investigation.

Comparable results were obtained in a child with severe sickle cell anemia. Upon the administration of 5 g of Fraction IV-7, the serum iron rose from 245γ/100 cc of serum to a maximum of 310 γ in approximately 4 hours. No latent iron-binding capacity appeared.

Whether Fraction IV-7 furnishes a factor required for hemoglobin synthesis and red cell formation could not be established with the amounts of this material employed in the present study and within the short observation periods. Table I, which provides figures for 24 hour periods of observation, reveals entirely insignificant deviations of the hemoglobin and hematocrit values from the pre-injection levels.

Conclusion. The administration of Fraction IV-7 of plasma to 4 patients with severe Mediterranean anemia resulted in a consistent rise in serum iron values in all, and in a transitory appearance of a latent capacity to further bind iron in 3 of the 4 subjects. These observations suggest the release of iron from the tissues to the blood when additional metal-combining globulin is supplied in this disease. Further studies are in progress to explore the clinical value of Fraction IV-7 both in Mediterranean anemia and exogenous hemochromatosis associated with multiple transfusions.

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Absorption and Excretion of Terramycin in Humans: Comparison with Aureomycin and Chloramphenicol.* (17872)

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Terramycin is a recently discovered antimicrobial compound which is produced by a species of Streptomyces(1). This drug has been found to be active against a number of bacterial species *in vitro* and to suppress experimental infections in mice(2). Clinical investigation of terramycin is now in progress (3). Studies in laboratory animals have indicated that terramycin, like aureomycin and chloramphenicol, is well absorbed following oral administration(4). It seemed desirable, therefore, to make a study of the absorption, distribution, and excretion of terramycin following its administration to humans and to compare the data with those obtained following the administration of aureomycin or chloramphenicol (Chloromycetin®). In addition, the serum concentrations of these drugs attained in patients under treatment with various dosage regimens have been determined.

Materials and methods. Preparations of drug. Terramycin‡ was supplied as the amphoteric base in 250 mg capsules and in 2.5 g vials for aqueous suspension and as the hydrochloride in 250 mg capsules and com-

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‡ Supplied by Charles Pfizer and Co., Brooklyn, N.Y.

TABLE I.
Serum Concentrations of Terramycin Base and Hydrochloride in 28 Subjects After a Single Oral or Intramuscular Dose.

	Subj.	Dose, mg/kg	Serum conc. in μg/cc ½	1	2	4	6	24 hr
Terramycin base, oral	1	50		2.5	5	10	10	2.5
	2			2.5	250	4	4	<2.5
	3			<2.5	<2.5	<2.5	2.5	<2.5
	4			62	200	250	250	<2.5
	5			<2.5	<2.5	2.5	2.5	<2.5
	6	25		<2.5	<2.5	<2.5	2.5	<2.5
	7			<2.5	2.5	5	5	<2.5
	8			<2.5	2.5	2.5	2.5	<2.5
	9			<2.5	<2.5	<2.5	2.5	<2.5
	10			<2.5	2.5	2.5	2.5	<2.5
Terramycin hydrochloride, oral	11	50		<2.5	6.2	12	6.2	<2.5
	12			6.2	6.2	12	12	<2.5
	13			6.6	6.6	6.6	4	<2.5
	14			6.6	4	4	2.5	<2.5
	15			12	13	13	13	12
	16			6.6	3.3	3.3	2.5	<2.5
	17				3.3	3.3	2.5	
	18				5	5	4	
	19	25		6.2	6.2	12	12	3.1
	20			<2.5	<2.5	<2.5	<2.5	<2.5
	21			<2.5	6.2	6.2	6.2	<2.5
	22			<2.5	<2.5	<2.5	2.5	<2.5
	23			<2.5	3.3	3.3	3.3	<2.5
Terramycin base, I.M.	24	10	16	5	2.5		<2.5	<2.5
	25	10	5	5	5		<2.5	<2.5
	26	5	2.5	2.5	2.5		2.5	<2.5
	27	5	5	5	5		<2.5	<2.5
	28	5	5	5	5		2.5	<2.5

N.B. 2.5 μg/cc = minimal measurable concentration.

pressed tablets. Aureomycin hydrochloride§ was available in 250 mg capsules and in 50 mg vials. Chloramphenicol‖ was provided in 250 mg capsules.

