Groups of litter-mate rats of the same sex were used. Some were (1) castrated, (2) some thyroidectomized, (3) some fed thyroid extract and some (4) gonadectomized and fed thyroid extract. Periods of 29 to 68 days were allowed to elapse after operation so as to allow time for structural changes to result in pituitary and lymphoid tissue. Those fed thyroid extract were given daily .1 g per 100 g body weight for several weeks. Of each pair, one rat was an uninjected control. In the second rat of each pair tissue damage was produced by subcutaneous injections in the legs of .5 cc 4% formalin, twice daily for 2 days, killing the rats on the 3rd day, according to the method used by Selye[11] in intact rats.

The table gives the results. Comparisons should be made between the injected and uninjected rats, in each category, which are litter-mates. As rats of one group, such as the castrated, are not the same age as another group, such as the thyroidectomized, no conclusions have been drawn as to whether rats in one group responded with greater or less degree than those in other groups. However, since the body weights are nearly the same in the different groups, except in the dwarfed thyroidectomized rats, a rough comparison can be made. The results within each group show that the rats injected with formalin had definite shrinkage of lymphoid tissue, no matter what the previous alteration was in lymphoid tissue. In fact, 2 castrated rats which showed chronic infection at the site of operation were not excluded from the series because this additional effect on lymphoid tissue did not prevent its response after acute injury. Since observations made in each group were of the same pattern of response, with few inconsistencies, it was not thought necessary to carry out a larger series of determinations.

Conclusions. Lymphoid tissue (lymph nodes, thymus and spleen) which had been altered by various experimental conditions was capable of shrinkage in response to injury of subcutaneous tissues by formalin injections.

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Studies on the Absorption and Excretion of Streptomycin in Animals.

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The introduction of streptomycin as a chemotherapeutic agent[1,2,3] has made apparent the need for a quantitative study of the distribution of this substance in body fluids. The present study was therefore undertaken to determine the relative concentrations of streptomycin in the blood, urine, feces, and tissues of various animal species following oral and parenteral administration.

Materials and Methods. The streptomycin* used in these studies ranged in potency from 150-400 units per mg of solid. The amount of drug administered was based on the findings in the efficacy and toxicity experiments reported by Robinson, Smith, and Graessle.[3]

The animal species used were normal monkeys, dogs, rabbits, and mice. All animals were maintained on adequate stock diet and housed under conditions of controlled temperature and humidity.

In all experiments streptomycin was admin-

1 Schatz, A., Bugie, E., and Waksman, S. A., PROC. SOC. EXP. BIOL. AND MED., 1944, **55**, 66.

2 Jones, D., Metzger, H. J., Schatz, A., and Waksman, S. A., *Science*, 1944, **100**, 103.

3 Robinson, H. J., Smith, D. G., and Graessle, O. E., PROC. SOC. EXP. BIOL. AND MED., 1944, **57**, 226.

* This material was prepared by Drs. Robert Denkewalter and Max Tishler from broth supplied by Dr. J. W. Foster of the Research Laboratories of Merck & Co., Inc.

