BLOOD STUDIES IN DOGS FOLLOWING THE ADMINISTRATION OF SULFAMERAZINE

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This is to certify that the

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BLOOD STUDIES IN DOGS FOLLOWING THE ADMINISTRATION OF SULFAMERAZINE

by

F. E. EADS

A THESIS

Submitted to the Graduate School of Michigan State College of Agriculture and Applied Science in partial fulfillment of the requirements for the degree of

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THESIS

To

My wife, DOLLY and

daughter, NANCY ANNE

ACKNOWLEDGMENT

It is with pleasure that the writer expresses his most sincere appreciation to Dr. C. S. Bryan for his encouragement and invaluable assistance in carrying out this project. Without his able guidance in solving many technical problems this investigation would not have been possible.

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INTRODUCTION

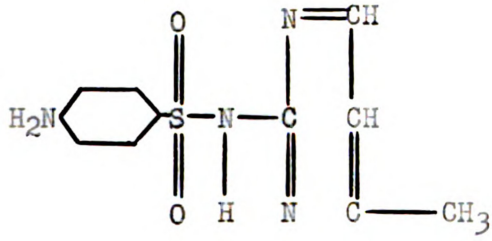
One of the major problems confronting the veterinary practitioner today is the paucity of basic knowledge underlying the use of drugs. With the advent of newer sulfonamides, which are more specific in action, and more specific in methods of administration this defect becomes increasingly apparent. If these drugs are to be used to the best advantage in the treatment of infections caused by susceptible organisms, basic studies are essential.

The following study was limited to certain observations on sulfamerazine in dogs. The four main objectives upon which this investigation was based were: (1) To determine the route which was the most convenient for administration. (2) To determine the route or routes which were equally effective or advantageous in maintaining therapeutic sulfamerazine blood plasma levels. (3) To determine the frequency of administration and the dosage necessary to maintain therapeutic blood plasma levels with sulfamerazine. And, (4) To determine the immediate effect of sulfamerazine on the blood picture of dogs following its administration.

REVIEW OF LITERATURE

The results obtained from blood studies following the administration of sulfamerazine in various animal species have been reported. Limited experimental data comparing the results obtained by the administration of sulfamerazine in dogs by various routes and dosages have been published.

Sulfamerazine (2-sulfanilamido-4-methylpyrimidine) is represented by the following structural formula.



Sulfamerazine

Sulfamerazine has distinct advantages over other sulfonamides in veterinary practice in that it is more rapidly and completely absorbed from the gastrointestinal tract and more slowly eliminated in the kidneys (Welch et al., 1943), (Goodwin et al., 1942), (Murphy et al., 1943), (Bryan, 1946) (Francis, 1947), (Wastrack and Lewis, 1947), and (Scheidy and Tillson, 1947). Therefore, therapeutic blood levels are maintained for longer periods and these levels can be produced and maintained by smaller and less frequent doses of the drug. In man, sulfamerazine is mainly absorbed from the small intestine and only slightly from the stomach with the large intestine intermediate in absorptive capacity (Murphy et al., 1943), and (Welch et al., 1943).

Sodium bicarbonate administered by mouth in sufficient amounts to increase the urinary pH from 6.8 to 7.8 increased the clearance of sulfamerazine two or three fold (Peters, Beyer, and Patch, 1944). This was likely due for the most part to increased electrolyte excretion rather than to pH change. Beyer et al., (1944) reported that the administration of sodium bicarbonate increased the urinary pH but it also increased the clearance value of sulfamerazine by interfering with its reabsorption. As the plasma level of the compound was increased, and more of the drug was filtered by the renal glomeruli, there was an increase in the amount of the sulfonamide that was reabsorbed through the renal tubules; therefore the ratio between plasma concentration and the amount excreted per unit of time remained fairly constant. Earle (1944) found an observable decrease in the reabsorption of sulfamerazine following the administration of sodium bicarbonate. Peters et al., (1944) reported that the clearance of sulfamerazine remained fairly constant when the plasma level was increased from 3.4 to 13.1 mg. percent indicating that maximum tubular reabsorption of the compound was not exceeded. Assuming complete glomerular filtration of unbound sulfamerazine, normally about 80 percent of the filtered compound is reabsorbed by the renal tubules of the dog. Similar results were reported in man by Reinhold et al., (1945). Earle (1944) calculated that in man 63 percent of the sulfamerazine is bound at a plasma concentration of 10 mg. percent. There is a more extensive binding of sulfamerazine in human than in dog plasma, and the renal clearance of sulfamerazine in man is considerably lower than in the dog. Davis (1943) has reported that essentially all the binding of sulfonamides is on the albumin fraction of plasma. Beyer et al., (1946)

found that the binding of these same agents varies from one species to another.

Welch and his co-workers (1943) found that Sulfamerazine and its acetyl derivatives were approximately 20 percent more soluble than the respective forms of sulfadiazine in both water and urine.

Sulfamerazine blood plasma concentrations have been studied in dogs, cats, horses, cattle, sheep, swine, foxes, mink, poultry and man. Welsh and his associates (1946) have demonstrated that, following the administration of a single oral dose of sulfamerazine (½ grain per pound, or 71.5 mg. per kilogram of body weight), it is possible to obtain and maintain higher blood concentrations in the dog than with sulfanilamide, sulfapyridine, sulfathiazole, sulfadiazine, sulfamethazine, or sulfaguanidine. Sulfamerazine also gave effective blood levels in cats but did not produce adequate blood concentrations in horses, cattle, sheep or swine. They concluded than an adequate dosage for animals is a maintenance blood level above 5 mg. percent, and preferably above 8 mg. percent twenty-four hours a day. Inadequate levels and fluctuating blood levels are responsible for the development of drug-fast strains of bacteria. The blood plasma level studies of Welsh et al., (1946) using ½ grain (71.5 mg. per kilogram) per pound of body weight are comparable to the data reported herein.

A similar study was made by Scheidy and Tillson (1947) comparing the blood concentrations of sulfadiazine, sulfamerazine, and sulfamethazine in cattle. Therapeutic blood levels of sulfamerazine were maintained for twenty-four hours by a single oral dose of

1 grain per pound of body weight. Intravenously, using ½ grain per pound of body weight in cattle, similar results were obtained to those shown in Table XIII.

In a more recent study on blood concentrations following the administration of sulfamerazine and sulfamethazine to cattle, Scheidy et al., (1949) found that only a very small percentage of the absorbed drug is acetylated by cattle. A combination of these two drugs caused no apparent serious toxic reactions so far as could be determined, and the values for creatinine, non-protein nitrogen, icterus index, hemoglobin and average blood values appeared to be within the normal range during each phase.

Welsh et al., (1948) in studies of sulfonamide blood concentrations in foxes found very low concentrations of sulfamerazine when ½ grain per pound was administered in a single dose per os. Similar studies were made by Langer et al., (1948) in mink. Much higher blood concentrations were found, in fact, ½ grain per pound in a single dose per os gave a therapeutic blood level for eight hours and compared favorably with sulfamethazine and sulfadiazine.

Sulfamerazine blood levels in poultry have been studied by Thorp et al., (1947), Gordeuk and Learned (1948) Kiser et al., (1948), Mattis et al., (1946), Severens et al., (1945), Bottorff and Kiser (1947), Anderson et al., (1947), Alberts and Graham (1948), and others. Thorp et al., (1947) observed that sodium sulfamerazine administered in a concentration of 0.2 percent in water for five days resulted in an average blood concentration of 13.68 mg. of free sulfamerazine per 100 ml. When 1.0 percent sulfamerazine was fed in the mash, an average blood level of 27.03 mg. of free drug per 100 ml.

of blood resulted. Comparable results were obtained with 0.25 percent concentration of sulfamerazine in the mash. An average of 8.26 mg. of free drug per 100 ml. of blood was obtained during a five-day treatment. Gordeuk and Learned (1948) and Kiser et al., (1948) reported similar results in chickens and turkey poults. Alberts and Graham (1948) obtained blood sulfonamide concentrations of 16.4 mg. percent within three hours after the fourth oral dose of sodium sulfamerazine (12 hour intervals, 0.5 grains per pound) was administered to turkeys. Using 0.2 percent sodium sulfamerazine in chickens, Anderson et al., (1947) obtained blood concentrations of 16 mg. percent. Mattis et al., (1946) obtained the same blood concentration when the dosage of sulfamerazine in the feed was increased to 0.5 percent. Blood concentrations of 30 mg. percent or higher were obtained by Severens et al., (1945) with 0.5 to 2.0 percent sulfamerazine. Bottorff and Kiser (1947) reported blood concentrations ranging between 12 and 31 mg. percent when 0.5 to 0.75 percent were administered.

Extensive work has been done in humans with the various sulfonamides. Gilligan (1943) in his studies on the plasma binding properties of sulfadiazine, sulfamerazine, and sulfamethazine in man found that plasma levels of sulfamerazine were considerably higher than for sulfadiazine suggesting that this drug is bound in the plasma to a greater extent. As the plasma concentration decreased, the percent of binding of the drug increased. In vitro dialysis studies show sulfamerazine to be bound in the plasma to a greater extent than sulfadiazine. The binding of the drug in the plasma in vivo was essentially the same as was found in the in vitro studies. Sieber and Clark (1945) stated that plasma binding alone cannot account for pro-

longed blood levels of sulfonamides. Other factors which determine the magnitude and maintenance of blood levels are: 1) solubility in and absorption from the intestinal tract; 2) presence or absence of food; 3) the emptying time of the gastro-intestinal tract; 4) excretion into the intestinal tract; and 5) urinary and fecal excretion.

Welch and his associates (1944) found that when an initial 2 gram dose of sulfamerazine, followed by daily maintenance doses of 1 gram was administered, the average daily concentration of free sulfamerazine in the blood of 8 normal men decreased from about 6.5 mg. to 3.5 mg. percent. Hageman et al., (1943) gave an initial dose of 4 grams by mouth and a maintenance dose of 1 gram every 8 hours and obtained blood plasma levels of 20 mg. percent at two hours which decreased to 14 mg. percent in five hours.

Murphy and his co-workers (1943) reported that when sulfamerazine was administered rectally in aqueous suspension by retention enema, at no time was more than a trace of the drug found in the blood.

Renal elimination and toxicity are the two primary concerns in sulfonamide therapy. Important toxic manifestations are granulocytopenia, hemolytic anemia, and kidney concrements (Green, 1944). The low toxicity of sulfamerazine has made it the drug of choice in the treatment of systemic bacterial infections. The lower concentration in the urine and the fact that both the drug itself and its acetyl derivative were more soluble in neutral and acid urine than the corresponding forms of the older sulfonamides, lessen the possibility of renal obstruction (Welch et al., 1943). Welch and

his associates also reported that the toxicity apparently was about the same for sulfamerazine and sulfadiazine when a comparison was made on the basis of the concentration of the compounds in the blood.

Schmidt et al., (1944) compared the toxicity of sulfamerazine, sulfamethazine and sulfadiazine. Insofar as acute and/or chronic toxicity are concerned there appeared to be little difference between the three compounds. However, both sulfamerazine and sulfamethazine seem to be more desirable than sulfadiazine as far as solubility and renal toxicity are concerned.

Studies by Lehr (1945) and Frisk et al., (1947) with various sulfonamide compounds have shown that there is a reduced chance of renal toxicity in small experimental animals and in humans when mixtures of sulfonamides, rather than one or the other of the components, are given.

Mattis et al., (1946) conducted toxicity studies with young chickens and administered 0.5 and 1.0 percent sulfamerazine in dry mash for 14 successive days. This dosage of the compound produced no evidence of toxicity in the chickens. Average concentrations of free sulfamerazine in the blood of the treated chickens were respectively 14.7 and 29.1 mg. percent when the drug was fed in these concentrations. Farr and Jaquette (1947) reported toxicity in chickens with sulfamerazine in the mash at levels higher than 0.25 percent or in 0.5 gram daily doses administered in capsules for a week. They observed a retarding of weight gain which they felt was due both to the unpalatability and to the toxic effects of the drug.

Hall and Spink (1943) reported a low incidence of toxicity in 115 human cases. Leukopenia with or without granulocytopenia

occurred in about one percent of the cases. Similar results were reported by Green (1944), and Clark and his associates (1943). Kracke (1947) stated that the sulfonamide drugs, in addition to causing leukopenia, may produce a marked degree of neutrophilic leukocytosis.

Schmidt et al., (1944) conducted toxicity tests on mice and found that animals receiving lethal doses of sulfamerazine died within 18 to 36 hours after treatment. When 5 grams per kilogram of body weight was administered by mouth a blood concentration of 148 mg. percent was obtained. This killed 93 percent of the mice. When a 2 gram per kilogram dose was administered subcutaneously, a blood concentration of 164 mg. percent was obtained which produced a 90 percent mortality. When the same dosage was administered intraperitoneally, a blood concentration of 161 mg. percent was obtained which was 95 percent fatal. They found the toxicity of sulfamerazine for the urinary tract to be evident when blood levels exceeded 40 mg. percent. In animals with such blood levels, ureters, bladders and kidney pelvis frequently contained sulfamerazine concretions composed of both free and conjugated drug. The incidence of these deposits rose progressively with the blood levels of the drug. Sulfamerazine seemed to have somewhat less urinary tract toxicity in the dog than in the rat (Schmidt et al., 1944).

Eads (1949) reported that sulfamerazine may be administered safely to dogs and cats in daily dosages of 1 grain per pound of body weight in divided doses. In this clinical study no visible untoward effects were noticed after 7 to 11 successive days of treatment.