Collection of specimens. In the studies of absorption and excretion following oral administration, all subjects were fasted for 12 hours preceding the ingestion of a single large dose of drug. Blood specimens were drawn with sterile precautions at intervals after the drug administration. The urine was likewise collected at intervals during the 8 or 24 hour period following the ingestion of drug, all specimens were pooled, and aliquots were sterilized by Seitz filtration prior to assay. Specimens of serum and other body fluids were collected at random from patients receiving daily maintenance doses of these drugs. Simultaneous withdrawals of blood and cerebrospinal fluid were made from a number of patients who had been given a single large dose of drug several hours before lumbar puncture or from patients receiving maintenance doses. All serum, urine, spinal fluid, and other specimens were kept frozen until time of assay.

Assay technic. Microbiologic assay was carried out both with a 2-fold serial dilution technic and a modification thereof which provided fractional dilutions of the unknown. In the latter technic(5) after a preliminary 1:4 dilution of the unknown serum in broth,

§ Supplied by Lederle Laboratories Division, American Cyanamid Co., Pearl River, N.Y.

‖ Supplied as Chloromycetin® by Parke, Davis and Co., Detroit, Mich.

5. Tompsett, R., Shultz, S., and McDermott, W., *J. Bact.*, 1947, v53, 581.

subsequent dilutions were made in 25% pooled serum broth. When the broth inoculum of culture had been added, the final concentration of serum in each tube became 20%. The final dilutions of the unknown were 1:5, 1:6.6, 1:8, 1:10, etc. This method obviated the effect which varying concentrations of serum proteins may exert upon such an assay procedure. For non-protein fluids such as cerebrospinal fluid, a preliminary 1:4 dilution was not made. The final dilutions of these specimens were 8:10, 7:10, and 6:10 in the first 3 tubes of the series respectively. The test organisms used in the assay procedures were *Bacillus cereus*¶ and *Streptococcus hemolyticus*, C203Mv. The tests were read after 18 to 24 hours incubation at 37°C. Standard solutions of the drugs for which the unknown specimens were being assayed were diluted, inoculated, and incubated concurrently with each series of unknowns.

Results. Absorption and excretion. A single oral or intramuscular dose of terramycin base or hydrochloride was administered to 28 subjects. The largest single doses given were 4 g by mouth and 0.6 g intramuscularly. Intramuscular injections of the base were irritating, so that only a few determinations were made with this preparation by this route. The serum concentrations obtained in this study are presented in Table I. The maximum concentrations were attained usually within 2 hours after an oral dose and within 30 minutes after intramuscular injection. These were of the order of 12 to 16 μg per ml after a dose of 50 mg per kilo by mouth or 10 mg per kilo intramuscularly. Two elderly persons attained serum concentrations of 250 μg per ml after an oral dose of 2.5 g of terramycin base. These were the highest serum concentrations measured and were out of proportion to the other serum determinations. Although renal function was not investigated in these 2 subjects, it is possible there may have been impairment of drug excretion. The median serum concentrations after oral administration of terramycin base and hydrochloride are shown in

¶ This strain was obtained from Mr. A. C. Dornbush, Lederle Laboratories.

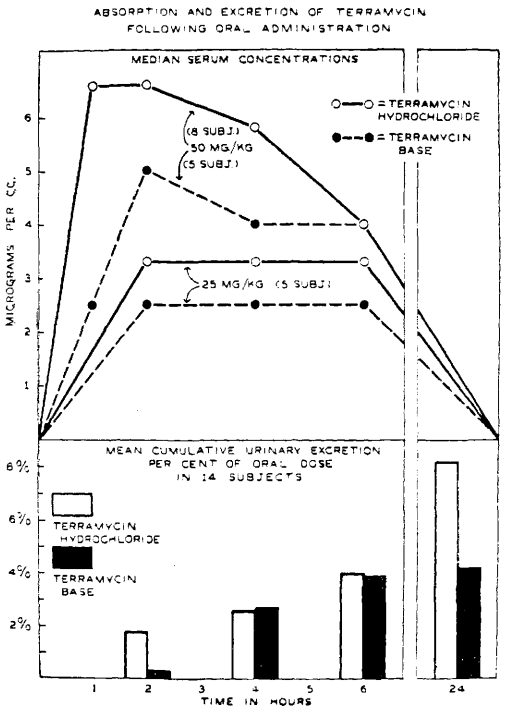

FIG. 1.