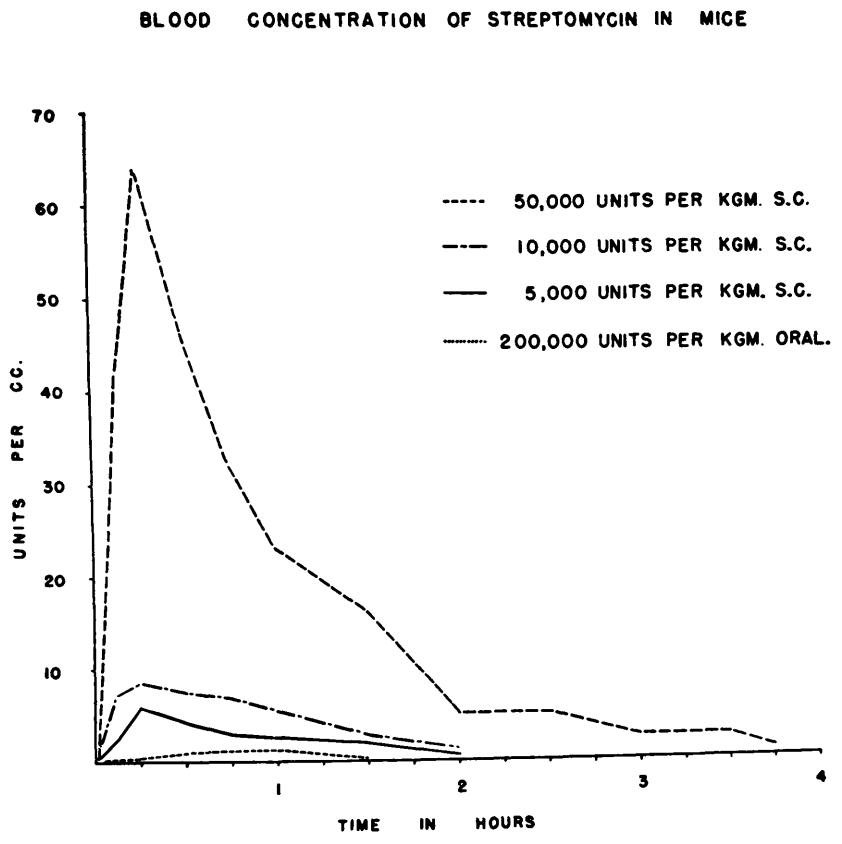

FIG. 1.

istered as an aqueous solution. In order to insure adequate urine flow tap water was administered by stomach tube to each animal 10-15 minutes before giving streptomycin.

Blood and urine samples were taken at frequent intervals. All assays were completed within a 24-hour period. The method of assay, employing *Staphylococcus aureus SM* as the test organism, was that reported by Stebbins and Robinson.[4]

The excretion of streptomycin into the bile was studied in dogs and rabbits. The animals were subjected to deep anesthesia with nembutal, the abdomen opened and the common bile duct cannulated. Samples of bile were obtained at frequent intervals, usually every 15 minutes, and assayed in the same manner as blood and urine.

In those cases where streptomycin was administered orally, or by duodenal instillation, the stools of the gastro-intestinal tract were collected in distilled water, centrifuged, and the supernatant fluid assayed as described above for blood.

Results. Blood Levels. Mice: From the results presented in Fig. 1, it is apparent that streptomycin is rapidly absorbed and excreted following subcutaneous administration in mice. A single therapeutic dose of 5,000 units per kg produced a maximum blood concentration of 6.5 units per ml within 15 minutes, which gradually declined to 2 units per ml during a 2-hour period. Larger doses of 10,000 and 50,000 units per kg subcutaneously resulted in correspondingly higher and more prolonged blood concentrations, although, here again, the drug was rapidly excreted (Fig. 1). Following oral administration of large doses of streptomycin (200,000 units per kg) only small amounts of the drug were de-

[4] Stebbins, R. B., and Robinson, H. J., PROC. SOC. EXP. BIOL. AND MED., 1945, **59**, 255.