Leukopenia following the administration of sulfamerazine occurs in man but not in experimental animals (Litchfield, 1948). Data reported herein show that a marked leukopenia developed upon the administration of large doses of sulfamerazine to normal healthy dogs. Clark et al., (1943) observed leukopenia in man with sulfamerazine therapy. They reported no cases of acute hemolytic anemia or agranulocytosis. Jennings et al., (1937) reported a few cases of agranulocytosis, all with fatal termination. No agranulocytosis or granulopenia occurred even when a leukopenia developed during sulfonamide therapy (Bigler et al., 1938). The action appeared to be independent of the leukocytes in that it did not produce an increase in total leukocytes or in the proportion of neutrophiles.

According to Denny and Menten (1946), blood dyscrasias are among the less frequently observed pathologic changes produced by the sulfonamides which include hemolytic anemia, aplastic anemia, and agranulocytopenia. Aplastic anemia was rare in both children and adults but hemolytic anemia was seen rather frequently. Favorite, Reiner and London (1944) reported a case of leukopenia in man due to sulfamerazine when 42 grams was administered during a 23 day period. No changes in the red cell count or hemoglobin content were noticed but a marked lymphocytosis (93 percent) and neutropenia (7 percent) was observed. Menten and Graff (1946) conducted a survey of the hemoglobin and red cell counts of children whose blood exhibited granulocytopenia and found that they did not usually reveal an accompanying anemia. The percentage of neutropenias developing from sulfonamides in their studies was about 20 percent of all patients receiving the drug.

Malkamus and Opperman (1944) reported the normal ranges

of blood constituents in the dog as follows: erythrocytes 5.5 to 8 million per cu. mm., leukocytes 9 to 10 thousand per cu. mm., and the leukocyte differential count was 60 to 80 percent neutrophiles, 2 to 4 percent eosinophiles, 0 to 0.5 percent basophiles, 13 to 32 percent lymphocytes, and 3 to 5 percent monocytes.

The normal ranges of the canine blood constituents reported by Coffin (1945) are as follows: erythocytes 5.5 to 8.8 millions per cu. mm., leukocytes 5 to 20 thousand per cu. mm., hemoglobin 12.5 to 17.3 grams per 100 ml., neutrophiles 3.6 to 15 thousand per cu. mm., eosinophiles 0.1 to 2 thousand per cu. mm., basophiles 0 to 0.4 thousand per cu. mm., lymphocytes 0.6 to 6 thousand per cu. mm., monocytes 0.1 to 2.4 thousand cu. mm.

Boddie (1946) reported the following as normal ranges in dogs: erythrocytes 5.1 to 7.6 million per cu. mm., leukocytes 8 to 14.6 thousand per cu. mm., hemoglobin 11.2 to 14.8 grams per 100 ml., and the leukocyte differential count was 0 to 4 percent eosinophiles, 67 to 81 percent neutrophiles, 14.6 to 25.4 percent lymphocytes, 1 to 7 percent monocytes.

METHODS

Estimation of Sulfamerazine in the Circulatory System.

The method employed by Hoffman (1941) was used. The concentration of free sulfamerazine in the blood was determined by the method of Bratton and Marshall (1939). The technique was as follows: Collect two or three drops of blood from the radial vein in a small tube containing a crystal or two of potassium oxalate. Transfer 0.1 ml. of this to a small test tube containing exactly 5 ml. of distilled water. The pipette should be washed out by sucking up the water and blowing it out several times. Add 1.0 ml. of 15 percent trichloroacetic acid. Mix thoroughly with the pipette, and again wash the pipette by sucking up the fluid and blowing it back. Centifuge the sample until the precipitate is completely separated from the filtrate. Transfer 4 ml. of the filtrate into a 10 ml. graduated tube; add 0.5 ml. of 0.1 percent sodium nitrite and mix. After 3 minutes, add 0.5 ml. of ammonium sulfamate. After 1 minute, add 0.5 ml. of N (1-naphthyl) ethylenediamine dihydrochloride. Let the color develop for at least 1 minute, dilute to 8 ml., mix, and compare readings in the Cenco-Sheard-Sanford Photelometer using the green filter with a standard stock solution of sulfamerazine. Preparation of this stock solution is as follows; Weigh out accurately 100 mg. of pure sulfamerazine* and transfer to a liter volumetric flask with a little distilled water. Add a few drops of 10 percent sodium hydroxide to hasten

* Sharp and Dohme Sulfamerazine Chemical Reagent (Powder).

the solution of the comparatively insoluble powder, dilute with distilled water to the 1000 ml. mark and shake until all the sulfamerazine is dissolved.

Calculation is made by reference to a calibration curve obtained by analysis of the stock solution. This calibration curve is determined as follows: Transfer 2.0, 1.5, 1.0, 0.5, and 0 ml. respectively of this stock solution to 50 ml. volumetric flasks. To each of these add 9 ml. of 15 percent trichloroacetic acid. Make up each solution to 50 ml. with distilled water, and mix thoroughly. These solutions represent 1 to 40 filtrates of blood specimens containing respectively 16, 12, 8, 4, and 0 mg. per 100 ml. Analyze 10 ml. of each of these solutions exactly as above for blood filtrates, making up finally to 20 ml., and read in the Photelometer, using the green filter. Plot the values on semi-logarithmic paper. They fall in a straight line in the lower concentrations but may have a slight tendency to form a convex curve in the region of 16 mg. per 100 ml.

Determination of Hemoglobin. The Photelometer was used in determining hemoglobin (Sanford et al., 1933). Preparation of the sample of blood was as follows: Twenty ml. of the diluting fluid (0.1 percent solution of sodium carbonate in water) was measured accurately into a suitable container such as 50 ml. Erlenmeyer flask. To this was added 0.1 ml. of blood. The mixture was thoroughly shaken and then transferred to the absorption cells and checked in the Photelometer, using the green filter. This reading was then obtained in grams of hemoglobin per 100 ml. of blood from a calculated table.

Leukocyte and Erythrocyte Enumeration. Leukocyte and erythrocyte counts were made within 24 hours after bleeding using Thoma diluting pipettes and Bright-Line Improved Neubauer Counting Chambers (Coffin, 1945). Two percent oxalic acid (Jones, 1927) was used as the diluting fluid for counting leukocytes, while Leake and Guy diluting fluid (Todd, 1943) was used for counting erythrocytes.

Differential Leukocyte Counts. Blood smears were prepared within one hour after bleeding by the two-slide method using oxalated blood and stained with Wright's stain (Todd, 1943) immediately following drying.

The dogs used in this study were obtained from a city dog pound. All dogs were hospitalized for several months prior to use. None of the dogs developed distemper or any other contagious disease throughout the course of study. The ages ranged from approximately fifteen months to five years. The animals were fed once each day; the ration consisted of one part of cooked meat scraps and two parts of commercial cereal dog food. This was fed at approximately three p.m. each day. Water was kept before the dogs at all times. Sulfamerazine administration and the drawing of blood samples were always begun in the morning, and blood samples were collected at regular intervals until that phase of the experiment was concluded. All of the animals gained weight steadily during the entire course of study.

The anti-coagulant used in collecting blood samples was 2 percent potassium oxalate*; one-half ml. of this preparation

* 1 Gm. of potassium oxalate was placed in 50 ml. of distilled water.

was placed in each bleeding vial and placed in a paraffin oven until dried. Each vial therefore contained 10 mg. of potassium oxalate which is enough oxalate to prevent the clotting of 5 ml. of blood.

The types of sulfamerazine products and routes of administration used in this experiment were: 1) in tablet form, 2) in powder form, 3) in an emulsion, 4) intravenously, and 5) subcutaneously. Each type of product was administered at the rate of $\frac{1}{2}$ grain (71.5 mg. per kilogram), 1 grain (143 mg. per kilogram), and $1\frac{1}{2}$ grains (214.5 mg. per kilogram) of sulfamerazine per pound of body weight respectively. Further blood plasma concentrations were also determined when sulfamerazine was administered once, twice and three times daily for a three day period as compared to a 24 hour period.

The first objective in this study was to compare sulfamerazine blood plasma levels following the single oral administration of $\frac{1}{2}$ grain (71.5 mg. per kilogram), 1 grain (143 mg. per kilogram), and $1\frac{1}{2}$ grains (214.5 mg. per kilogram) per pound of body weight. Dogs numbered 101, 102, 103, 104, and 105 were used. Tables IX through XIII. In this series of trials dogs were weighed immediately prior to administering the drug in tablet form. One hour following the administration of sulfamerazine, the first blood sample (approximately 1 ml.) was drawn from the right radial vein. Blood samples were then taken at 4 hour intervals for the next twelve hours followed by another at twenty-four hours. Beginning with the first sample drawn, blood was taken at specified intervals and checked for red and white cell counts, hemoglobin content, and leukocyte differential counts, to determine if sulfamerazine had any effect on alter-

ing these in the normal healthy animal. Similar series of trials were conducted on dogs numbered 115, 116, and 117 (Tables XIV through XVI) using the powdered sulfamerazine given by mouth in capsules, and on dogs numbered 118, 119, and 120 (Tables XVII through XIX) using an emulsion measured in a 10 ml. syringe and deposited in the mouth to assure proper dosage. In these trials, blood samples, red and white cell counts, hemoglobin determinations and leukocyte differential counts were studied and compared with the results when the tablet form of the drug was used.

In the next series of trials a sterile aqueous solution containing 6 percent sulfamerazine was used on dogs numbered 121, 122, and 123 (Tables XX through XXII). Subcutaneous injections were made in the nuchal region approximately on the midline. This location was chosen as it is readily accessible and caused the least discomfort to the animal. A 20 gauge needle was used. Following the subcutaneous injections blood samples were drawn one hour following the injection, and then at 3 hours, 5 hours, 8 hours, 12 hours, 24, 36, and 48 hours. In this series of studies samples were taken at the beginning and at twenty-four hours of each individual trial to determine the effects of sulfamerazine on the blood picture when given by this route.

Intravenous injections were made in the right radial veins of dogs numbered 124, 125, and 126 (Tables XXIII through XXV) using a 20 gauge needle. The first blood sample was drawn immediately following injection before the needle was removed from the vein, using a clean syringe to draw the blood sample. Another blood sample was drawn one hour after injection and thereafter at two-hour inter-

vals for eight hours, then at 12, 24, 36, and 48 hours. As with the other routes of administration, the blood picture was determined at specific intervals.

Subsequent trials were conducted on dogs numbered 106, 109, 112, 107, 110, 113, 108, 111, and 114 (Tables XXVI through XXXIV) when sulfamerazine was administered in tablet form by mouth once, twice, and three times daily for a three day period.

The results of Tables IX through XXXIV are summarized in Tables I through VIII.

Table I. Average Blood Plasma Levels Following the Oral Administration of a single dose of Sulfamerazine in Tablet Form. Dogs No. 101, 102, 103, 104, 105, 106, 109, 112 in appendix.

Hours after Administration	Gr./lb. body weight administered		
	½	1	1½
	mg. %	mg. %	mg. %
1	1.3	2.2	4.0
4	3.6	6.3	6.9
8	3.3	6.0	6.6
12	2.1	4.8	6.3
24	1.7	3.2	5.0

Table II. Average Blood Plasma Levels Following the Oral Administration of a single dose of Sulfamerazine in Powder Form. Dogs No. 115, 116, 117 in appendix.

Hours after Administration	Gr./lb. body weight administered		
	½	1	1½
	mg. %	mg. %	mg. %
1	1.2	2.5	3.5
4	4.8	6.6	7.2
8	4.2	6.2	7.1
12	2.5	5.5	6.5
24	2.0	4.5	5.0

Table III. Average Blood Plasma Levels Following the Oral Administration of a single dose of Sulfamerazine in an emulsion. Dogs No. 118, 119, 120 in appendix.

Hours after Administration	Gr./lb. body weight administered		
	½	1	1½
	mg. %	mg. %	mg. %
1	0.6	2.5	4.3
4	4.3	6.2	7.8
8	4.0	5.9	6.8
12	2.0	4.9	6.0
24	1.5	4.8	5.3

Table IV. Average Blood Plasma Levels Following the Administration of a single dose of Sulfamerazine Subcutaneously. Dogs No. 121, 122, 123 in appendix.

Hours after Administration	Gr./lb. body weight administered		
	½	1	1½
	mg. %	mg. %	mg. %
1	10.5	16.0	19.5
3	12.5	19.0	22.7
5	5.4	19.2	21.8
8	2.1	17.6	19.2
12	1.0	13.1	16.6
24	0.2	8.0	10.5
36		2.4	8.7
48		1.0	5.8

Table V. Average Blood Plasma Levels Following the Administration of a single dose of Sulfamerazine Intravenously. Dogs No. 124, 125, 126 in appendix.

Hours After Administration	Gr./lb. body weight administered		
	½	1	1½
	mg. %	mg. %	mg. %
0	13.0	21.3	37.1
1	9.0	12.3	20.1
2	8.2		
3		11.1	17.8
4	5.0		
5		9.4	17.4
6	4.7		
8	3.5	9.0	16.9
12	3.2	6.8	14.5
24	1.4	4.4	10.5
36		3.7	7.7
48		1.4	5.0

RESULTS

The objective of this study was to compare sulfamerazine blood plasma level curves following the oral administration of sulfamerazine with the plasma levels obtained following both intravenous and subcutaneous injection.