Fig. 1. The differences between the 2 preparations were not statistically significant ($p > 0.05$). Data for the urinary excretion of terramycin in 14 subjects are presented in Table II. The mean values for the amount of drug recovered in biologically active form in the urine after oral administration are shown in Fig. 1. These differences were likewise not significant ($p > 0.20$). Six adult males received a single oral dose of 50 mg per kilo of chloramphenicol; 3 days later the same 6 subjects were given an equal dose of aureomycin. The individual doses ranged from 3.25 to 4.25 g. The serum concentrations of these drugs are presented in Table III, and the median concentrations are shown in Fig. 2. Data concerning the urinary excretion are presented in Table IV. The mean cumulative excretion of the 2 drugs in biologically active form is shown in Fig. 2. The serum concentrations and the urinary excretion of chloramphenicol in biologically active form were consistently and significantly higher than those of aureomycin ($p < 0.01$).

Serum concentrations during maintenance

TABLE II.
Urinary Excretion of Terramycin Base and Hydrochloride After Oral and Intramuscular Administration in 14 Subjects.

	Subj.	Dose, mg/kg	Urine conc. μg/cc 0-2	2-4	4-6	6-24 hr	% dose recovered 2	4	6	24 hr
Terramycin, base oral	1	50	26	1790	102		.5	5.9	7.0	
	2	25	1.6	6.4	6.4	13	.4	3.2	5.4	6.5
	3	25	.2	51	51	102	.0	1.1	1.4	4.5
	4	25	6.4	13	13	6.4	.2	.8	1.2	1.8
Terramycin hydrochloride, oral	5	50	26	819	205	205	1.1	3.3	5.0	13
	6	50	26	103	103	205	.4	1.5	2.4	6.8
	7	25	2500	410	102	1250	4.7	7.8	10.6	26
	8	25	<.4	.8	13	51	.0	.0	.3	1.7
	9	25	26	410	410	625	.8	1.0	1.6	3.4
Terramycin, base intramuscular	10	10	22				.2			
	11	10	2.8		45*	11	.1		1.3	3.7
	12	5	.4		.8*	6.4	.0		.1	.4
	13	5	.4		<.4*	6.4	.0		.1	1.3
	14	5	410		26*	.8	10.8		11.3	11.6

* 2-6 hr grouped urine.

TABLE III.
Serum Concentrations of Aureomycin and Chloramphenicol in the Same 6 Subjects After a Single Oral Dose of 50 mg/kg.

Time after administration		Serum conc. in μg/cc Aureomycin	Chloramphenicol
15 min.	Sera	3	3
	Range	<1.5	<12.5–13.2
	Median	<1.5	<12.5
30 "	Sera	3	6
	Range	<1.5	<12.5–26.6
	Median	<1.5	18.3
1 hr	Sera	6	6
	Range	<1.5–2	<12.5–33
	Median	1.5	25.5
2 "	Sera	6	6
	Range	2–4	<12.5–40
	Median	3.3	33
4 "	Sera	3	3
	Range	3.3–8.3	33–50
	Median	6.2	50
6 "	Sera	3	3
	Range	2.5–12.5	25–33
	Median	5	25
8 "	Sera	6	6
	Range	2–6.2	16–26.6
	Median	3.2	22.5
24 "	Sera	3	3
	Range	<1.5	<12.5
	Median	<1.5	<12.5

N.B. 1.5 μg/cc = minimal measurable concentration of aureomycin.
12.5 μg/cc = minimal measurable concentration of chloramphenicol.

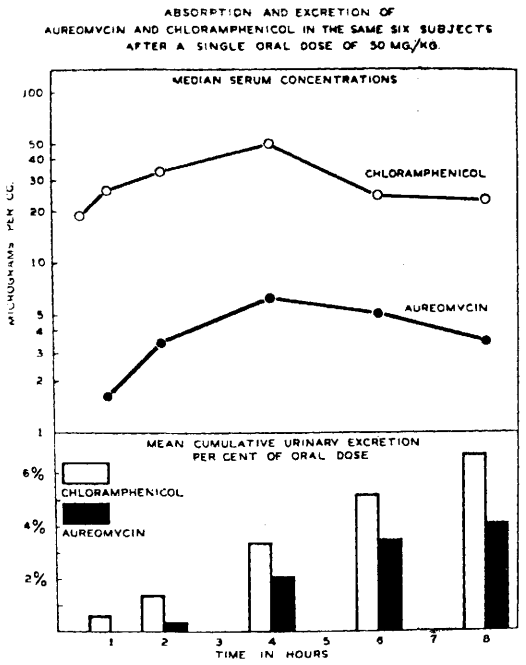

FIG. 2.

