large that the animal is overwhelmed and cannot utilize the normal protective mechanism which might be sufficient ordinarily to control the infection. The temperatures of the treated animals were usually 1 to 2 degrees F lower than the controls. The skin lesions of the treated animals are ordinarily less extensive, while the controls frequently presented more edema, swelling and necrotic areas.

Conclusion. 1. A blood level of 7 to 10 mg total sulfapyridine is effective in controlling bacteremia with Type I pneumococcus in 66.6% of treated rabbits. 2. In animals treated with sulfapyridine the local skin lesions are usually less extensive and the temperatures are 1 to 2 degrees lower than the controls.

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Observations on the Toxicology of Sulfathiazole and Sulfapyridine.

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Fosbinder and Walter as well as Lott and Bergeim[1] have prepared the thiazole analogue of sulfapyridine, 2(*p*-amino-benzene-sulfonamido)thiazole, which in this and the following report will be called sulfathiazole. The pharmacology of this drug is of interest because it resembles sulfapyridine in respect of its chemotherapeutic efficacy in combating pneumococcus and other infections in mice.[2]

Experiments which have been performed to determine the acute lethal effects of a single dose of the sodium salt of either drug are listed in Table I.* Each experiment represents observations in a single group of albino mice homogeneous as to breed and size. In experiments B, C, D, and E, the toxicity of the sodium salts of sulfapyridine and sulfathiazole after subcutaneous injection was compared at the same dose levels. In experiment B, the drugs appeared to be equally toxic; however, in experiments C, D, and E, when larger

[1] Fosbinder, R. J., and Walter, L. A., *J. Am. Chem. Soc.*, 1939, **61**, 2032; Lott, W. A., and Bergeim, F. H., in press.

[2] McKee, C. M., Rake, Geoffrey, Greep, R. O., and van Dyke, H. B., PROC. SOC. EXP. BIOL. AND MED., 1939, **42**, 417.

* The molecular weight of sulfathiazole is 2.5% greater than sulfapyridine. Usually doses were corrected for this difference.

doses were administered, sulfapyridine sodium was much the more toxic. In experiment F, both drugs were administered by stomach tube. In this experiment, as in the others, sulfapyridine sodium appears to be the more toxic. "Characteristic" curves drawn from the data of experiments A-E indicate that the LD50 of sulfapyridine Na is about 0.95 g per kilo, whereas that of sulfathiazole Na is about 1.45 g per kilo. The determination of the toxicity of a single dose of the free acids was not attempted because their solubility is low and their absorption is probably extremely variable.[3]

The effects of repeated doses of the free acids were, however, investigated in mice, rats, and monkeys. Mice were fed the Sherman No. 1 diet containing the drugs in concentrations of 1 and 2%. The technics of Bieter and coworkers[4] and Litchfield and coworkers[5] were employed to insure accurate estimation of the amounts of food and drug eaten. The data, summarized in Table II, indicate that

TABLE I.

Toxicity of Sodium Salts of Sulfapyridine and Sulfathiazole Administered as a Single Dose to Mice.

Experiment	Drug	Route of administration	Dose, g/kg	No. of mice	Mortality, %	Time of observation, hr
A	Sulfathiazole Na	Subcut.	0.5	25	4	45
	" "	"	1.0	25	20	45
	" "	"	2.0	25	92	45
	" "	"	3.5	25	100	6
	Alkali control	"	—	25	0	45
B	Sulfapyridine Na	"	0.500	23	4	72
	Sulfathiazole Na	"	0.512	22	4	72
C	Sulfapyridine Na	"	0.750	30	17	72
	Sulfathiazole Na	"	0.768	30	3	72
D	Sulfapyridine Na	"	1.000	30	98	72
	Sulfathiazole Na	"	1.025	30	13	72
E	Sulfapyridine Na	"	1.000	30	100	4
	Sulfathiazole Na	"	1.025	30	17	72
F	Sulfapyridine Na	Stomach tube	1.000	16	19	24
	" "	" "	1.500	16	25	24
	" "	" "	2.000	16	31	24
	Sulfathiazole Na	" "	1.025	15	0	24
	" "	" "	1.537	15	0	24
	" "	" "	2.050	16	6	24

[3] Marshall, E. K., Jr., Bratton, A. C., and Litchfield, J. T., *Science*, 1938, **88**, 597.