ORAL ADMINISTRATION

With a single oral administration of sulfamerazine in tablet form the highest blood plasma concentration was obtained in four hours. When ½ grain (71.5 mg. per kilogram) per pound of body weight was administered a concentration of 3.4 mg. percent was found. This concentration was maintained for two hours followed by a gradual fall so that 2.0 mg. percent was still present at twenty-four hours. With 1 grain (143.5 mg. per kilogram) per pound of body weight, a blood plasma concentration of 6.5 mg. percent was obtained at 4 hours, with 5.1 mg. still present at 8 hours; at twenty-four hours the concentration of 3.2 mg. percent. Because of this remaining blood plasma concentration, a forty-eight hour sample was drawn which contained 0.8 mg. percent. Using 1½ grains (214.5 mg. per kilogram) per pound of body weight a concentration of 6.4 mg. percent was obtained at four hours, with 5.5 mg. per 100 ml. at twelve hours and 5.0 mg. still present at twenty-four hours. Comparable results were obtained using sulfamerazine in powder form as well as an emulsion.

SUBCUTANEOUS ADMINISTRATION

The blood plasma levels obtained after subcutaneous administration of the sulfamerazine compared very closely with the

intravenous injection both in regard to the highest concentration obtained with each dosage and to the rapid drop in concentration from this point to the forty-eight hour readings. The highest concentration of sulfamerazine, when given by this route, required approximately three hours after which time it was rapidly eliminated from the circulatory system. The 24 hour samples contained 0.2, 8.0, and 10.5 mg. percent of free sulfamerazine respectively when ½, 1, and 1½ grains per pound of body weight of the drug were injected. The 36 hour samples had 2.4 and 8.7 mg. percent while the 48 hour samples yielded blood plasma readings of 1.0 and 5.8 mg. percent, respectively, of sulfamerazine when 1 and 1½ grains per pound was the dose.

INTRAVENOUS ADMINISTRATION

Since the sulfamerazine was introduced directly into the circulatory system when given intravenously it was found almost immediately at the concentration of 13.0 mg. percent with ½ grain per pound of body weight, 21.3 mg. percent with 1 grain per pound of body weight and 37.1 mg. percent with 1½ grains per pound of body weight when injected in a single dose. The concentrations fell quite rapidly so that at twenty-four hours blood plasma levels of 1.4, 4.4, and 10.5 mg. percent respectively were obtained. Since rather high blood levels were still present at twenty-four hours when 1 grain and 1½ grains per pound of body weight were used, samples were drawn at thirty-six and forty-eight hours. At thirty-six hours the blood plasma concentration using 1 grain per pound was 3.7 mg. percent while 7.7 mg. were obtained using 1½ grains per pound of body weight.

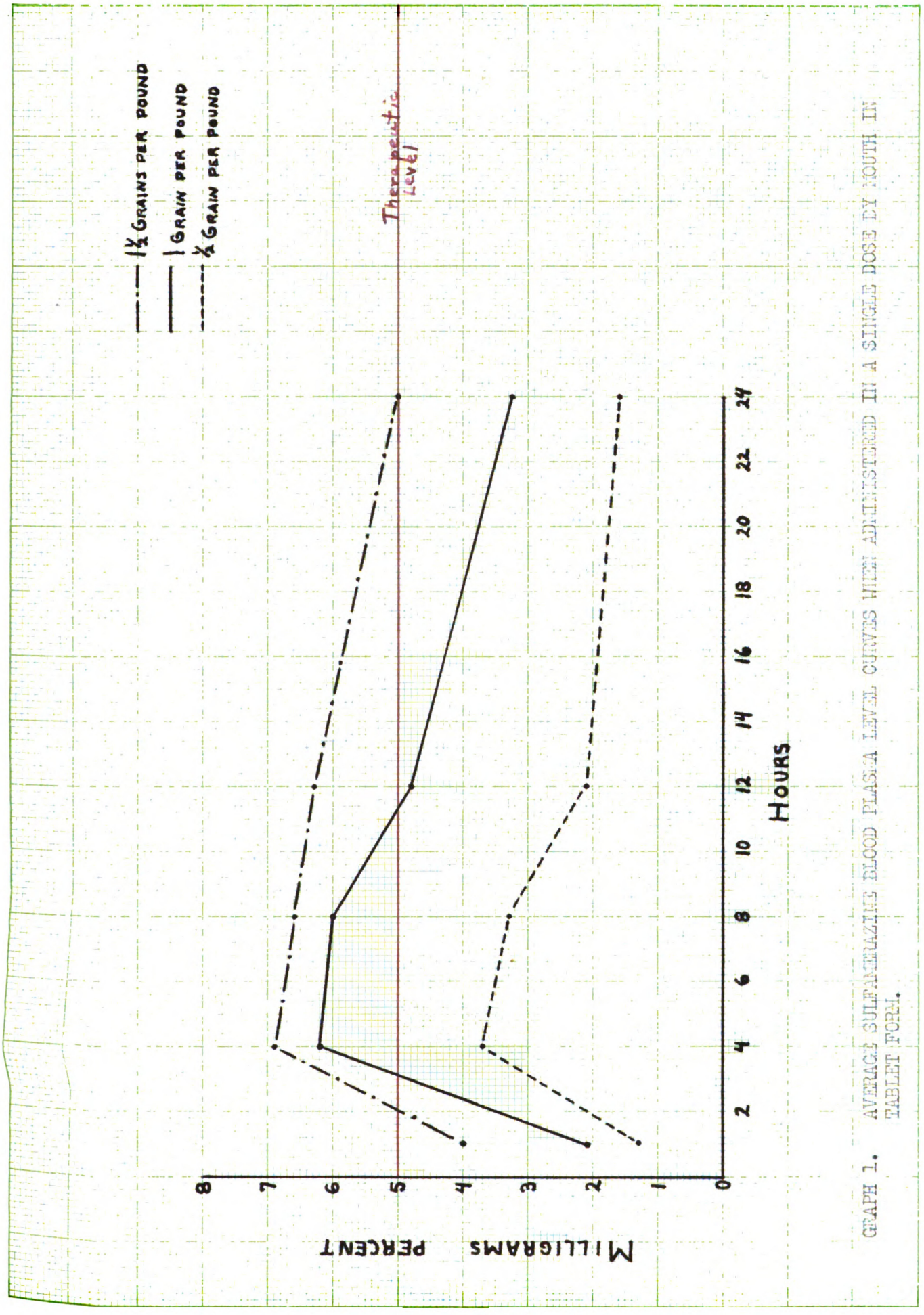
At forty-eight hours the concentration had dropped to 1.4 mg. percent with 1 grain per pound and 5.0 mg. percent with 1½ grains per pound of body weight.

DISCUSSION

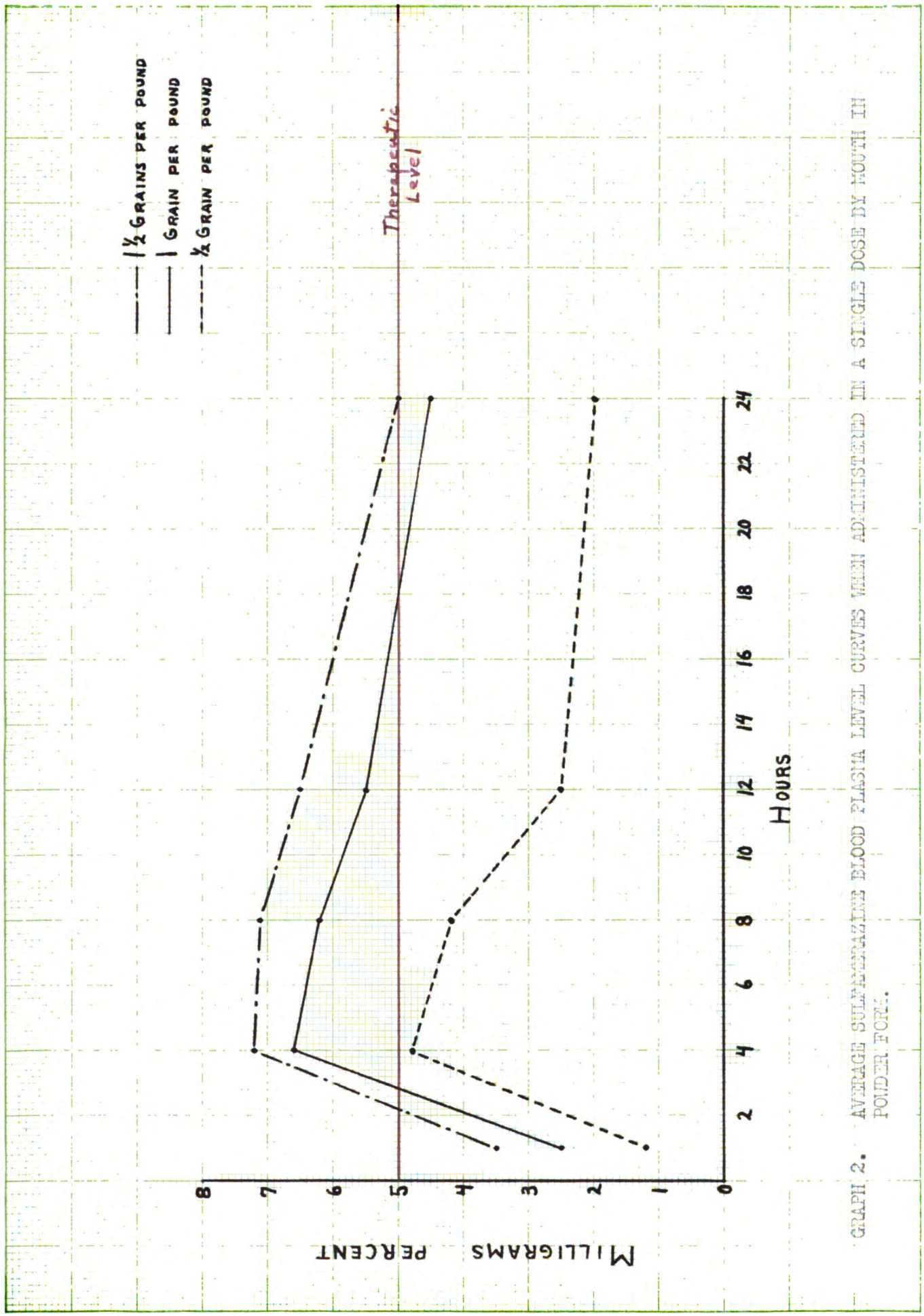
Graph 1 presents the average sulfamerazine blood plasma level curves following a single oral administration of ½, 1, and 1½ grains per pound of body weight in tablet form. When these curves obtained on canines were compared with curves on human sulfamerazine blood plasma levels (Welch et al., 1944) administered in single daily doses, they were found to be quite similar. Comparable results were also obtained by Welsh et al., (1946) when they used ½ grain of sulfamerazine per pound of body weight in a single dose per os. In mink slightly higher concentrations were found using a single oral administration of ½ grain of sulfamerazine per pound of body weight (Langer et al., 1948), while in foxes the concentrations were slightly lower (Welsh et al., 1948). The average plasma concentration of free sulfamerazine in cattle following a single oral dose of approximately 1 grain per pound of body weight was found to be lower than those found in the dog (Scheidy and Tillson, 1947).

The data of Graphs 2 and 3 present results obtained when similar doses of powdered sulfamerazine and an emulsion of sulfamerazine were used.