therapy. Twenty-two determinations of terramycin in the serum were made in 13 patients who were receiving terramycin hydrochloride in total daily doses of 5 g by mouth given in divided amounts at 6 hour intervals. On a weight basis this represented a daily intake of drug of between 50 and 100 mg per kilo. Most of these patients were suffering from a variety of infections and were febrile at the start of therapy. Measurable amounts of drug were quickly attained in the serum with these doses and ranged from 2.5 to 12.6 μg per ml. With maintenance doses of 4 g per day, the drug could frequently not be measured in the serum. The assay method was not capable, however, of accurately measuring concentrations of terramycin in the serum which were below 2.5 μg per ml. Thirty-five serum determinations of aureomycin were made in 12 patients receiving daily doses of 3 to 12 g of aureomycin hydrochloride given orally in divided amounts. These doses were equivalent to 50 to 200 mg per kilo per day. Several of these patients, especially those receiving the larger doses, were vomiting, however, and did not always retain the whole amount of drug given each day. Serum concentrations of 2.1 to 12.8 μg per ml were attained in patients receiving 200 mg per kilo per day, 3.2 to 10.2 μg per ml in those receiving 100 mg per kilo per day, and 1.5 to 5.1 μg per ml in those receiving 50 mg per kilo per day. Ninety-three determinations of chloramphenicol were made in the sera from 18 patients. The treatment regimens were the same as in the patients receiving maintenance doses of aureomycin. Observations were made on 4 dosage schedules. Maximum concentrations of 50 to 100 μg per ml were found in patients receiving 100 or 200 mg per kilo per day. With doses of 50 mg per kilo per day, amounts of drug in the sera ranged from 6.2 to 100 μg per ml, but most sera contained fewer than 25 μg per ml; with doses of 25 mg per kilo per day the concentration of drug was usually too low to measure, although a few patients attained serum concentrations as high as 12.5 μg per ml.

Concentrations in other body fluids. Small amounts of terramycin were found to be present in the cerebrospinal fluids of 4 patients after oral doses of terramycin hydrochloride. Two of these patients had been receiving maintenance doses of 4 g per day for several days before lumbar puncture. Although no drug was detectable in the sera at the time of lumbar puncture, concentrations of 0.5 and 0.7 μg per ml respectively were found in the cerebrospinal fluid. In 2 patients who had received only 3 g and 750 mg orally for a single day preceding lumbar puncture respectively, no drug could be measured in the cerebrospinal fluid. Three other patients received a single oral dose of 2.5 g 4 hours preceding lumbar puncture. In 2 of these terramycin was present in the cerebrospinal fluid in concentrations of 0.6 and 0.7 μg per ml and simultaneously in the serum in concentrations of 9.5 and 2.5 μg per ml respectively. In the third patient no drug was measured in the cerebrospinal fluid in spite of a serum concentration of 5 μg per ml. The assay procedure was capable of measuring as little as 0.5 μg per ml in the cerebrospinal fluid in contrast to not less than 2.5 μg per ml in the serum. Pleural fluid for terramycin assay was obtained from a patient 2 and 7 hours

TABLE IV.
Urinary Excretion of Aureomycin and Chloramphenicol in the Same 6 Subjects After a Single Oral Dose of 50 mg/kg.

Grouped urine, hr		Urine conc. in μg/cc		Hr after administ.	% of dose recovered in urine	
		Aureomycin	Chloramphenicol		Aureomycin	Chloramphenicol
0–½	No.	6	6	½	6	6
	Range	.8–.15	10–133		.0–.1	.0–.2
	Median	3.8	48		.0	.0
½–1	No.	6	6	1	6	6
	Range	16.5–63	13.3–1250		.0–.2	.0–1.1
	Median	48	175		.0	0.6
1–2	No.	6	6	2	6	6
	Range	50–380	20–1250		.2–.7	1–2.2
	Median	112	275		.3	1.2
2–4	No.	3	3	4	3	3
	Range	284–760	375–1875		1.6–2.4	2.9–3.7
	Median	568	625		2.4	3.7
4–6	No.	3	3	6	3	3
	Range	350–950	825–1875		2.3–4.4	4.7–5.6
	Median	570	1250		3.8	5.6
6–8	No.	3	3	8	6	6
	Range	175–1520	625–1250		1.7–8.4	3.9–9.3
	Median	570	825		3.1	7
23–24	No.	3	3			
	Range	33–100	<10–67			
	Median	33	20			
47–48	No.	3	3			
	Range	6.6–24	<10			
	Median	20	<10			

respectively following the ingestion of a single dose of 1.25 g of terramycin hydrochloride. Terramycin was present in concentration of 512 μg per ml in the 2 hour specimen but could not be detected in the 7 hour specimen. A concentration of 2.5 μg per ml of terramycin was obtained in the ascitic fluid of a patient who had been receiving 3 g by mouth daily for 7 days prior to paracentesis.