[4] Bieter, R. N., Larson, W. P., Cranston, E. M., and Levine, M., *J. Pharm. and Exp. Ther.*, 1939, **66**, 3.

[5] Litchfield, J. T., Jr., White, H. J., and Marshall, E. K., Jr., *J. Pharm. and Exp. Ther.*, 1939, **66**, 23.

when the drugs are repeatedly consumed by mice sulfathiazole is distinctly more toxic than sulfapyridine if the concentration of the drug fed is 2%. No greater toxic effect of sulfathiazole could be detected in mice eating diets containing 1% of either drug. Groups of growing albino rats were fed diets containing 0.5 and 1% of the drugs for a period of 57 days. The weight-curves of the female animals are shown in Fig. 1 and demonstrate that in the rat sulfapyridine is the more toxic drug.

TABLE II.
Effects of Repeated Doses of Sulfapyridine and Sulfathiazole on Mice.

Drug	Amount in food, %	Days on diet	Mean wt at start, g	Mean wt at end, g	No. of survivors on day 0	5	10	15	20	25	28	Mean total dose of drug eaten by survivors, g
Sulfapyridine	1	28	12.06	19.74	14	14	14	14	14	14	14	0.814
"	2	28	15.55	18.56	28	28	28	28	27	27	27	1.477
Sulfathiazole	1	28	14.23	19.49	15	15	15	15	15	15	15	0.908
"	2	28	15.11	17.5	30	26	18	11	9	7	6	1.523

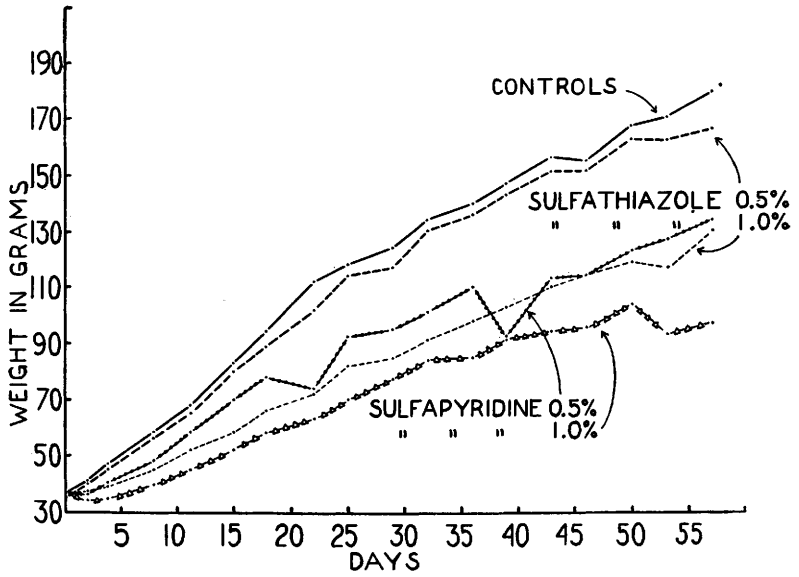

FIG. 1.

Average growth of homogeneous groups of 9 female rats receiving sulfathiazole or sulfapyridine in the food. No deaths occurred.

Comparable results were obtained in a similar experiment with growing males. However, during the experimental period, a sudden severe unexplained loss of weight occurred in the control animals and in those receiving sulfathiazole; subsequent recovery was rapid.