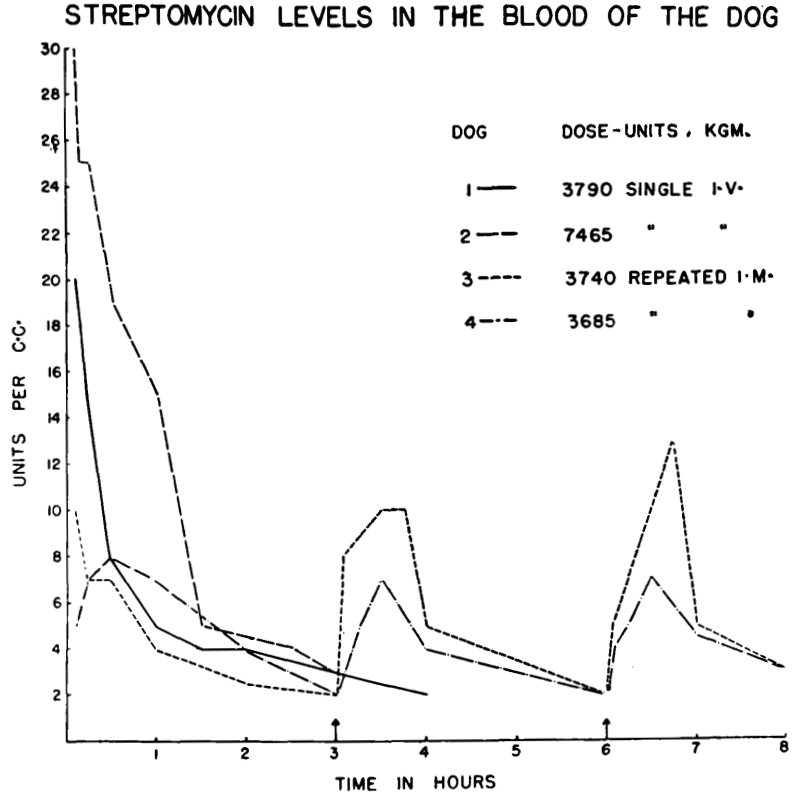

FIG. 2.

tected in the blood (2 units per ml). The blood level curve which was obtained following oral administration of the drug differed from that which was obtained following parenteral injection, in that the maximal concentration following oral administration did not occur until 45 to 60 minutes following drug administration.

Dogs and Monkeys: The findings in dogs and monkeys (Fig. 2 and 3) are similar to those obtained following parenteral administration of streptomycin in mice. Single intravenous dosage of 3700 and 7400 units per kg (100,000 units per dog) of streptomycin produced blood levels of 20 and 30 units per ml respectively, immediately following injection. At the end of 4 hours the blood concentrations decreased to 2-3 units per ml. By repeated intramuscular injection of streptomycin every 3 hours in dogs and monkeys (Fig. 2 and 3) therapeutically effective blood concentrations ranging between 3-18 units per cc could be maintained. Peak blood levels of the drug were reached within 30 minutes after each injection and then gradually declined in much the same manner as that following a single dose.

Urine Levels. The rapid disappearance of streptomycin from the blood following parenteral administration can be largely accounted for by its early appearance in the urine. In a series of ten dogs approximately 50-80% of the drug was excreted in the urine within a 24-hour period. During the first 2 hours after injection 50% of the initial dosage appeared in the urine.

In a group of 2 monkeys receiving 10,000 units per kg of streptomycin per day for 5 days, approximately 60% of the drug was accounted for in the urine during this period (Table I). Examination of the urine collected during the subsequent 5-day period showed that it contained an additional 5-6% although the concentration in the blood was below the level that could be measured.

When the dose of streptomycin was

TABLE I.
Total Urinary Excretion by Monkeys Following Divided Intramuscular Dosage of Streptomycin.

Monkey No.	Time, days	Dose, units/kg × 1000	Total daily dose, units × 1000	Units excreted in 24 hrs × 1000	% daily dose excreted	Total % recovered
1	1	10	28.9	19.9	69	
	2	10	28.9	20.3	70	
	3	10	28.9	14.9	52	
	4	10	28.9	13.6	47	
	5	10	28.9	18.5	63	60
	6	—	—	4.		
	7	—	—	.4		
	8	—	—	1.		
	9	—	—	.6		
	10	—	—	.7		
	11	—	—	.0		65
2	1	10	32.4	19.8	61	
	2	10	32.4	20.5	63	
	3	10	32.4	17.1	53	
	4	10	32.4	16.4	51	
	5	10	32.4	24.5	76	60
	6	—	—	4.3		
	7	—	—	.8		
	8	—	—	1.1		
	9	—	—	1.2		
	10	—	—	.9		
	11	—	—	.0		66
3	1	50	167	70.	42	
	2	50	167	90.	54	
	3	50	167	78.4	47	
	4	50	167	72.	43	
	5	50	167	54.	32	44
	6	—	—	7.2		
	7	—	—	1.6		
	8	—	—	1.3		
	9	—	—	1.1		
	10	—	—	.6		
	11	—	—	.0		45
4	1	50	154	58.4	38	
	2	50	154	37.5	24	
	3	50	154	98.8	64	
	4	50	154	64.	42	
	5	50	154	32.5	21	38
	6	—	—	7.		
	7	—	—	1.4		
	8	—	—	.8		
	9	—	—	.6		
	10	—	—	.3		
	11	—	—	.0		39

increased to 50,000 units per kg in a second group of two monkeys (Table I) and administered by the same dosage schedule, 38-44% of the drug was excreted in the urine during treatment, followed by a further urinary output of 1% of streptomycin in the 5-day period after injection had been discontinued.