Aureomycin could not be measured in the cerebrospinal fluids of 4 patients who had been receiving daily oral doses of 4 g for several days prior to lumbar puncture. An additional patient was given a single dose of 3 g by mouth. Four hours later blood and cerebrospinal fluid specimens were obtained simultaneously for assay. Although the serum concentration of aureomycin was 7.6 μg per ml, no drug was measured in the cerebrospinal fluid. The assay technic was capable of detecting as little as 0.4 μg per ml of aureomycin in the cerebrospinal fluid. One patient with hydrarthrosis of the knee received 4.5 g of aureomycin hydrochloride daily for 3 days. At the end of this time there was present in the joint fluid a concentration of 0.3 μg per ml of the drug. Other observers have reported the accumulation of aureomycin in the cerebrospinal fluid and in other fluids of the body in patients taking the drug by mouth(6).

Nineteen determinations of chloramphenicol were made on cerebrospinal fluids obtained from 7 patients who were receiving daily doses of 25 to 50 mg per kilo of this drug. These patients had concentrations of chloramphenicol in the serum which ranged from less than 6.2 μg to 100 μg per ml. The cerebrospinal fluid concentrations ranged from less than 4 μg per ml, which was the smallest amount detectable by the assay technic, to

6. Dowling, H. F., Lepper, M. H., Caldwell, E. R., Jr., Whelton, R. L., and Brickhouse, R. L., *J. Clin. Invest.*, 1949, v28, 983.

50 μg per ml. All patients receiving the larger doses of chloramphenicol had measurable concentrations of drug in the cerebrospinal fluid after 2 or 3 days of treatment, and these were of the order of one-fourth to one times the serum concentrations. Chloramphenicol was also found in concentration of 12.5 μg per ml in the joint fluid of one patient who had a coexisting serum concentration of 25 μg per ml.

Discussion. It is well recognized that studies of the concentrations of an antimicrobial drug which are attained in the blood may bear only a very loose relationship to the therapeutic results which will be achieved with the administration of that drug. Nevertheless, the results of such studies can be valuable in serving as a guide to the dosage regimens to be employed in therapy and in comparing the absorption and excretion of various forms of the same drug. Of the 3 compounds studied, chloramphenicol provided the most rapid accumulation of antimicrobial substance in the serum and yielded the highest serum concentrations. It also accumulated in the cerebrospinal fluid to the greatest extent. Terramycin was found to compare favorably with chloramphenicol and aureomycin in its absorption following oral administration and in its excretion in the urine. Concentrations of terramycin in the serum were obtained following single large doses or with daily maintenance doses of terramycin hydrochloride which were similar in magnitude to those concentrations attained with equivalent doses of aureomycin given by mouth. The persistence of measurable serum concentrations of terramycin for periods of 6 hours and longer after single doses and the high serum concentrations attained when 1 to 1.25 g maintenance doses of drug were given at 6 hour intervals indicate that total daily doses of 4 to 5 g by mouth will provide satisfactory drug concentrations in the body in terms of the *in vitro* sensitivities of the various microorganisms against which terramycin has been demonstrated to be effective(2).

Summary. A study has been made of the absorption, distribution, and urinary excretion of terramycin, aureomycin, and chloramphenicol in humans. The determinations of absorption and excretion of aureomycin and chloramphenicol following a single oral dose of drug were made in the same subjects. All 3 drugs were readily absorbed after oral administration. The maximum serum concentrations attained after oral doses of 50 mg per kilo were 25 to 50 μg per ml for chloramphenicol, 12 to 16 μg per ml for terramycin, and 3.3 to 12.5 μg per ml for aureomycin. Reasonably high serum concentrations of all 3 drugs were maintained with daily doses of 50 to 100 mg per kilo by mouth. Measurable concentrations of the compounds were obtained in the cerebrospinal fluid and in other body fluids when sufficiently large doses of the drugs had been administered. Urinary excretion of the 3 substances in biologically active form was similar, and high concentrations of drug were attained in the urine.

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