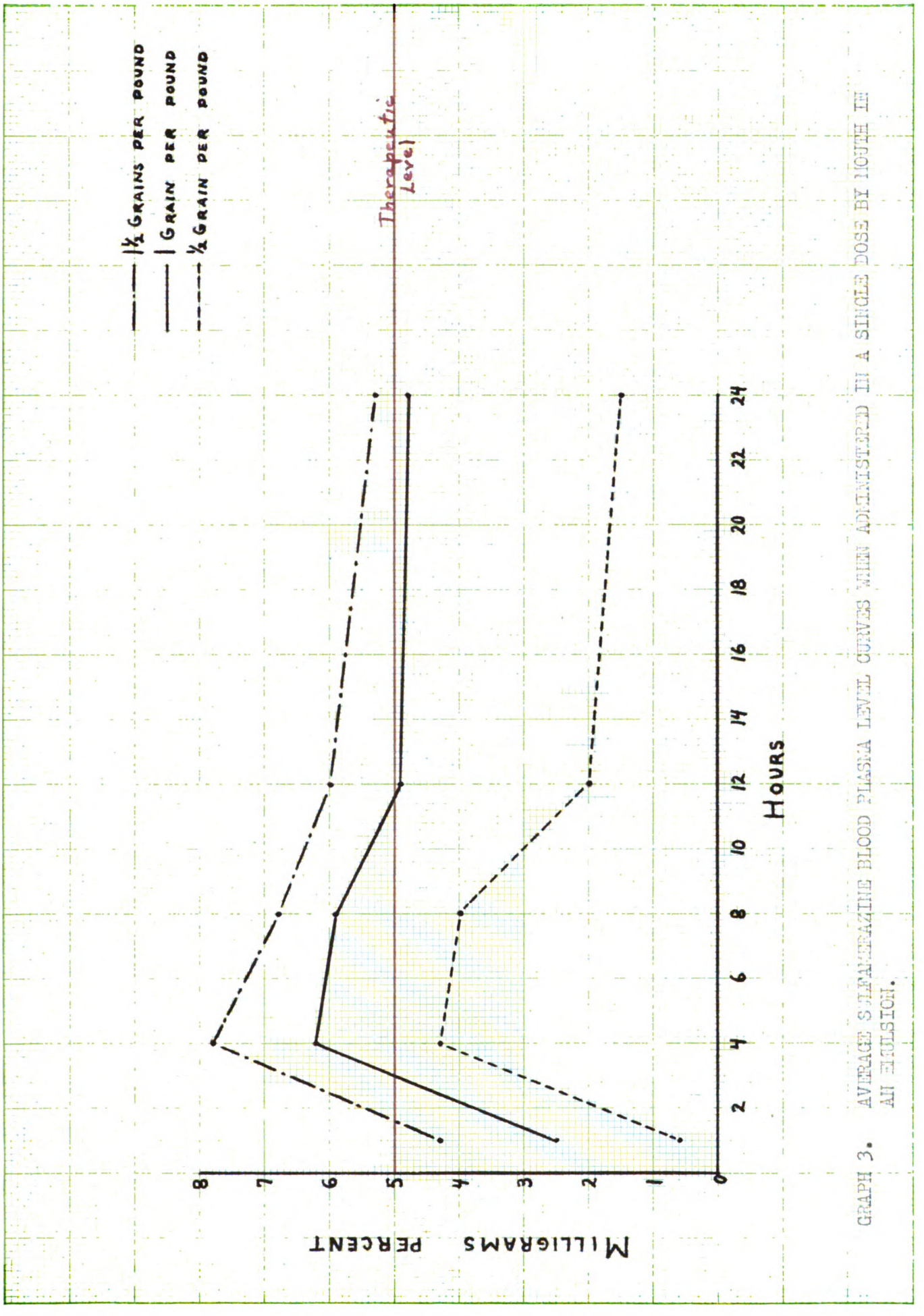
Graph 4 shows the average blood plasma level curves following the single intravenous administration of a 6 percent aqueous solution of sodium sulfamerazine when ½, 1, and 1½ grains per pound of body weight of sulfamerazine were injected. Graph 5 presents the average blood plasma level curves when a 6 percent aqueous solution of sodium sulfamerazine was injected subcutaneously



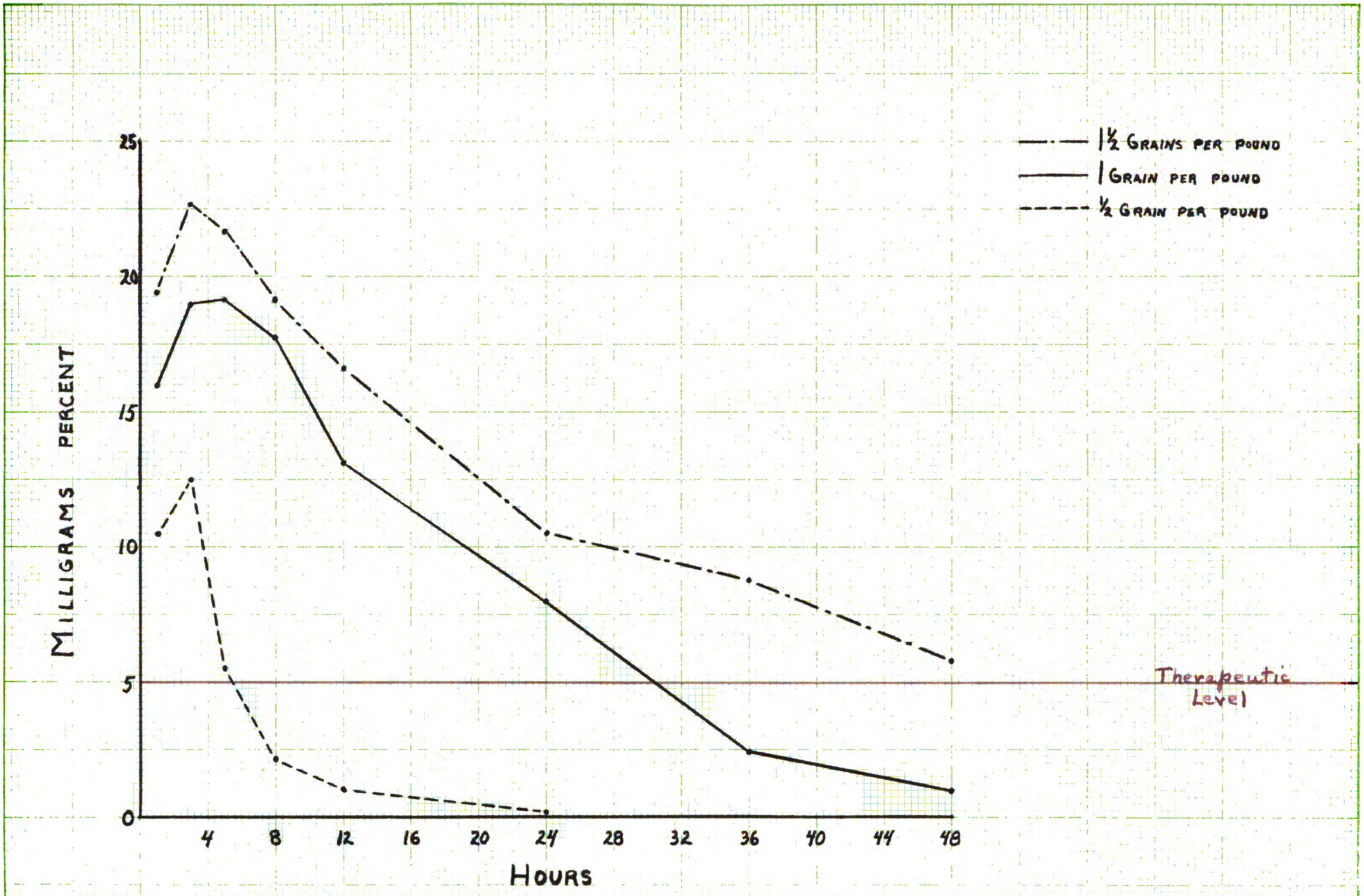
GRAPH 1. AVERAGE SULFATHIAZOLE BLOOD PLASMA LEVEL CURVES WHEN ADMINISTERED IN A SINGLE DOSE BY MOUTH IN TABLET FORM.



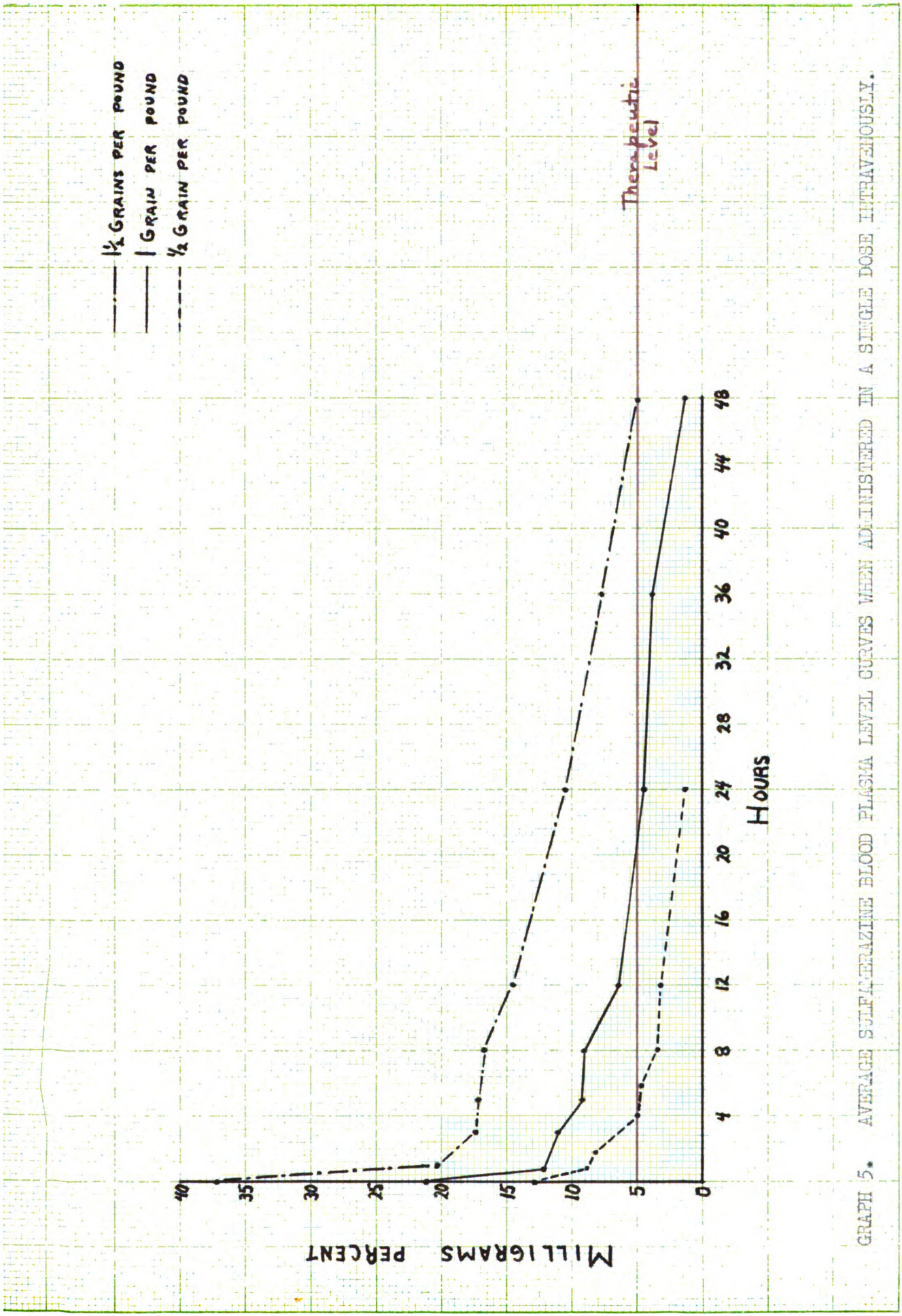
GRAPH 2. AVERAGE SULFANILAMIDE BLOOD PLASMA LEVEL CURVES WHEN ADMINISTERED IN A SINGLE DOSE BY MOUTH IN POWDER FORM.



GRAPH 3. AVERAGE SULFAMERAZINE BLOOD PLASMA LEVEL CURVES WHEN ADMINISTERED IN A SINGLE DOSE BY MOUTH IN AN EMULSION.



GRAPH 4. AVERAGE SULFAMERAZINE BLOOD PLASMA LEVEL CURVES WHEN ADMINISTERED IN A SINGLE DOSE SUBCUTANEOUSLY.



GRAPH 5. AVERAGE SULFADIAZINE BLOOD PLASMA LEVEL CURVES WHEN ADMINISTERED IN A SINGLE DOSE INTRAVENOUSLY.

in dosages of ½, 1, and 1½ grains per pound of body weight. When the sulfamerazine blood plasma curve on canines was compared with the curve obtained with cattle (Scheidy and Tillson, 1947) following the single oral administration of ½ grain per pound of body weight, they were found to be considerably lower than those found in cattle.

From a clinical standpoint the primary interest was 1) to determine the route which was the most convenient for administration, 2) to determine the route or routes which were equally effective or advantageous in maintaining therapeutic sulfamerazine blood plasma levels, and 3) the frequency of administration necessary to maintain therapeutic plasma levels with sulfamerazine.

After analyzing the results of the three routes of administration, the oral route seemed to more nearly meet the clinical qualifications required. The intravenous route was temporarily discarded because of occasional difficulty in administration (objection by dog) but primarily because of the large volume of 6 percent sulfamerazine solution that must be given to maintain sustained blood plasma levels. This latter objection is also encountered in the subcutaneous use of the 6 percent solution.

SULFAMERAZINE ADMINISTERED SEVERAL TIMES PER DAY

The next series of experiments were designed to determine the sulfamerazine blood plasma levels when the drug was administered by mouth once daily, twice daily, and three times daily for a three day period using ½, 1, and 1½ grains per pound of body weight, respectively. Studies on the blood picture were continued.

In the series of experiments where the drug was administered once daily for three days the following dogs were used:

½ grain per pound of body weight - 106

1 grain per pound of body weight - 109

1½ grain per pound of body weight - 112

Tables VI, XXVI, XXVII, and XXVIII.

When administered twice daily for three days the following dogs were used:

½ grain per pound of body weight - 107

1 grain per pound of body weight - 110

1½ grain per pound of body weight - 113

Tables VII, XXIX, XXX, and XXXI.

When administered three times daily for three days the following dogs were used:

½ grain per pound of body weight - 108

1 grain per pound of body weight - 111

1½ grain per pound of body weight - 114

Tables VIII, XXXII, XXXIII, and XXXIV.

Graph 6 shows the average sulfamerazine blood plasma level curves for the three dosages when administered once daily for three days. Graph 7 shows the same for twice daily for three days and Graph 8 shows the blood plasma level curves when sulfamerazine was administered three times daily for three days.

The sulfamerazine blood plasma level curves were found to be quite regular. The height and maintenance of the sulfamerazine concentrations were also found to be directly proportional to the dosage administered. When sulfamerazine was administered orally once

daily for three days it was observed that a slight increase in the blood plasma level occurred on each succeeding administration since normally about 80 percent of the filtered compound is resorbed by the renal tubules of the normal dog (Peters et al., 1944).

Table VI. Blood Plasma Levels Following the Oral Administration of Sulfamerazine Once Daily For Three Days. Dogs No. 106, 109, 112 in Appendix.