Biliary and Fecal Excretion Studies. Assay of the bile obtained from rabbits for an 8-hour period following single intravenous administration of streptomycin (5,000 units per kg) accounted for only 5-10% of the dose.

Dogs receiving 100,000 units per kg by mouth and rabbits which received 8,000 units per kg intraduodenally in a single dose showed no detectable levels in the bile for a similar time period.

Dogs given 100,000-200,000 units per kg perorally on an empty stomach and sacrificed 24 hours later showed 60-80% of the drug

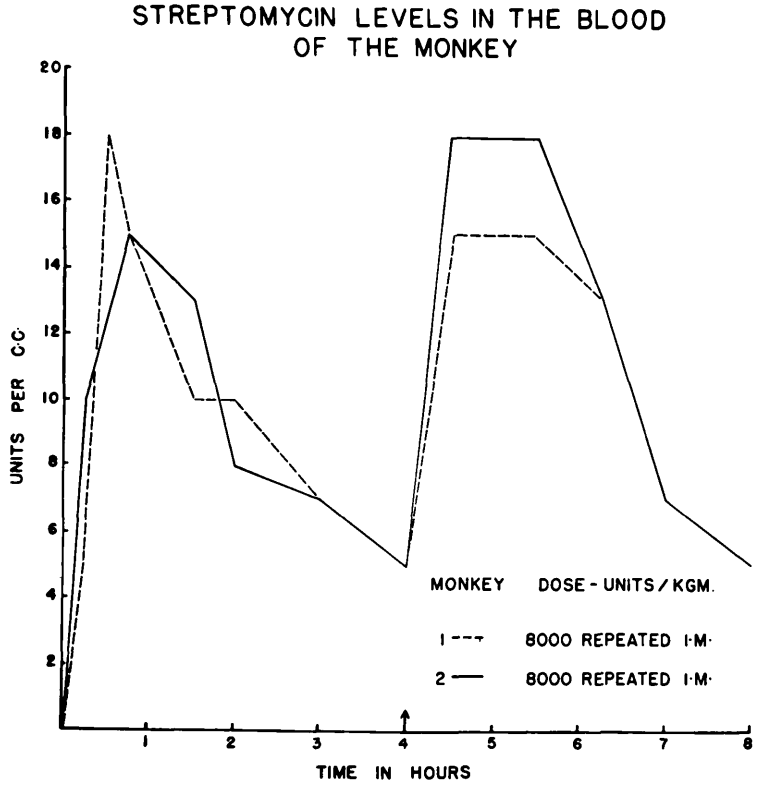

FIG. 3.

unabsorbed from the gastro-intestinal tract. About 5-10% of the drug appeared in the urine during this time. Only traces of streptomycin could be detected in the blood of these animals.

A series of *in vitro* studies on streptomycin in the presence of artificial gastric and duodenal juice indicated that the potency of this antibiotic agent was not materially affected when mixed with these substances for 24 hours at 37.5°C.

Summary and Conclusions. The findings reported in this communication show that streptomycin is rapidly absorbed and excreted following parenteral administration. The rapid disappearance of streptomycin from the blood is largely accounted for by its early appearance in the urine. Approximately 60-80% of the drug is excreted in the urine of dogs within a 24-hour period after parenteral administration. Somewhat smaller amounts were excreted in the urine of monkeys.

When the drug is given perorally relatively small amounts are detected in the blood. This is largely due to the lack of absorption of streptomycin from the gastro-intestinal tract as shown by the large amount of the drug recovered in the feces.

Therapeutic blood concentrations can be maintained by repeated intramuscular injection.

Following intravenous administration of streptomycin, only 5-10% of the dose can be demonstrated in the bile.