The drugs as the free acids were administered by stomach tube to monkeys in a daily dose of 0.4 g per kilo body weight.† It was estimated that this dose represents about 3 times the maximum human therapeutic dose of sulfapyridine according to present recommendations. Sulfathiazole and sulfapyridine were fed to 3 monkeys each for 14 days, and to 4 each for 21 days. With one exception, the loss of weight was negligible in the monkeys receiving sulfathiazole, whereas it was definite and often pronounced in those given sulfapyridine. One monkey receiving sulfathiazole died on the eighteenth day apparently as a result of uremia due to the accumulation of acetylsulfathiazole in the collecting tubules; in addition, there was distension of the ureter on the left side because of an accretion of the conjugated drug in the ureter. No renal damage could be detected grossly or microscopically in the other 6 monkeys receiving sulfathiazole. Six of 7 monkeys receiving sulfapyridine began to exhibit hematuria and albuminuria after a few days and at necropsy were found to be suffering from unilateral or bilateral hydroureter and hydronephrosis, confirming the reports of others.[6] The chronic toxic effects of sulfathiazole in rats and monkeys are much less pro-

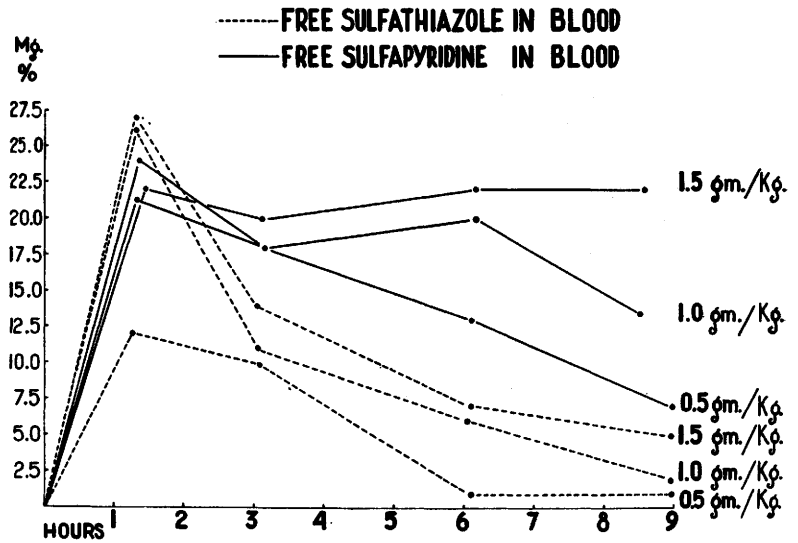

FIG. 2.

The level of sulfathiazole or sulfapyridine in the blood of mice after the oral administration of the free acids. See text.

Ordinate: concentration of drug in blood. Abscissa: time in hours.

† These experiments were performed by Dr. Warren C. Corwin.

[6] Antopol, W., and Robinson, H., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 428, and others.

nounced probably because the drug as shown by our metabolic studies is more rapidly metabolized and undergoes much less conjugation than sulfapyridine.

Metabolic studies have been made in mice, rats, and monkeys. In Fig. 2, the levels of free sulfapyridine and sulfathiazole in the blood of mice are plotted against time in hours.‡ The doses of the free acids as suspensions in gum acacia were administered by stomach tube. Nineteen of the points represent the average values of 5 animals each; the remaining 5 points are the average values of 4 animals each. It is evident that sulfathiazole disappears from the blood more rapidly than sulfapyridine; however, the data do not indicate whether this difference is due to more rapid destruction, more rapid tissue fixation, more rapid excretion, or to a combination of these factors. In a second experiment in mice, the sodium salts in a dose of 1 g per kilo body-weight were injected subcutaneously. At the end of 1, 3, and 6 hours the blood levels were: sulfapyridine, 74, 69, and 47 mg %; sulfathiazole, 90, 79, and 6 mg %. Fifty percent of the mice receiving sulfapyridine were dead at the end of 3 hours, whereas none of the mice receiving sulfathiazole had died spontaneously at the end of 6 hours. Such observations indicate that early deaths cannot be correlated with the levels of the drugs in the blood.