Hours After Administration	Gr./lb. body weight administered		
	½	1	1½
	mg. %	mg. %	mg. %
1	0.4	2.6	3.0
4	4.0	6.2	7.4
8	3.8	6.8	8.0
12	2.5	5.5	7.0
24 *	2.0	3.5	5.5
28	4.5	6.4	8.1
32	4.2	6.4	9.0
36	2.7	5.2	7.8
48 *	2.4	4.6	5.6
52	4.9	8.1	9.4
56	4.4	7.9	9.4
60	2.9	7.2	8.0
72	2.5	6.0	6.8

* indicates another administration of the drug.

Table VII. Blood Plasma Levels Following the Oral Administration of Sulfamerazine Twice Daily for Three Days. Dogs No. 107, 110, 113 in Appendix.

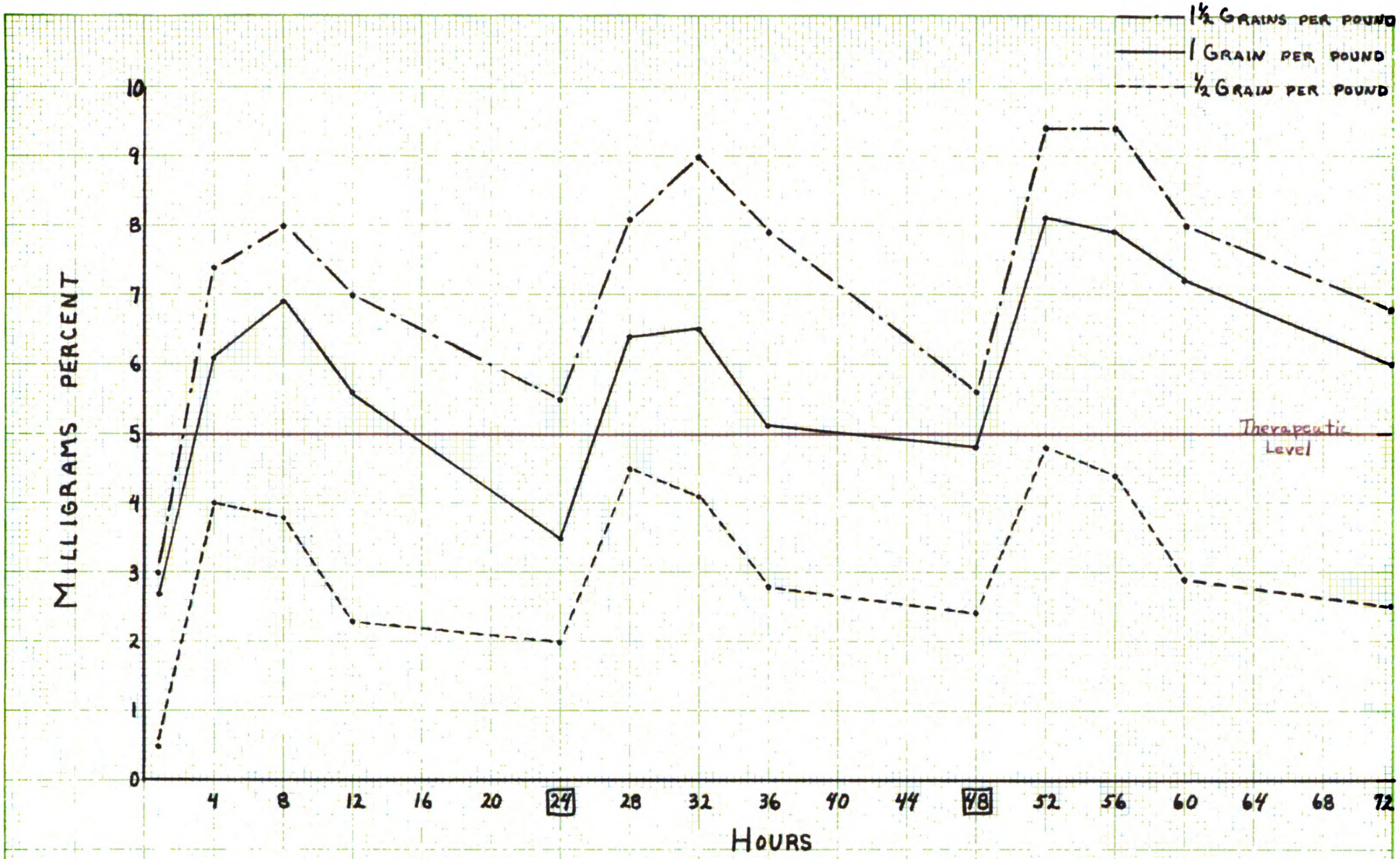
Hours After Administration	Gr./lb. body weight administered		
	½	1	1½
	mg. %	mg. %	mg. %
1	1.0	3.0	3.5
4	4.5	4.9	8.9
8 *	3.2	7.2	10.9
12	11.8	12.8	17.1
24 *	8.0	12.8	17.9
28	12.5	16.4	24.4
32 *	11.4	23.5	26.5
36	16.4	27.8	36.7
48 *	13.8	25.0	34.8
52	15.2	27.9	36.0
56 *	15.0	27.2	35.3
60	18.2	32.1	39.8
72	12.5	29.9	36.5

* indicates another administration of the drug.

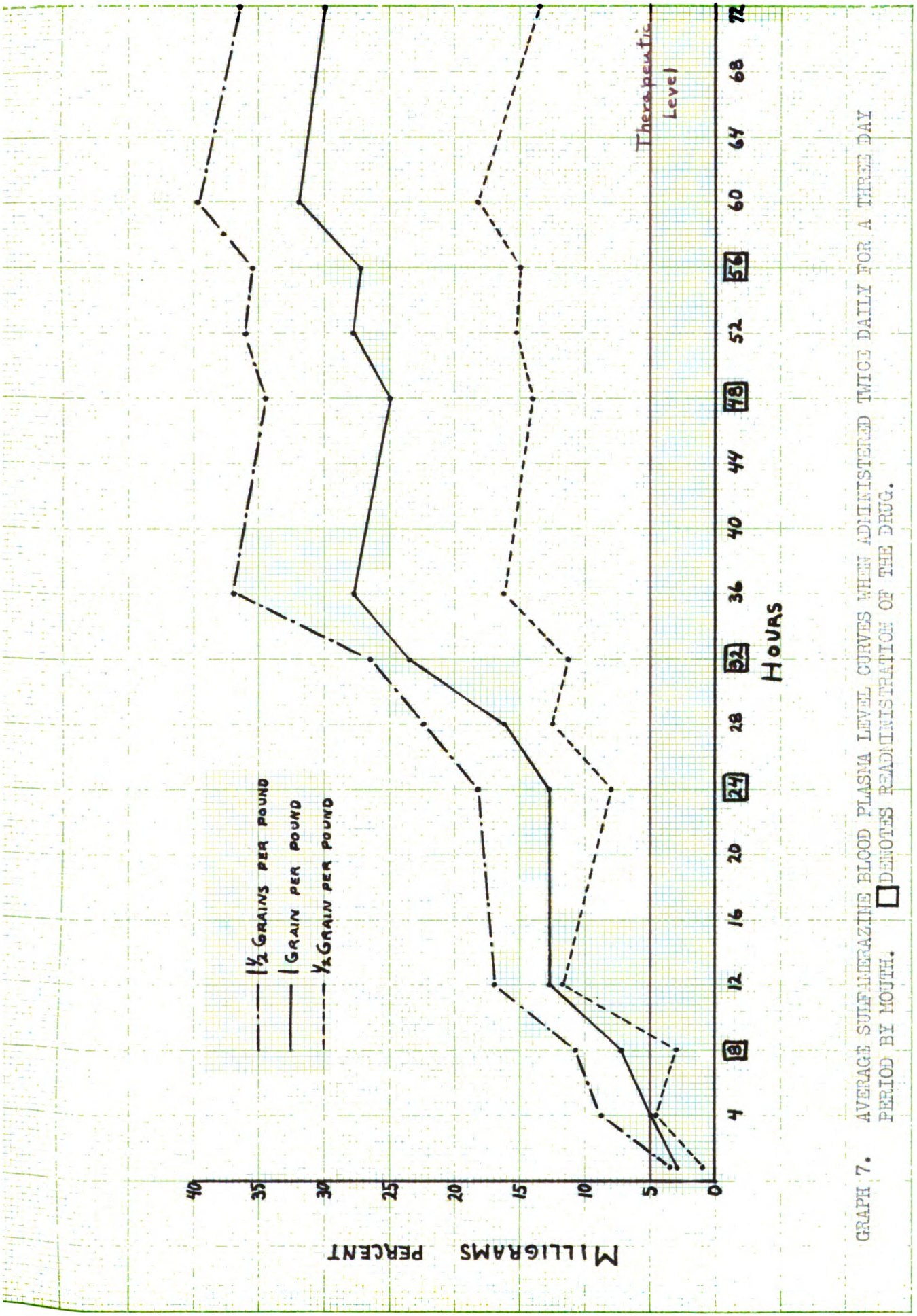
Table VIII. Blood Plasma Levels Following the Oral Administration of Sulfamerazine Three Times Daily for Three Days. Dogs No. 108, 111, 114 in Appendix.

Hours After Administration	Gr./lb. body weight administered		
	½	1	1½
	mg. %	mg. %	mg. %
1	1.8	3.8	5.6
4 *	3.0	5.0	8.4
8 *	6.3	6.8	10.5
12	11.4	14.1	17.3
24 *	6.1	14.1	22.4
28 *	7.4	21.2	24.0
32 *	11.0	27.8	32.0
36	12.4	32.0	38.5
48 *	10.0	29.0	36.2
52 *	13.8	31.9	38.6
56 *	16.0	34.8	40.1
60	19.0	37.1	42.2
72	14.8	32.1	40.8

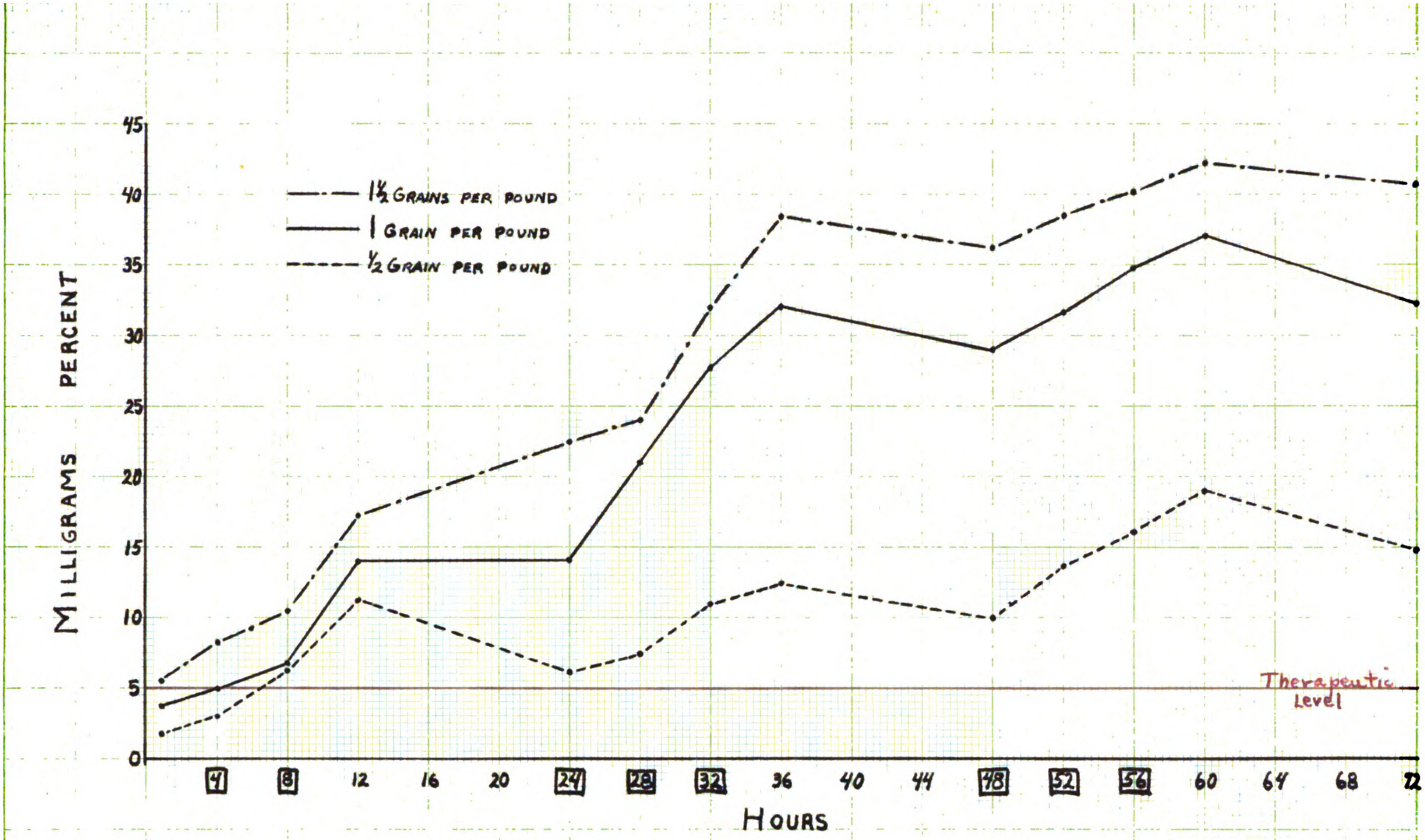
* indicates another administration of the drug.



