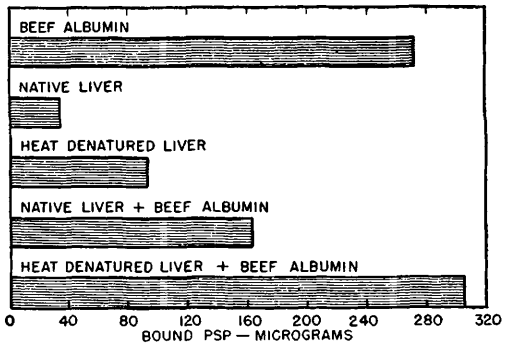

FIG. 1.

Effect of adding native, and heat denatured, homogenized tissue on the binding of phenolsulfonephthalein by crystallized bovine plasma albumin, 50 mg.

tissue proteins with the albumin molecule thus decreasing the available points of attachment for PSP in the system while heat denaturation of the tissue proteins decreased their attraction for albumin increasing the amount of dye anions bound by the mixture.

Summary. 1. Tissue proteins are capable of binding the anionic dye, phenolsulfonephthalein, in small amounts. All of the tissues examined bound less of the dye than did serum and different tissues appear to have different capabilities in this respect. Of the tissues tested liver bound most and pancreas least.

2. Denaturation of tissue proteins by heat or urea materially increased the ability of the tissue protein to bind the dye.

3. Native tissue proteins of the rat considerably decreased the binding of phenolsulfonephthalein by crystallized bovine albumin; this property was abolished by heat denaturation of the tissue proteins.

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Effect of Aureomycin on the Herpes Simplex Virus in Embryonated Eggs.* (17498)

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Aureomycin was shown by Wong and Cox to have marked therapeutic activity against the viruses of the psittacosis-lymphogranuloma group.(1) They demonstrated that 49 of 55 treated chick embryos survived experimental infection with psittacosis through the 13th day, contrasted with survival of only 3 of 57 control embryos. These results were found to be essentially true for the other members of the psittacosis-lymphogranuloma group (lymphogranuloma venereum, psittacosis, SF strain human pneumonitis, mouse pneumonitis, feline pneumonitis, and meningopneumonitis F97 strain) as well as for the rickettsiae. In contrast with the marked activity *in vivo,* they conclude that the drug had no *in vitro* effect against these agents. Harned *et al.,* in a study on the acute toxicity of intravenously administered aureomycin hydrochloride for several laboratory animals, found the LD50 for the drug to be 134 mg/kg for mice and 118 mg/kg for rats.(2) Clinical accounts of successful treatment of

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The aureomycin used in this study was supplied by Lederle Laboratories Division of American Cyanamid Company, New York City.

Much of the preliminary work incident to this study was done by Dr. Lewis L. Coriell.

The activity levels for inactivated aureomycin were performed by Dr. Coleman Whitlock.

1. Wong, S. C., and Cox, H. R., *Ann. N. Y. Academy Sci.,* 1948, v51, 290.

2. Harned, B. K., Cunningham, R. W., Clark, M. C., Cosgrove, R., Hine, C. H., McCauley, W. J., Stokey, E., Vessey, R. E., Yuda, N. N., and SubbaRow, Y., *Ann. N. Y. Acad. Sci.,* 1948, v51, 182.

diseases thought to be caused by herpes simplex such as Kaposi's Varicelliform Eruption,(3) dendritic ulcers of the cornea(4) and herpetic stomatitis(5) are being reported in increasing numbers. Because of the effectiveness of aureomycin against the larger viruses and the optimistic clinical reports against herpes simplex infections it seemed important to determine the *in vitro* effects of this antibiotic on the herpes simplex virus and to evaluate its *in vivo* action under the most favorable possible conditions in embryonated chick eggs.

Method and Materials. The toxicity of aureomycin was determined in chick embryos 10 to 12 days of age which averaged 50 g in weight. The eggs were prepared by the false air sac technique,(6) and the individual aureomycin dose dissolved in 0.2 ml of gelatin saline solution, was inoculated onto the "dropped" chorioallantois. The eggs were incubated at 96°F and 76% relative humidity. They were candled at least twice daily and death of the embryo was used as the end point. All dead eggs were cultured in Brewer thioglycollate medium for possible bacterial contamination. For drug controls penicillin in a concentration of 500 units/ml was used in some experiments and inactivated aureomycin in others. The aureomycin control was prepared by heating aureomycin at 56°C for 5 days, and was shown by bioassay to contain less than 0.1% of its original activity. Two lots of aureomycin were used. The first series of experiments was performed with batch A (Lederle lot No. unknown) and the later series with batch B (Lederle lot No. 7-9022B).

