these drugs could be given as rapidly as 1 cc/15 seconds.

Analyses of the rat and its excreta for the distribution of mercury were very inconclusive as to the mechanism of the decidedly increased toxicity of Thiomerin. Actually a quite varied distribution of mercury was found in 9 animals which were analyzed. In general, about 60 to 75% of the administered mercury was excreted in the 2 to 4 days of survival of the animal, although one rat which was given Thiomerin excreted less than 20% of the total in this period. Median retention in the kidneys was only 48 μg of mercury with extremes of 17 and 388 μg. There was no essential difference in distribution between

14. DeGraff, A. C., and Lehman, R. A., *J.A.M.A.*, 1942, v119, 998.

the two diuretics used.

Discussion. A higher incidence of delayed deaths occurred from the administration of Mercurophylline to which Sodium Thioglycollate had been added. This substantiated preliminary testing in which it was found that 0.1 cc of Sodium Thioglycollate and 0.1 cc of Mercurophylline (4.0 mg of mercury) when mixed and injected subcutaneously had a greater toxicity than either drug administered alone. Control injections of 30 mg of Sodium Thioglycollate subcutaneously were non-toxic. The toxicity of the combined Mercurophylline and Sodium Thioglycollate following subcutaneous administration is approximately double that of Mercurophylline and is even greater than that of Mercurophylline with benzyl alcohol when given by this route. The toxicity of this combination of mercurial and thiol is, however, of the same order as that of Thiomerin given subcutaneously in rats, a fact which might be anticipated from their similarity of chemical composition. Thus, the addition of the thiol compound, even though it eliminates the danger of acute death following intravenous injection, so increases the chronic toxicity of Mercurophylline by either subcutaneous or intravenous routes that its addition would seem to be a decided hazard.

Summary. Data presented on the comparative toxicity of Thiomerin, Mercuhydrin and Mercurophylline indicate that the compound containing the thiol group definitely is more toxic in rats. Other reports, both clinical and laboratory, have indicated such a possibility. Sodium Thioglycollate potentiates the chronic toxicity of Mercurophylline when combined with it and given by either the subcutaneous or intravenous route. There is, however, elimination of the acute cardiac effect with intravenous administration. Studies of the distribution of mercury in the rat and its excreta following subcutaneous administration of two of the drugs failed to indicate the mechanism of potentiation of toxicity.

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Pancreatic Secretion to Peptic Ulcer. II. Effect of Hypoglycemia With and Without Pancreatectomy.* (17957)

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A definite relationship exists between pancreatic function and peptic ulcer formation. When the alkaline pancreatic secretions are deviated from the duodenum by external pancreatic fistula, ligation of the pancreatic ducts or by implantation of the ducts into the lower reaches of the gut without ablation of the pancreas, peptic ulcers develop spontaneously (1-4). However, if the duodenum is deprived of the alkaline external secretinos by means of pancreatectomy, ulcers form spontaneously only rarely. It was suggested by Poth, Manhoff, and DeLoach(5) that this difference might be due to the fact that the hyperglycemia due to pancreatectomy suppresses secretion of the gastric acid-pepsin factor sufficiently to protect the gastric and duodenal mucosa from ulceration. The experiments reported in this paper are directed toward the evaluation of this assumption.

Experimental method. Twenty healthy, mongrel dogs were divided into 2 equal groups. Their food consisted of 50 g of raw

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