GRAPH 6. AVERAGE SULFAMERAZINE BLOOD PLASMA LEVEL CURVES WHEN ADMINISTERED ONCE DAILY FOR A THREE DAY PERIOD BY MOUTH. ☐ DENOTES READMINISTRATION OF THE DRUG.



GRAPH 7. AVERAGE SULFAMERAZINE BLOOD PLASMA LEVEL CURVES WHEN ADMINISTERED TWICE DAILY FOR A THREE DAY PERIOD BY MOUTH. □ DENOTES READMINISTRATION OF THE DRUG.



GRAPH 8. AVERAGE SULFAMERAZINE BLOOD PLASMA LEVEL CURVES WHEN ADMINISTERED THREE TIMES DAILY FOR A THREE DAY PERIOD BY MOUTH. ☐ DENOTES READMINISTRATION OF THE DRUG.

The highest concentration found on the second day of administration varied slightly depending on the dosage used. With ½ grain per pound of body weight, the highest concentration was observed four hours following the second daily dosage when 4.5 mg. percent was found. With 1 grain per pound, a blood plasma level of 6.4 mg. percent was obtained at this same bleeding and this plasma level was maintained for another four hours before a decrease in concentration was found while with 1½ grains per pound the plasma level continued to rise until eight hours following the second daily administration when a concentration of 9.0 mg. percent was found. Following the third daily administration, a concentration of 4.9 mg. percent was found in four hours with ½ grain sulfamerazine per pound of body weight after which time the plasma level gradually dropped until 2.5 mg. percent was present seventy-two hours after the initial dose had been given. With 1 grain per pound of body weight a concentration of 8.1 mg. was obtained four hours after the third daily dosage had been administered. This plasma level dropped slowly so that 6.0 mg. percent was still present seventy-two hours following the initial dose. Using 1½ grains per pound of body weight a level of 9.4 mg. percent was obtained four hours after the third daily dose. This level was maintained for another four hours after which time there was a gradual drop with a blood plasma level of 6.8 mg. percent still present seventy-two hours after the initial dose. These data show that to approach a concentration of 5.0 mg. percent, and preferably above 6 mg. percent twenty-four hours a day, which is considered the therapeutic blood plasma level (Welsh et al., 1946) 1½ grains of sulfamerazine per pound of body weight was necessary when administered in a single daily dose per os.

When sulfamerazine was administered by mouth twice daily (9 A.M. and 5 P.M.) for three days the following results were found: The blood plasma concentration made steady increases with both 1 and 1½ grains per pound but ½ grain per pound gave an irregular although constantly increasing plasma curve. As one would expect, the highest concentration obtained was found at sixty hours or four hours after the last dosage had been administered. With ½ grain per pound of body weight a concentration of 4.5 mg. percent was found in four hours while concentrations of 4.9 and 8.9 mg. percent were found with 1 and 1½ grains per pound of body weight respectively. Eight hours after administration the blood concentration with ½ grain per pound had dropped slightly to 3.2 mg. percent while the concentrations with 1 and 1½ grains per pound was 7.2 and 10.9 mg. percent. At this point a second dose was administered causing another increase in blood plasma levels so that in another four hours the concentrations for ½, 1, and 1½ grains per pound of body weight were 11.8, 12.8, and 17.1 mg. percent, respectively. Twelve hours elapsed before another bleeding was made. At this time the blood concentrations were 8.0, 12.8, and 17.9 mg. percent using ½, 1, and 1½ grains per pound. Another dose of sulfamerazine was given immediately. Four hours later (at 28 hours) the blood concentration had reached 12.5, 16.4, and 24.4 mg. percent. At thirty-two hours blood plasma levels 11.4, 23.5, and 26.5 were obtained. The second dosage for that twenty-four hour period was administered. At thirty-six hours blood concentrations of 16.4, 27.8, and 36.7 mg. percent were found; and at forty-eight hours blood plasma levels of 13.8, 25.0, and 34.8 mg. percent were observed. Another dose of sulfamerazine was administered at

this point. Again a slight increase in blood plasma levels were noticed. With ½ grain per pound the plasma level was 15.2 mg., with 1 grain per pound 27.9 mg., while with 1½ grains per pound 36.0 mg. percent was obtained. In another four hours (at 56 hours) the concentrations were 15.0, 27.2, and 35.3 mg. percent respectively. The second dose for that day was given each dog immediately after bleeding. At sixty hours blood concentrations of 18.2, 32.1, and 39.8 mg. percent were found. This represented the highest concentration obtainable with two administrations of sulfamerazine daily. The following morning, at seventy-two hours, the blood plasma levels had dropped to 12.5, 29.9, and 36.5 mg. percent. The data reported herein show that therapeutic blood plasma level curves may be obtained with the administration of ½ grain of sulfamerazine per pound of body weight twice daily to normal dogs.

When sulfamerazine was administered by mouth three times a day (9 A.M., 1 P.M., and 5 P.M.) for three days, that is, 1½, 3, and 4½ total grains per pound of body weight daily, the following results were found: With the exception of the administration of ½ grain of sulfamerazine per pound of body weight, there was a very steady and increasing blood plasma concentration. When ½ grain per pound was administered there was an irregular although increasing plasma level curve. Again it was observed that the highest blood plasma concentration was found at sixty hours after the initial dose had been given. Four hours after administration concentrations of 3.0, 5.0, and 8.4 mg. percent were found when sulfamerazine had been given in dosages of ½, 1, and 1½ grains per pound respectively. Sulfamerazine was again administered in the same dosages. In another four

hours blood plasma levels of 6.3, 6.8, and 10.5 mg. percent were found. Again sulfamerazine was administered. At twelve hours blood plasma levels of 11.4, 14.1, and 17.3 mg. percent respectively were observed. No more sulfamerazine was administered until the next morning. Blood samples prior to another dosage of sulfamerazine gave blood plasma levels of 6.1, 14.1, and 22.4 mg. percent. As shown by these figures, the blood concentration when ½ grain of sulfamerazine per pound of body weight was administered was eliminated rather rapidly. With 1 grain per pound the concentration remained the same but when 1½ grains per pound were given, the blood plasma level continued to increase. These results are comparable to those obtained when sulfamerazine was administered twice daily in the same dosages. The first dose of the second day was given. In four hours or twenty-eight hours after the initial dose had been given the blood plasma concentrations respectively were 12.5, 21.2, and 24.0 mg. percent. Another dose of sulfamerazine was administered orally. At 32 hours, blood plasma levels were 11.0, 27.6, and 32.0 mg. percent when ½, 1, and 1½ grains per pound respectively were administered. Again each dog was redosed. At 36 hours blood plasma levels of 12.4, 32.0, and 38.5 mg. percent were found. No more sulfamerazine was administered and no blood samples were drawn until the next morning when the blood contained 10.0, 29.0, and 36.2 mg. percent of sulfamerazine. This represented a slight drop in the blood concentration of the drug overnight. The dogs were redosed immediately after this forty-eight hour bleeding. At 52 hours the blood plasma level had again started to rise. At this time the blood concentrations obtained were 16.0, 34.8, and 38.6 mg. percent respectively. The dogs were again redosed after the

drawing of the blood samples. At 56 hours with the blood plasma levels still rising, 16.0, 34.8, and 40.1 mg. percent were observed. Following this bleeding the third dose for the day was administered and in four hours, or sixty hours from the initial dosage, the highest concentrations were obtained. These blood plasma concentrations were 19.0, 37.1, and 42.2 mg. percent respectively. With no more sulfamerazine administered, the blood plasma concentration slowly dropped so that at seventy-two hours after the initial dosage had been given, plasma concentrations of 14.8, 32.1, and 40.8 mg. percent were found. Considering 5 mg. percent as a therapeutic level, one can readily see that it is unnecessary to administer sulfamerazine three times a day to maintain therapeutic blood concentrations.

The same brand of sulfamerazine was used throughout the course of this study, including the tablet form, powder form, emulsion, and sterile powder for the preparation of intravenous and subcutaneous solutions. All three types of sulfamerazine products given orally gave similar blood plasma level curves. The first study, comparing the single oral administration of ½, 1, and 1½ grains per pound, was begun March 1, 1948 and was completed August 6, 1948. The study comparing the various dosages given either intravenously or subcutaneously as well as the oral administration of the drug for a three day period was begun August 9, 1948 and completed September 3, 1948.

Blood studies were made at specified intervals with each dosage of sulfamerazine administered. The results of each trial were further tabulated to find out if there was a trend towards lowering or raising of the cellular constituents following the administration of sulfamerazine. It was found that no changes occurred in the

red and white cell counts, hemoglobin content, and differential leukocyte counts when sulfamerazine was administered at the rate of ½, 1, or 1½ grains per pound of body weight once daily for three days. Blood changes did not occur even when sulfamerazine was administered by mouth at the rate of 1½ grains (214.5 mg. per kilogram) per pound of body weight daily in three divided doses for a three day period. Similar results were obtained when the drug was administered at the rate of 2 grains (286 mg. per kilogram) per pound of body weight daily in two divided doses for a three day period. Fifty-two hours after the initial dose of sulfamerazine had been administered, when given at the rate of 3 grains (429.0 mg. per kilogram) per pound of body weight daily in three divided doses, a leukopenia developed. The leukocytic count dropped 39 percent at seventy-two hours. At the same time no changes in the hemoglobin content were found but a slight monocytosis developed as the drug was continued. When 3 grains per pound of body weight were administered daily in two divided doses for three days a slight leukocytosis developed in twenty-four hours which gradually decreased. A slight hemoglobinemia and neutropenia were found at seventy-two hours. Upon increasing the dosage to 4½ grains (643.5 mg. per kilogram) per pound of body weight daily in three divided doses, it was found that a marked leukopenia developed eight hours after the initial dose had been given. This drop in the white cell count continued throughout the course of administration of the drug until, at seventy-two hours, the leukocyte count had decreased 60 percent from normal. Hemoglobin changes that occurred were significant. Although the red cell count remained fairly constant, the hemoglobin content decreased markedly as the dosages were continued until it had

dropped 28 percent, (Table XXXIV). Analysis of the cellular constituents showed a marked neutropenia and lymphocytosis. With this dosage normal healthy dogs showed anorexia which lasted several days after the course of administration of the drug.

It was interesting to observe the blood changes when sulfamerazine was administered either intravenously or subcutaneously. Both routes of injections produced a leukocytosis even when ½ grain per pound of body weight was administered in a single injection. No major changes were found in the hemoglobin content or in the cellular constituents except a very slight hemoglobinemia of approximately 20 percent when sulfamerazine was injected intravenously in single 1 and 1½ grain per pound doses. Also a slight neutropenia was found when 1½ grains per pound of the drug was injected intravenously.

Two hundred forty-six sulfamerazine trials were run during the course of this experiment. In 85 of these, blood samples were taken at specific intervals on each individual trial and checked for red and white cell counts and hemoglobin content. Differential leukocyte counts were made on 69 of the dogs.

The greatest difference between the initial and final count in any one trial was 990,000 red blood cells per cu. mm., 7,250 white blood cells per cu. mm., and 5.2 grams of hemoglobin. On the differential counts, in any one given trial, the largest difference between initial and final counts on neutrophils was 16 cells, lymphocytes 15 cells, monocytes 6 cells, basophils 4 cells, and eosinophils 5 cells.

Berkson et al., (1940) stated that following the usual practice of taking twice the standard error as significant limits, the

erythrocyte count is determined significantly within ± 16 percent. Similarly, for a leukocyte count the count is determined significantly within ± 21 percent.

Each trial was further tabulated to show the trend towards lowering and raising of the cellular constituents and hemoglobin content following the administration of sulfamerazine. Results of this tabulation are shown in Table XXXV.

Table XXXV. Blood Picture Following the Administration of Sulfamerazine Compared with the Initial Blood Picture.

Test	No. of Trials	No. showing decrease	Trials showing increase	Trials showing identical results
RBC	85	53	31	1
WBC	85	33	50	2
Hemoglobin	85	53	22	10
% Neutrophils	69	42	23	4
% Lymphocytes	69	32	34	3
% Monocytes	69	12	41	16
% Basophils	69	7	33	29
% Eosinophils	69	23	29	17

The results reveal a noticeable decrease in erythrocytes, hemoglobin content, and neutrophils and an increase in leukocytes, monocytes, and basophils. In many of the trials the increase or decrease was almost negligible.

The greatest variation in the red cell count obtained in any animal was ± 450,000; in the majority there was very little

change. There was a definite change in the white cell count. Smaller doses gave no significant change but increased dosages of sulfamerazine produced a leukopenia. Hemoglobin changes were rather insignificant except where larger doses were administered for several days when a hemoglobinemia was produced; this varied as much as 5.2 grams per 100 ml. of blood. Marked changes were found in the cellular constituents as the dosage of sulfamerazine was increased. There was a decrease in the percent of neutrophils, an increase in the percent of lymphocytes and a slight increase in the percent of monocytes as the dosage of the drug was increased. From these results it was concluded that small doses of sulfamerazine were non-toxic but when doses of two or more grains per pound of body weight were administered, a definite tendency towards toxicity resulted. The range of blood constituents in the latter trials was not found to be within the normal variations reported by Coffin (1945), Malkamus (1944), and Boddie (1946). Results of this tabulation are shown in Tables XXXVI and XXXVII.