The urinary excretion of the drugs in groups of rats is summarized in experiment 1 of Table III. The total excretion of sulfathiazole was slightly greater in the 24-hour period. However, at the end of 4 and 8 hours the excretion of sulfathiazole had taken place at more than double the rate of excretion of sulfapyridine. During the succeeding 16 hours, sulfapyridine was excreted more and more rapidly. It is of great interest that, in comparison with sulfathiazole, about twice as much sulfapyridine was excreted in the form of the acetylated derivative.|| These findings were confirmed and extended in monkeys (experiments 2-4, Table III) in which an even greater difference in the nature of the excretion-products was detected. The monkey excreted about 3 times as much acetylated sulfapyridine as acetylated sulfathiazole. Total excretion of sulfathiazole was always higher. Particularly in the first 24 hours the rate of excretion of sulfathiazole was approximately twice to 4 times that of sulfa-

‡ Only insignificant amounts of the drugs in the blood appeared to be conjugated. All determinations were made by the method of Bratton and Marshall, *J. Biol. Chem.*, 1939, **128**, 537.

|| It is assumed that the conjugated forms excreted by rats are acetyl derivatives. Acetyl sulfathiazole was isolated from the urine of a monkey receiving sulfathiazole by stomach tube.

TABLE III.
Excretion of Sulfapyridine or Sulfathiazole in Urine of Rats and Monkeys.

Experiment	Drug	Dose, g/kg	Route	Period of observation, hr	% of dose recovered	Distribution of drug in urine	
						Free, %	Conjugated, %
1*	Na Sulfapyridine	0.75	Subcut.	24	25.9	55.8	44.2
	" "		"		24.2	58.2	41.8
	Na Sulfathiazole		"		33.0	77.9	22.1
	" "		"		29.6	81.6	18.4
2	Sulfapyridine	1.00	"	72	10.2	36.5	63.5
	Sulfathiazole		"		23.3	86.2	13.8
3	Na Sulfapyridine	0.500	"	96	55.6	39.6	60.4
	Na Sulfathiazole	0.513	"		67.6	78.6	21.4
4	Sulfapyridine	1.000	Stomach tube	95	19.4	30.2	69.8
	Sulfathiazole	1.025	" "		28.0	76.2	23.8

* There were 4 adult rats in each group in this experiment. All the remaining experiments were performed with single monkeys *(Macaca mulatta)*.

TABLE IV.
Distribution of Sulfapyridine or Sulfathiazole in Blood of Monkeys.*

Experiment†	Drug	Interval after treatment, hr	Drug in blood		
			Total, mg %	Free, %	Conjugated, %
3	Na Sulfapyridine	1.1	5.1	78	22
Subcutaneous	" "	3.2	14.1	65	35
injection	" "	6.1	17.0	63	37
	" "	24.6	21.0	14	86
	" "	48.0	5.7	19	81
	Na Sulfathiazole	1.1	11.7	88	12
	" "	3.1	18.5	83	17
	" "	6.1	15.3	84	16
4	Sulfapyridine	2.9	4.0	50	50
By stomach	"	6.6	10.0	38	62
tube	"	23.3	11.3	8	92
	"	30.8	5.7	11	89
	Sulfathiazole	2.6	3.8	79	21
	"	6.3	4.8	75	25
	"	23.0	1.8	72	28

* No blood-levels below 1 mg % are included. In both experiments observations were concluded after 95-96 hours.
† See Table III for additional data on treatment.

pyridine. In experiment 3, for example, 96% of the total excretion of sulfathiazole occurred within 24 hours after injection whereas 61% of the total sulfapyridine excreted was found in the urine during the succeeding 24-hour periods.

In the monkey the proportion of free sulfathiazole in the blood is also much higher than that of free sulfapyridine (Table IV). This is especially noteworthy in experiment 4 after administration of the free drugs by stomach tube. Although, contrary to our other experiments, the levels of total sulfapyridine are usually the higher, the levels of free drugs are essentially alike, indicating that sulfapyridine would here have no chemotherapeutic advantage since the acetylated form is valueless.

Summary. If the sodium salts of the two drugs be used, the toxicity of sulfathiazole appears to be about 65% of the toxicity of sulfapyridine for LD50. Repeated administration of the drugs in the food of mice indicates that sulfathiazole is more toxic than sulfapyridine at a high dose level but that there is no difference at a dose level which is effective therapeutically. In monkeys and growing rats, receiving either drug for 14-57 days, sulfapyridine is clearly the more toxic. The principal pathological change in all 3 species appears to be renal damage.

Metabolic studies indicate that sulfathiazole is more rapidly metabolized and undergoes much less conjugation than sulfapyridine.