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Comparison of Synthetic and Fermentation Chloramphenicol (Chloromycetin) in Rickettsial and Viral Infections.

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Synthetic chloromycetin, recently prepared by Crooks and his coworkers,[1] has been shown to have the same physical properties and antibacterial activities as chloromycetin produced by the mold *Streptomyces venezuelae*, N. Sp. The present report provides comparative data on the rickettsiostatic and virustatic effects of crystalline chloromycetin obtained from two sources, *i.e.*, from fermentation liquor of cultures of *Streptomyces venezuelae*, N. Sp.[2] and from chemical synthesis.[1] Both types were supplied by the Research Laboratories of Parke, Davis and Company. Earlier publications have provided information on the use of the fermentation type of drug in the treatment of rickettsial and viral infections of experimental animals[3-7] and man.[8-11]

1 Controulis, J., Rebstock, M. C., and Crooks, H. M., Jr., *J.A.C.S.*, 1949, in press.

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TABLE I.
Comparison of Effect of Two Types of Chloromycetin in Infected Embryonated Eggs.

Infecting agent	Drug dose mg/egg	Mean prolongation of life in days of embryos treated with chloromycetin	
		Fermentation	Synthetic
R. rickettsi	.125	1.65	1.78
	.25	2.91	2.91
R. akari	.125	1.89	2.06
	.25	3.67	3.48
R. mooseri	.125	1.23	1.09
	.25	2.26	2.63
R. prowazcki	.125	1.20	1.00
	.25	2.82	2.82
R. tsutsugamushi (Gilliam strain)	.125	1.02	1.07
	.25	3.20	3.09
(Seerangayee strain)	.25	2.30	2.40
Psittacosis virus (6 BC strain)	.25	1.63	1.83
	.50	3.33	3.63
(P 4 strain)	.25	1.29	1.45
	.50	3.20	2.96
Lymphogranuloma virus (J.H. strain)	.25	3.93	3.89
	.50	7.12	7.23

Methods. The methods employed in the present work were similar to those used in previous studies [3,6-8,10] and need not be discussed.

Results. Table I summarizes the data obtained in embryonated eggs treated with varying doses of the 2 types of chloromycetin one-half hour before inoculation with a given rickettsial or viral strain. In this work the causal agents of epidemic, murine and scrub typhus, spotted fever and rickettsialpox, and of psittacosis and lymphogranuloma venereum were employed. Inspection of Table I reveals that the mean prolongations of life (MPL) of the groups infected with a given agent and treated with the same amounts of fermentation or synthetic drug were essentially identical. Statistical analysis of the basic data confirmed this. In these tests the MPL represented the difference between the mean day of death of infected embryos in treated and control groups. Each group contained 20-24 living embryos at the beginning of the period for calculating the mean day of death; depending on the infectious agent, deaths within the first 3 or 4 days after inoculation were considered non-specific.

Mice infected with the Karp strain of *R. tsutsugamushi* received a single daily intraperitoneal injection of solutions containing varying amounts of the 2 types of drug. The results obtained in one such test in which treatment was begun the day following injection of rickettsiae and continued for 20 days are summarized in Table II. Both types of drug were highly effective in preventing death of the animals.

In other experiments mice were infected intraperitoneally with the 6 BC strain of psittacosis virus and treated by intraperitoneal injection or oral administration of varying doses of the 2 types of chloromycetin. The results were essentially indistinguishable from those previously obtained[7] when the fermentation

[9] Payne, E. H., Knaudt, J. A., and Palacios, S., *J. Trop. Med. and Hyg.*, 1948, **51**, 68.

[10] Smadel, J. E., Woodward, T. E., Ley, H. L., Jr., Philip, C. B., Traub, R., Lewthwaite, R., and Savoor, S. R., *Science*, 1948, **108**, 160.

[11] Pincoffs, M. C., Guy, E. G., Lister, L. M., Woodward, T. E., and Smadel, J. E., *Ann. Int. Med.*, 1948, **29**, 656.

TABLE II.
Comparison of Chemotherapeutic Effect of Two Types of Chloromycetin in Mice Infected with *R. tsutsugamushi.*

Drug treatment I. P.		Dilution of infectious inoculum		
Type	mg/day/mouse	10^{-5}	10^{-6}	10^{-7}
None	(Controls)	8/8*	4/8	4/8
Fermentation	2.5	0/8		
	1.5	0/8		
	0.75	0/8		
	0.375	2/8		
Synthetic	2.5	0/8		
	1.5	0/8		
	0.75	1/8		
	0.375	3/8		

* Numerator = No. of mice dying; denominator = No. of mice in group.

All mice in control groups receiving 10^{-2}, 10^{-3}, 10^{-4} dilutions of challenge material succumbed while one died in the 10^{-8} group. The titer of the inoculum was $10^{-6.6}$, therefore, the drug treated mice received approximately 40 MLD's intraperitoneally.