The oral route of administration was found to be the one of choice for ease of administration. Either the tablet form, powder form, or the emulsion may be used with equal ease. With vicious dogs where placing the hand inside the dog's mouth is undesirable, the emulsion is preferred since it may be administered while the dog is muzzled. The subcutaneous and intravenous routes of injection are impractical with sodium sulfamerazine since large doses are required and the dogs showed more objection to these routes when given orally.

Only one dog exhibited any local or systemic reactions

Dog No.	Dosage Sulfamerazine (grains per lb.)	Pre-medication R.B.C.	W.B.C.	Hb.	Post-medication R.B.C.	W.B.C.	Hb.
102	½ once (Tablet)	7.02	13.1	15.2	7.1-6.5	12.9-13.0	14.8-15.2
105	½ " (Powder)	5.47	10.9	18.0	5.1-5.28	10.7-11.25	17.3-17.6
118	½ " (Emulsion)	6.34	12.5	14.0	6.12-6.28	12.4-12.6	12.8-13.1
124	½ " (Intravenous)	6.34	12.5	14.0	5.55-6.2	16.2-23.8	12.4-13.9
121	½ " (Subcutaneous)	5.47	10.9	18.0	5.12-5.57	11.35-13.3	17.3-17.9
103	1 " (Tablet)	6.75	9.2	15.0	6.57-6.89	9.2-10.2	15.0-15.5
116	1 " (Powder)	4.26	9.7	14.5	4.47-4.7	9.5-9.9	14.3-14.5
119	1 " (Emulsion)	4.26	9.7	14.5	4.05-4.14	9.7-9.95	13.0-14.1
125	1 " (Intravenous)	6.75	9.2	15.0	5.94-6.22	10.55-11.15	12.0-12.6
122	1 " (Subcutaneous)	5.47	10.9	18.0	5.34-5.87	13.2-14.0	16.4-16.9
105	1½ " (Tablet)	6.34	12.5	14.0	5.25-6.25	10.75-11.75	13.3-14.2
117	1½ " (Powder)	5.7	12.1	16.5	5.5-6.1	12.35-13.35	16.5-16.7
120	1½ " (Emulsion)	6.34	12.5	14.0	5.97-6.1	12.35-11.9	13.9-14.0
126	1½ " (Intravenous)	6.34	12.5	14.0	6.79-6.02	13.75-16.1	14.1-11.4
123	1½ " (Subcutaneous)	6.18	9.9	15.0	5.9-6.53	12.35-12.9	13.2-14.1
106	½ once daily 3 days*	4.26	9.7	14.5	4.63-4.85	9.9-10.1	15.2-16.2
107	½ twice " " " *	5.7	12.1	16.5	5.07-5.65	10.7-11.9	13.4-15.7
108	½ 3 times" " " *	6.75	9.2	15.0	6.0-6.38	9.05-10.0	13.7-15.4
109	1 once " " " *	6.18	9.9	15.0	6.2-6.87	9.8-10.6	15.2-16.5
110	1 twice " " " *	4.26	9.7	14.5	4.23-4.51	9.05-10.4	13.9-15.3
111	1 3 times" " " *	5.7	12.1	16.5	5.67-6.67	14.5-7.45	15.7-15.3
112	1½ once " " " *	6.75	9.2	15.0	6.55-5.99	12.7-8.1	15.2-14.1
113	1½ twice " " " *	6.18	9.9	15.0	6.75-7.1	15.4-11.45	15.7-13.7
114	1½ 3 times" " " *	5.7	12.1	16.5	5.49-6.09	11.6-4.85	15.9-12.0

* denotes administration of the drug in tablet form

TABLE XXXVI. BLOOD PICTURE IN DOGS FOLLOWING THE ADMINISTRATION OF SULFANILAMIDE COMPARED WITH THE BLOOD PICTURE IN NORMAL DOGS.

Dog No.	Dosage Sulfamerazine (grains per lb.)	Pre-medication N.%	L.%	M.%	B.%	E.%	Post-medication N.%	L.%	M.%	B.%	E.%
102	½ once (Tablet)	65	18	11	0	6	67-69	19-21	6-10	0	3-5
115	½ " (Powder)	64	30	1	0	5	64	28	3	1	4
118	½ " (Emulsion)	74	24	0	0	2	72	25	0	0	3
124	½ " (Intravenous)	74	24	0	0	2	67-73	21-32	0-3	0-2	0-4
121	½ " (Subcutaneous)	64	30	1	0	5	69-75	19-28	1-5	0-1	1-3
103	1 " (Tablet)	68	28	1	1	2	68-76	23-32	0	1-2	1-3
116	1 " (Powder)	68	31	0	1	0	70	28	0	1	1
119	1 " (Emulsion)	68	31	0	1	0	66	40	2	2	0
125	1 " (Intravenous)	68	28	1	1	2	64	29	3	2	2
122	1 " (Subcutaneous)	64	30	1	0	5	63	32	0	1	4
105	1½ " (Tablet)	74	24	0	0	2	70-75	24-28	0	0	1-2
117	1½ " (Powder)	64	36	0	0	0	68	29	2	0	1
120	1½ " (Emulsion)	74	24	0	0	2	67	26	3	3	1
126	1½ " (Intravenous)	74	24	0	0	2	63-78	19-28	0-4	0-3	1-3
123	1½ " (Subcutaneous)	66	33	1	0	0	68	29	2	0	1
106	½ once daily 3 days*	68	31	0	1	0	57-65	31-39	1-2	1-2	2-3
107	½ twice " " " *	64	36	0	0	0	63-68	28-31	1-4	0-2	0-2
108	½ 3 times" " " *	68	28	1	1	2	61-68	22-33	1-4	1-3	2-3
109	1 once " " " *	66	33	1	0	0	63-65	24-30	2-6	1-4	2-5
110	1 twice " " " *	68	31	0	1	0	62-67	27-32	0-4	0-4	0-3
111	1 3 times" " " *	64	36	0	0	0	57-66	29-35	0-6	1-4	0-1
112	1½ once " " " *	68	28	1	1	2	62-66	27-34	0-2	0-2	0-4
113	1½ twice " " " *	66	33	1	0	0	53-62	34-38	1-4	1-4	1-3
114	1½ 3 times" " " *	64	36	0	0	0	48-69	25-51	1-5	0	0-1

N. - Neutrophils L. - Lymphocytes M. - Monocytes B. - Basophils E. - Eosinophils.
* denotes administration of the drug in tablet form.

TABLE XXXVII. BLOOD PICTURE IN DOGS SHOWING THE CELLULAR CONSTITUENTS FOLLOWING THE ADMINISTRATION OF SULFAMERAZINE COMPARED WITH THE BLOOD PICTURE IN NORMAL DOGS.

following the administration of sulfamerazine. Anorexia and depression resulted from the oral administration of 4½ grains (643.5 mg. per kilogram) per pound of body weight daily in three divided doses for a three day period.

SUMMARY AND CONCLUSIONS

The dogs exhibited the least objection to oral administration and the most objection to the intravenous route when sulfamerazine was injected. Sulfamerazine blood plasma levels following oral administration were found to be slightly higher and persisted for a longer period of time than those following the intravenous or subcutaneous routes of administration. A concentration of 5 mg. percent (therapeutic level) or higher was maintained for an average of 20 hours following a single oral administration of 1 grain (143 mg. per kilogram) per pound of body weight, and 22 hours following 1½ grains (214.5 mg. per kilogram) per pound. Therapeutic blood levels were not obtained for a sufficiently long period of time by a single oral dosage of ½ grain (71.5 mg. per kilogram) per pound of body weight in the dog.

Therapeutic blood levels of free sulfamerazine were maintained over a prolonged period when given either subcutaneously or intravenously only when 1 or 1½ grains per pound of body weight (1.1 and 1.6 ml. per pound respectively) were administered. These two methods of administration were more objectionable to the dog than the oral administration.

Sulfamerazine blood plasma levels of 5 mg. percent or higher were maintained with the administration of ½ grain (71.5 mg. per kilogram) per pound twice daily per os. Larger and/or more frequent doses produced an unnecessarily high blood concentration.

There was no significant difference between the oral

use of sulfamerazine in the tablet form, in powder form, or in an emulsion in the dog as far as blood plasma levels were concerned.

Studies of the blood picture indicate that the greatest variation in the red cell count obtained in any animal was ± 450,000; in the majority there was very little change. Increased dosages of sulfamerazine during a three-day study produced a leukopenia in the normal healthy dog. Intravenously the trend was towards a leukocytosis. Hemoglobinemia was produced when sulfamerazine was administered by mouth at the rate of 4½ grains (643.5 mg. per kilogram) per pound of body weight daily in three divided doses for a three day period. With the larger doses, the dogs showed a marked anorexia and depression which lasted several days after the administration of the drug.

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Table IX — Determinations on Dog No. 101 — 3/1/48

Route - Oral (Tablet) — Male — 52 lbs. — 2 yrs.

Dose - ½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm.	tubular	R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	1.6	1.3								
2 hr.	2.0	1.7								
3 hr.	2.8	2.4								
4 hr.	3.4	2.6	5,540,000	10,400	15.5	78	19	1		2
5 hr.	3.4	2.6								
6 hr.	3.4	2.8								
7 hr.	3.3	2.1								
8 hr.	3.2	2.0								
12 hr.	2.0	0.9	5,690,000	12,600	16.5	80	19			1
24 hr.	1.6	0.8	5,700,000	11,900	17	80	18			2

Sulfamerazine mg. % - Milligrams of sulfamerazine per 100 ml. of blood plasma
R.B.C. - Red blood cells
W.B.C. - White Blood cells
Hb. g./100 ml. - Grams of hemoglobin per 100 ml. of blood
N. - Neutrophils L. - Lymphocytes M. - Monocytes B. - Basophils E. - Eosinophils

Table X — Determinations on Dog No. 102 — 3/13/48

Route - Oral (Tablet) — Male — 38 lbs. — 2 yrs.

Dose - ½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	N.	Differential L.	M.	B.	E.
1 hr.	1.9	1.6								
2 hr.	2.2	1.9								
3 hr.	3.0	2.6								
4 hr.	3.4	2.8	6,850,000	12,900	15	68	21	6		5
5 hr.	3.4	2.8								
6 hr.	3.3	2.6								
7 hr.	3.2	2.4								
8 hr.	3.0	2.0	7,100,000	13,000	15.2	69	19	8		4
12 hr.	2.0	1.0								
24 hr.	1.8	0.9	6,500,000	12,900	14.8	67	20	10		3

Table XI — Determinations on Dog No. 103 — 6/3/48

Route - Oral (Tablet) — Male — 56 lbs. — 3 yrs.

Dose - 1 gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm.	tubular	R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	1.8	1.5	6,850,000	10,100	15	75	21		1	3
2 hr.	5.0	3.5								
3 hr.	5.8	4.0								
4 hr.	6.5	4.3	6,575,000	9,800	15	72	28			
5 hr.	6.4	5.6								
6 hr.	6.2	5.4								
7 hr.	5.6	4.9	6,750,000	9,600	15	65	32		2	1
8 hr.	5.1	3.9								
12 hr.	4.2	2.9	6,740,000	9,200	15	76	23			1
24 hr.	3.2	2.0	6,890,000	10,200	15.5	68	30			2
48 hr.	0.8	1.0								

Table XII — Determinations on Dog No. 104 — 8/6/43

Route - Oral (Tablet) — Male — 25 lbs. — 15 mos.

Dose - ½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	4.9	4.5								
2 hr.	6.1	5.4								
3 hr.	8.2	8.0								
4 hr.	6.4	5.6	5,840,000	11,100	17.5	71	28	1		
5 hr.	6.1	5.4								
6 hr.	6.1	5.4								
7 hr.	6.1	5.4	5,700,000	11,700	17.5	71	29			
8 hr.	6.1	5.3								
12 hr.	5.6	4.4								
24 hr.	5.0	3.5	5,730,000	12,350	18	69	29	2		

Table XIII — Determinations on Dog No. 105 — 8/6/43

Route - Oral (Tablet) — Male — 29 lbs. — 15 mos.

Dose - 1½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfanerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	1.8	1.5								
2 hr.	3.0	2.6								
3 hr.	4.6	3.9								
4 hr.	5.8	5.0	6,250,000	10,750	13.8	75	24			1
5 hr.	5.8	5.4								
6 hr.	5.6	5.0								
7 hr.	5.4	4.8	6,120,000	11,300	14.2	74	26			
8 hr.	4.7	4.2								
12 hr.	4.0	3.6								
24 hr.	3.0	2.8	6,250,000	11,750	14.2	70	28			2

Table XIV — Determinations on Dog No. 115 — 8/20/43

Route - Oral (Powder) — Male — 25 lbs. — 15 mos.

Dose - ½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R. B. C.	W.B.C.	Hb. g./100 ml.	N.	L.	M.	B.	E.
1 hr.	1.2	1.5	5,280,000	10,700	17.6					
4 hr.	4.8	5.0								
8 hr.	4.2	4.9								
12 hr.	2.5	2.5								
24 hr.	2.0	2.2	5,100,000	11,250	17.3	64	28	3	1	4

Table XV — Determination on Dog No. 116 — 8/30/48

Route - Oral (Powder) — Female — 37 lbs. — 2½ yrs.