The virus used was egg adapted in this laboratory from the HF Rockefeller strain by 129 serial passages on the chorioallantoic membrane. The inoculum for each experiment was prepared from virus maintained in this laboratory by frequent passage. Different passages, each of which had a titer of approximately 10^9 infectious units per ml were used for each experiment. This strain of virus when properly diluted kills 95-100% of the chick embryos between the 69th and 90th hours. Embryos which died with virus plus drug were subcultured in other eggs to establish the presence of virus.

The *in vitro* effect was determined by mixing the drug and 10% suspensions of the infected membranes; incubating at room temperature for 4 hours; diluting the mixture sufficiently to obviate any *in vivo* effect; and injecting the inoculum onto the chorioallantoic membrane. Penicillin (500 units/ml) which had previously been shown to have no virucidal action was used as control.(7) To determine an *in vivo* effect one experiment was performed in which the virus and drug were inoculated into the yolk sac, the virus follow-

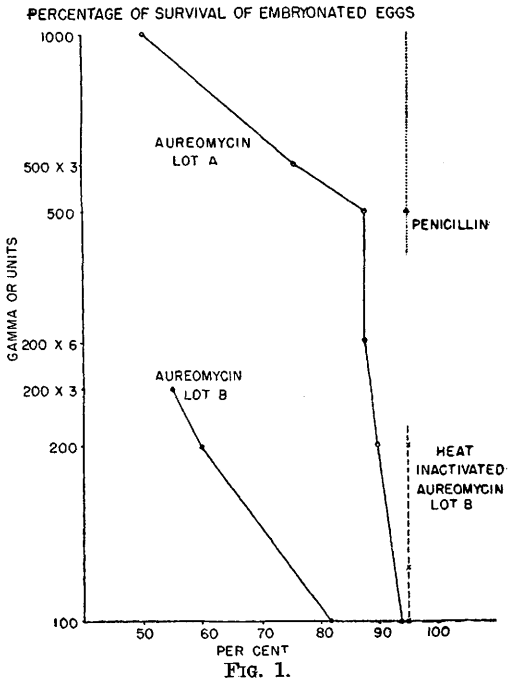

FIG. 1.

The toxicity of aureomycin and penicillin for embryonated eggs. The drug was inoculated onto the chorioallantoic membrane and death within 60 hours was taken as end point. Most points are the results from 24 or more eggs.

3. Baer, R. L., and Miller, O. B., *J. Invest. Derm.*, 1949, v13, 5.

4. Braley, A. E., and Sanders, M., *Ann. N. Y. Acad. Sci.*, 1948, v51, 280.

5. Fisher, A. A., and Schwartz, S., *J. Invest. Derm.*, 1949, v13, 51.

6. Scott, T. F. McNair, American Public Health Association, *Diagnostic Procedures for Virus and Rickettsial Diseases*, p. 243, 1948.

7. Coriell, L. L., Blank, H., and Scott, T. F. M., *J. Lab. Clin. Med.*, 1949, v34, 402.

TABLE I.
In vitro Effect of Aureomycin on Herpes Simplex Virus.

No. of infectious units inoculated into each egg	Conc. of aureomycin *in vitro* before dilution and inoculation	Amt. of aureomycin inoculated into each egg	Mortality of eggs inoculated with aureomycin-treated virus	Mortality of eggs inoculated with control (penicillin)-treated virus
5×10^5	4000 γ/ml	2 γ	12/12	12/12
5×10^4	4000 γ/ml	0.2 γ	10/10	12/12
5×10^3	4000 γ/ml	0.02 γ	12/12	12/12

ing the drug by 30 minutes. To detect minimal effects and insure maximal contact of drug with virus, additional experiments were done on the chorioallantoic membrane with the technique of inoculation as described in the toxicity studies. In one experiment the virus preceded the drug but in the others the drug was inoculated 30 minutes before the virus. In addition, in several experiments the drug was reinoculated at 12-hour intervals for a total of 3 or 6 doses.