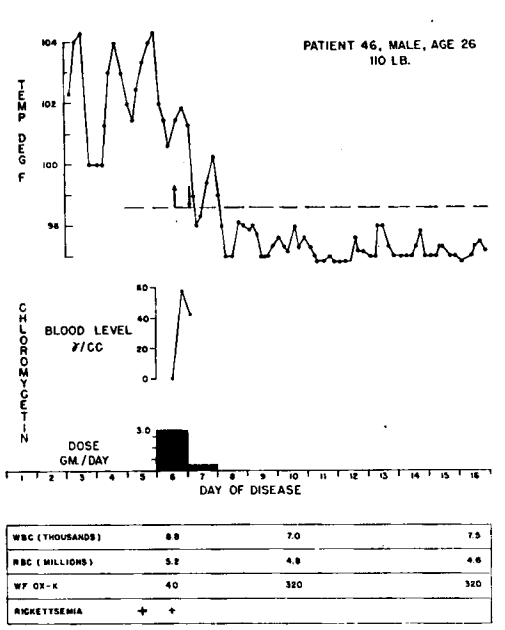

FIG. 1.

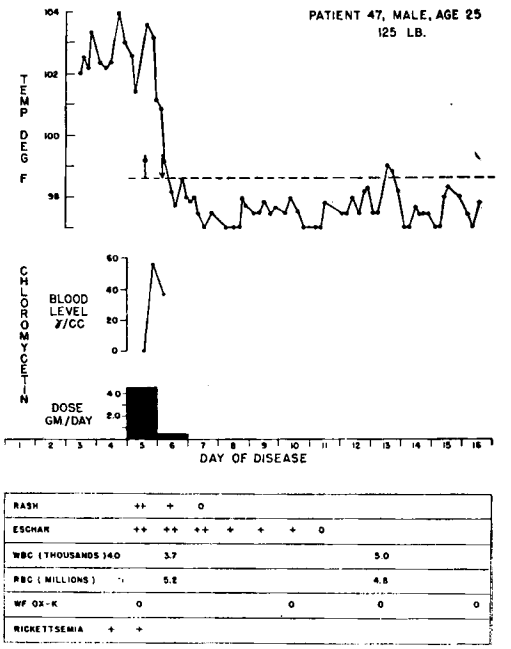

FIG. 2.

type of antibiotic was employed under similar conditions. Chloromycetin from both sources was equally virustatic in the present tests.

The clinical findings in 2 patients with scrub typhus* who were treated with synthetic chloromycetin are summarized in Fig. 1 and 2. The diagnosis was established in each individual by recovering *R. tsutsugamushi* on 2 occasions from samples of blood which were drawn during the febrile phase and promptly injected intraperitoneally into mice. Furthermore, during convalescence Patient 46 developed agglutinins for the OX-K strain of *B. proteus*. Both patients were con-

* The authors wish to thank Major C. J. Williams, RAMC, and other members of the Staff of the Military Hospital, Kuala Lumpur, who were responsible for the care of these patients.

sidered to have had typical scrub typhus although neither presented all the classical signs of the disease. Thus, Patient 46 failed to show a rash or eschar while Patient 47 failed to develop a positive Weil-Felix reaction. Both persons received the drug orally over a period of 16 hours. Patient 46 was given an initial dose of 3.0 g followed by 4 doses of 0.25 g at 3-hour intervals. The regime for Patient 47 was similar except that the initial amount was 4.0 g. The response of these 2 patients following therapy was entirely comparable to that observed in other cases of scrub typhus treated with fermentation chloromycetin. It will be recalled that the febrile period of scrub typhus, which usually lasts for 14 days in untreated cases, was terminated in 31 hours (average for 25 patients) after treatment with chloromycetin was instituted.[10]

Our experience, although limited, indicates that synthetic chloromycetin possesses the same low level of toxicity for embryonated eggs, mice and men that has characterized the fermentation type[3,5,10] of drug.

Conclusion. Chloromycetin prepared by chemical synthesis appears to possess the same rickettsiostatic and virustatic properties in experimental infections and the same usefulness in treating patients with scrub typhus that have been demonstrated for chloromycetin produced by *Streptomyces venezuelae,* N. Sp.

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Condensation of 2, 3-Dimercaptopropanol (BAL) with Oxophenarsine Hydrochloride: Toxicity and Chemotherapeutic Effect.

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(Introduced by E. K. Marshall, Jr.)

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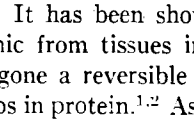
The use of 2,3-dimercaptopropanol (BAL) in the treatment of arsenical poisoning has been well established, and the pharmacological and biochemical basis for its action has been thoroughly explored. It has been shown that BAL mobilizes arsenic from tissues in which the metal has undergone a reversible binding with sulfhydryl groups in protein.[1,2] As a result BAL increases the excretion of administered arsenic from the body[3,4] and is a useful antidote in poisoning from arsenical gases or complications following administration of oxophenarsine hydrochloride.[2-7]

It has been shown that when arsenic reacts with keratin the ratio of combination with sulfur of the protein is 1 As : 2 S, and this suggested that a relatively stable ring of the type



is formed.[1] It is therefore postulated that BAL detoxifies by the removal of arsenic from the protein system and incorporation *in vivo* into a more stable cyclic

* Eli Lilly Fellow in Pharmacology and Experimental Therapeutics.

1 Stocken, L. A., and Thompson, R. H. S., *Biochem. J.*, 1946, **40**, 529.

2 *ibid.*, p. 535.

3 *ibid.*, p. 548.

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