Dose - 1 gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	2.5	3.1	4,700,000	9,500	14.3					
4 hr.	6.6	7.0								
8 hr.	6.2	6.5								
12 hr.	5.5	5.8								
24 hr.	4.5	4.7	4,470,000	9,900	14.5	70	28		1	1

Table XVI — Determination on Dog No. 117 — 8/30/48

Route - Oral (Powder) — Male — 55½ lbs. — 3 yrs.

Dose - 1½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	3.5	3.5	5,500,000	12,350	16.5					
4 hr.	7.2	7.0								
8 hr.	7.1	7.2								
12 hr.	6.5	6.2								
24 hr.	5.0	5.3	6,100,000	13,350	16.7	68	29	2		1

Table XVII — Determination on Dog No. 118 — 8/20/48

Route - Oral (Emulsion) — Male — 29 lbs. — 15 mos.

Dose - $\frac{1}{2}$ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	0.6	1.3	6,120,000	12,400	13.1					
4 hr.	4.3	4.8								
8 hr.	4.0	4.1								
12 hr.	2.0	1.9								
24 hr.	1.5	1.8	6,280,000	12,600	12.8	72	25			3

Table XVIII — Determination on Dog No. 119 — 8/23/43

Route - Oral (Emulsion) — Female — 37½ lbs. — 2½ yrs.

Dose - 1 gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	2.5	2.7	4,050,000	9,950	14.1					
4 hr.	6.2	6.5								
8 hr.	5.9	6.0								
12 hr.	4.9	5.1								
24 hr.	4.8	4.8	4,140,000	9,700	13.0	66	40	2	2	

Table XIX — Determination on Dog No. 120 — 9/1/48

Route - Oral (Emulsion) — Male — 28½ lbs. — 15 mos.

Dose - 1½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	4.3	4.6	6,100,000	12,350	14					
4 hr.	7.8	8.2								
8 hr.	6.8	7.2								
12 hr.	6.0	6.4								
24 hr.	5.3	5.5	5,970,000	11,900	13.9	67	26	3	3	1

Table XX — Determination on Dog No. 121 — 8/13/48

Route - Subcutaneous — Male — 25 lbs. — 15 mos.

Dose - ½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	N.	Differential L.	M.	B.	E.
1 hr.	10.5	11.0	5,360,000	13,300	17.8	69	27	1		3
3 hr.	12.5	12.9	5,120,000	11,650	17.3	72	23	2	1	2
5 hr.	5.4	6.5	5,460,000	11,350	17.6	75	19	5		1
8 hr.	2.1	3.5								
12 hr.	1.0	2.0								
24 hr.	0.2	1.1	5,570,000	12,100	17.9	70	28	1		1

Table XXI — Determination on Dog No. 122 — 8/27/48

Route - Subcutaneous — Male — 25 lbs. — 15 mos.

Dose - 1 gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	16.0	16.0	5,870,000	13,200	16.4					
3 hr.	19.0	18.9								
5 hr.	19.2	19.2								
8 hr.	17.6	17.9								
12 hr.	13.1	13.3								
24 hr.	8.0	8.8	5,340,000	14,000	16.9	63	32		1	4
36 hr.	2.4	2.6								
48 hr.	1.0	1.3								

Table XXII — Determinations on Dog No. 123 — 8/25/48

Route - Subcutaneous — Male — 46 lbs. — 3 yrs.

Dose - 1½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	19.5	19.7	6,530,000	12,350	13.2					
3 hr.	22.7	22.2								
5 hr.	21.8	21.8								
8 hr.	19.2	19.2								
12 hr.	16.6	17.2								
24 hr.	10.5	11.0	5,900,000	12,900	14.1	68	29	2		1
36 hr.	8.7	8.8								
48 hr.	5.8	6.0								

Table XXIII — Determinations on Dog No. 124 — 8/13/48

Route - Intravenous — Male — 29 lbs. — 15 mos.

Dose - ½ gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	N.	Differential L.	M.	B.	E.
0 hr.	13.0	13.9								
1 hr.	9.0	11.2	5,720,000	17,100	13.9	67	32	1		
2 hr.	8.2	10.6	5,550,000	16,200	12.6	73	22	2	2	1
4 hr.	5.0	6.6	5,575,000	17,400	12.4					
6 hr.	4.7	6.5								
8 hr.	3.5	5.9								
12 hr.	3.2	4.0	6,200,000	13,800	13.2	70	21	3	2	4
24 hr.	1.4	2.5	5,850,000	14,800	12.5	71	27		1	1

Table XXIV — Determinations on Dog No. 125 — 8/25/48

Route - Intravenous — Male — 55½ lbs. — 3 yrs.

Dose - 1 gr./lb. body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
0 hr.	21.3	22.0								
1 hr.	12.3	14.1	5,940,000	10,550	12.0					
3 hr.	11.1	13.3								
5 hr.	9.4	11.4								
8 hr.	9.0	9.5								
12 hr.	6.8	7.4								
24 hr.	4.4	5.0	6,220,000	11,150	12.6	64	29	3	2	2
36 hr.	3.7	3.9								
48 hr.	1.4	1.7								

Table XIV — Determinations on Dog No. 126 — 8/27/43

Route - Intravenous — Male — 28½ lbs. — 15 mos.

Dose - 1½ gr./lb body weight — Times per day - once — No. days - one

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	N.	Differential L.	M.	B.	E.
0 hr.	37.1	35.2								
1 hr.	20.1	20.3								
3 hr.	17.8	18.1								
5 hr.	17.4	17.2	6,350,000	13,750	14.1	71	26	1		2
8 hr.	16.9	16.9	6,790,000	14,800	13.8	78	19		2	1
12 hr.	14.5	14.8	6,400,000	15,650	12.3	65	27	3	2	3
24 hr.	10.5	10.9	6,020,000	16,100	11.4	63	28	4	3	2
36 hr.	7.7	8.0								
48 hr.	5.0	5.0								

Table XXVIII — Determinations on Dog No. 112 — 8/18/43

Route - Oral (Tablet) — Male — 57 lbs. — 3 yrs.

Dose - ½ gr./lb. body weight — Times per day - once — No. days - three

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	N.	Differential L.	M.	B.	E.
1 hr.	3.0	2.8								
4 hr.	7.4	7.0								
8 hr.	8.0	7.2								
12 hr.	7.0	5.9								
24 hr.*	5.5	5.0	6,550,000	11,200	15.2	62	34	2	2	
28 hr.	8.1	7.5								
32 hr.	9.0	8.1								
36 hr.	7.8	7.1								
48 hr.*	5.6	6.0	6,470,000	12,700	15.2	65	27	2	2	4
52 hr.	9.4	10.0								
56 hr.	9.4	9.0								
60 hr.	8.0	7.1								
72 hr.	6.3	6.6	5,990,000	8,100	14.1	66	32			2

* indicates another administration of the drug.

Table XXVII — Determinations on Dog No. 109 — 8/11/48

Route - Oral (Tablet) — Male — 46 lbs. — 3 yrs.

Dose - 1 gr./lb. body weight — Times per day - once — No. days - three

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm.	tubular	R.B.C.	W.B.C.	Hb. g./100 ml.	N.	Differential L.	M.	B.	E.
1 hr.	2.6	2.4								
4 hr.	6.2	6.5	6,870,000	9,800	16.5	64	30	2	1	3
8 hr.	6.8	7.0								
12 hr.	5.5	5.4								
24 hr.*	3.5	4.0	6,420,000	10,600	15.2	65	24	6	4	1
28 hr.	6.4	6.6								
32 hr.	6.4	7.0								
36 hr.	5.2	6.2								
48 hr.*	4.6	5.1	6,300,000	10,200	15.4	65	28	3	2	2
52 hr.	8.1	9.0								
56 hr.	7.9	8.5								
60 hr.	7.2	8.5								
72 hr.	6.0	7.0	6,200,000	10,000	15.2	63	30	3	2	2

* indicates another administration of the drug.

Table XXX — Determinations on Dogs No. 110 — 8/16/48

Route - Oral (Tablet) — Female — 40 lbs. — 2½ yrs.

Dose - 1 gr./lb. body weight — Times per day - two — No. days - three

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine Mg. % 1 cm.	tubular	R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	3.0	2.5								
4 hr.	4.9	4.5								
8 hr.*	7.2	6.4								
12 hr.	12.8	14.0								
24 hr.*	12.8	13.8	4,380,000	9,050	13.9	64	32	2	1	1
28 hr.	16.4	16.0								
32 hr.*	23.5	23.0								
36 hr.	27.8	26.5								
48 hr.*	25.0	24.4	4,510,000	10,400	14.9	67	30		1	2
52 hr.	27.9	26.0								
56 hr.*	27.2	26.5								
60 hr.	32.1	30.0								
72 hr.	29.9	28.9	4,230,000	9,300	15.3	62	27	4	4	3

* indicates another administration of the drug.

Table XXXI — Determinations on Dog No. 113 — 8/18/48

Route - Oral (Tablet) — Male — 46 lbs. — 3 yrs

Dose - 1½ gr./lb. body weight — Times per day - two — No. days - three

History - Apparently healthy

Blood Studies

Time Interval	Sulfamerazine mg. % 1 cm.	tubular	R.B.C.	W.B.C.	Hb. g./100 ml.	N.	Differential L.	M.	B.	E.
1 hr.	3.5	3.0								
4 hr.	8.9	8.1								
8 hr.*	10.9	10.3								
12 hr.	17.1	16.0								
24 hr.*	17.9	18.1	6,930,000	15,400	15.7	53	38	4	4	1
28 hr.	24.4	22.0								
32 hr.*	26.5	27.0								
36 hr.	36.7	36.2								
48 hr.*	34.8	35.0	7,100,000	11,450	15.9	62	34	2	1	1
52 hr.	36.0	34.3								
56 hr.*	35.3	34.8								
60 hr.	39.8	38.0								
72 hr.	36.5	36.5	6,750,000	13,250	13.7	58	36	1	2	3

* indicates another administration of the drug.

Table XXXIV — Determinations on Dog No. 114 — 8/23/48

Route - Oral (Tablet) — Male — 47½ lbs. — 3 yrs.

Dose - 1½ gr./lb. body weight — Times per day - three — No. days - three

History - Apparently healthy

Blood Studies

Time Interval	Sulfanerazine mg. % 1 cm. tubular		R.B.C.	W.B.C.	Hb. g./100 ml.	Differential N.	L.	M.	B.	E.
1 hr.	5.6	8.0	5,550,000	11,600	15.9					
4 hr.*	8.4	10.0	5,920,000	11,150	14.0					
8 hr.*	10.5	15.0	5,670,000	9,100	13.7					
12 hr.	17.3	19.5	5,900,000	7,700	13.7					
24 hr.*	22.4	22.0	6,090,000	6,900	12.6	69	25	5		1
28 hr.*	24.0	23.0								
32 hr.*	32.0	32.6								
36 hr.	36.5	36.9								
48 hr.*	36.2	36.3	5,400,000	6,700	11.3	50	46	4		1
52 hr.*	39.6	38.0								
56 hr.*	40.1	39.5								
60 hr.	42.2	42.0								
72 hr.	40.8	40.0	5,520,000	4,850	12.0	48	51	1		

* indicates another administration of the drug.

BLOOD STUDIES IN DOGS FOLLOWING THE ADMINISTRATION OF SULFAMERAZINE

Thesis for degree of Master of Science

Michigan State College

F. E. Eads

1949

An Abstract

Sulfamerazine was administered by various routes and dosages to determine (1) the blood plasma levels of sulfamerazine at different intervals following its administration. (2) the frequency of administration and the dosage necessary to maintain therapeutic blood plasma levels, and (3) the effect on the peripheral blood. Two hundred forty-six sulfamerazine trials were completed using 7 normal healthy dogs.

Following the single oral administration of ½, 1, and 1½ grains of sulfamerazine per pound of body weight the blood plasma concentration was found to be approximately the same when given in tablet form, in powder form or in an emulsion.

Sulfamerazine blood plasma levels following oral administration were found to be slightly higher and persisted for a longer period of time than those following the intravenous or subcutaneous routes of administration. A concentration of 5 mg. percent (therapeutic level) or higher was maintained for an average of 20 hours following a single oral administration of 1 grain (143 mg. per kilogram) per pound of body weight, and 22 hours following 1½ grains (214.5 mg. per kilogram) per pound. Therapeutic blood levels were

Abstract cont'd - F. E. Eads

not obtained for a sufficiently long period of time by a single oral dosage of ½ grain (71.5 mg. per kilogram) per pound of body weight in the dog.

Therapeutic blood levels of free sulfamerazine were maintained over a prolonged period when given either subcutaneously or intravenously only when 1 or 1½ grains per pound of body weight (1.1 and 1.6 ml. per pound respectively) were administered. These two methods of administration were more objectionable to the dog than the oral administration.

Sulfamerazine blood plasma levels of 5 mg. percent or higher were maintained with the administration of ½ grain (71.5 mg. per kilogram) per pound twice daily per os. Larger and/or more frequent doses produced an unnecessarily high blood concentration.

Studies of the blood picture indicate that the greatest variation in the red cell count obtained in any animal was ± 450,000; in the majority there was very little change. Increased dosages of sulfamerazine during a three-day study produced a leukopenia in the normal healthy dog. Intravenously the trend was towards a leukocytosis. Hemoglobinemia was produced when sulfamerazine was administered by mouth at the rate of 4½ grains (643.5 mg. per kilogram) per pound of body weight daily in three divided doses for a three day period. With the larger doses, the dogs showed a marked anorexia and depression which lasted several days after the administration of the drug.

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