Results. The toxicity of aureomycin for embryonated eggs is indicated in Fig. 1. The two batches of aureomycin, which were obtained at different times showed an appreciable difference in toxicity. Because of this, experiments to determine the effect of the larger doses on the virus could not be repeated with

TABLE II.
Mortality of Embryonated Eggs Inoculated on the Chorioallantois with Drugs and Then with Herpes Simplex Virus.

Approx. No. of infectious units inoculated	Aureomycin batch A						Aureomycin batch B (No. 7-9022 B)					
	100 γ/egg		200 γ/egg		500 γ × 3/egg		100 γ/egg		200 γ/egg		200 γ × 3/egg	
	Aureomycin	Control	Aureomycin	Control	Aureomycin	Control	Aureomycin	Control	Aureomycin	Control	Aureomycin	Control
5×10^5	20/21	22/22	17/23	27/27								
5×10^4	19/21	21/22	18/26	25/27	4/10	13/13	8/8	9/9	7/7	10/10	14/14	18/18
5×10^3	16/22	15/22	16/25	20/26	5/13	12/12	7/8	10/10	8/9	10/10	7/12	14/18
5×10^2					1/9	4/12	6/8	10/10	2/3	10/10	4/7	10/17
5×10^1							4/9	6/10	2/5	5/10		
5×10^0							3/9	2/10	1/10	2/10		

the second batch of aureomycin. Under the experimental conditions, aureomycin inactivated by heat, and penicillin were non-toxic. The LD50 of the least toxic lot of aureomycin for the chick embryo was 20 mg/kg. The *in vitro* effect of aureomycin on the herpes simplex virus is shown in Table I. The drug apparently has no *in vitro* action on the virus, under the conditions of the present experiments. The *in vivo* effect of aureomycin on the herpes simplex virus is shown in Table II. Because of the difference in toxicity and a possible difference in activity in the two lots of aureomycin, the results with each lot are recorded separately. There seems to be little difference in the effectiveness of the two lots of drug. Although in some of the dilutions small differences occurred between the controls and the drug treated embryos, at the critical dilutions with moderately large doses of drug and small amounts of virus there is no significant increased survival of aureomycin treated eggs. Analysis of the data also failed to show any significant delay in time of death of treated infected eggs.

Conclusions. (1) Aureomycin appears to be more toxic for the chick embryo than for most other laboratory animals. (2) There seemed to be a significant difference in toxicity between the 2 lots of aureomycin used. Incubation of aureomycin at 56°C for five days destroys the embryo-toxic principle as well as its antibiotic activity. (3) Aureomycin has no *in vitro* activity against herpes simplex virus. (4) As compared with its activity against the larger viruses, no similar *in vivo* effect against herpes simplex virus could be demonstrated.

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Remission of Disseminated Lupus Erythematosus Induced by Adrenocorticotropin.* (17499)

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This report describes the effect of pituitary adrenocorticotropic hormone (ACTH) in inducing 3 temporary dramatic remissions in 2 patients with classic disseminated lupus erythematosus. There was a virtually complete clinical remission while the ACTH was being administered. A description of this phenomenon has not been previously published.

Case I: A.F., a 36-year-old white housewife was admitted complaining of fever, weakness, weight loss, and arthralgia of 9 months' duration. She appeared acutely ill, with a daily temperature of 101°F. There were erythematous, scaly plaques on both cheeks which also showed telangiectatic vessels and areas of atrophy. The eruption, in addition, involved the forehead, chin, ears, scalp, arms and legs. Scalp and pubic hair were sparse. There was a soft, blowing, systolic murmur over the 4th left intercostal space. The spleen was not palpable. Examination of the blood showed anemia, leukopenia, thrombocytopenia and an elevation of the sedimentation rate; protein and microscopic collections of red blood cells were constantly present in the urine. A section of the involved skin of the face showed changes usually seen in disseminated lupus erythematosus.

Beginning May 24, 1949, 150 mg of ACTH was injected intramuscularly in 6 divided doses daily for 5 days when they had to be stopped because further supplies of this scarce hormone were unavailable.

A remarkable change in the patient's condition was apparent within 12 hours after the injections were started. She appeared more

* The assistance and cooperation of Dr. Dale Doherty and Dr. S. Biloon in the clinical study of these cases is appreciated.