frequent accompaniment of chemical or heavy metal toxicity, with or without evidence of hepatic functional impairment.[8,9] The failure to observe excesses in this experiment is in better agreement with the belief that the acute massive hepatic necrosis is not due to a chemical or poison. The histologic appearance of these livers suggests that the lesion develops very rapidly since there appears to be little evidence of antecedent damage as judged by infiltration or any attempt at a reparative process.

Summary. 1. The urinary and fecal excretion of coproporphyrin was studied at varying intervals in 21 young rats maintained on a diet containing 18% yeast as a source of protein.

2. Fourteen of the rats died suddenly between the 30th and the 125th day of acute massive hepatic necrosis. The gross and microscopic findings were identical with those described by Himsworth as characteristic of this lesion.

3. No significant increase in the excretion of coproporphyrin was observed during the course of these experiments. This is believed to minimize the likelihood of chemical or metal toxicity as the cause of the necrosis.

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Comparative Effect of Aureomycin and Chloromycetin on Psittacosis Infection in Chick Embryos.* (17433)

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Aureomycin[1] and chloromycetin[2,3] are antibiotics which have a similar spectrum of activity that includes many bacteria, rickettsias and viruses of the psittacosis-lymphogranuloma venereum group. With respect to psittacosis, Wong and Cox[4] found aureomycin to be highly effective against infection of embryonated hen's eggs with the 6BC strain, and Smadel and Jackson,[5] using the same strain, demonstrated similar beneficial effects from chloromycetin. The conditions of the experiments of these 2 groups of workers, however, were not entirely comparable. It seemed of interest, therefore, to make a direct comparison of the effects of these 2 agents under more nearly identical conditions. Such a comparison is reported in this paper.

Materials and methods. A uniform source of virus for the present experiments was obtained in the following manner: A stock yolk-sac suspension of the 6BC strain of psittacosis virus was injected into the yolk sac of a number of 7-day chick embryos which were then incubated at 35°C. On the 5th day, when deaths began to occur, the yolk sacs were harvested and pooled. An equal volume of broth was added and the mixture homogenized in a Waring blendor. The resulting 50% yolk sac suspension which, on culture, was found to be free of bacterial contamination, was distributed in sealed pyrex ampoules, quick-frozen and stored at −70°C. The LD_{50} of this suspension was about 10^{-8} throughout these experiments. Yolk sac smears of infected embryos stained by Macchiavello's technic showed numerous characteristic elementary bodies.

A fresh solution of crystalline aureomycin

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1 Duggar, B. M., and others, *Ann. N. Y. Acad. Sci.*, 1948, **51**, 175.

2 Gottlieb, D., Bhattacharyya, P. K., Anderson, H. W., and Carter, H. E., *J. Bact.*, 1948, **55**, 409.

3 Smith, R. M., *et al.*, *J. Bact.*, 1948, **55**, 425.

4 Wong, S. C., and Cox, H. R., *Ann. N. Y. Acad. Sci.*, 1948, **51**, 290.

5 Smadel, J. E., and Jackson, E. B., *Science*, 1947, **106**, 418.

hydrochloride‡ was prepared just prior to use in each experiment. A solution containing 8 mg of crystalline chloroamphenicol§ (chloromycetin) per ml was made, also in broth, but this was heated to 100°C for 15 minutes and and then stored at 4°C for use in 2 or 3 experiments. The chloromycetin remained in solution throughout this study and retained its potency when assayed against a standard test organism. Both antibiotics were diluted for use so that the desired dose was contained in 0.25 ml of broth.

All inoculations were made into the yolk sac of 7-day-old embryonated eggs. In each experiment, groups of 6 to 12 eggs per dose were each injected with aureomycin in amounts ranging from 1.0 mg to 0.0625 mg and comparable groups were inoculated with similar amounts of chloromycetin. These eggs were then inoculated with a 10^{-3} dilution of the virus suspension. A titration of the virus was included in each experiment and this inoculum was regularly found to contain about $10^5 LD_{50}$. The time elapsed between injections of drug and virus was about 45 minutes. Uninfected drug controls and untreated virus-infected controls were each given a second injection of plain broth. The eggs were incubated at 35°C; they were candled and the deaths noted daily for 12 days, after which the experiment was terminated. Deaths occurring before the third day were considered traumatic. The dose of drugs that protected 50% of embryos (PD_{50}), and the LD_{50} were calculated by the method of Reed and Muench.[6] This method was also used to calculate the day when 50% of the embryos died (DD_{50}).‖

Results. Similar results were obtained in 6 separate experiments. The data for all 6 experiments have been combined in Table I.

TABLE I. Effect of Aureomycin and Chloromycetin on Psittacosis (6BC) Infection of Chick Embryos. Summation of 6 experiments.

Antibiotic	Dose, mg	No. of embryos infected*	No. of embryos surviving: Days after infection 3	4	5	6	7	8	9	10	11	12	DD_{50}†	MPL_{50}‡ days	% survivals: Days after infection 8	10	12
Aureomycin	1.0	22	22	21	21	20	19	19	15	13	12	10	11.8	7.2	86	59	45
"	.5	54	54	53	52	50	47	38	37	30	27	21	11.6	7.0	70	56	39
"	.25	56	56	55	54	54	51	37	24	15	9	6	9.7	5.1	66	27	11
"	.125	57	57	56	55	46	41	22	10	5	0		8.1	3.5	39	9	0
"	.0625	35	35	35	33	23	15	5	1	0			7.0	2.4	14	0	0
Chloromycetin	1.0	43	43	42	39	36	34	24	19	11	6	3	9.5	4.9	56	26	7
"	.5	50	50	50	49	48	37	26	10	3	0		8.3	3.7	52	6	0
"	.25	53	53	52	47	37	17	3	1	0			6.7	2.1	6	0	0
"	.125	52	52	52	44	21	2	0					5.9	1.3	0	0	0
"	.0625	12	12	11	7	0							5.1	0.5	0	0	0
Broth		57	57	51	10	0							4.6		0	0	0

Infecting dose per embryo = 85,000 LD_{50}, calculated on 8th day. (See also Table II).
* Number which survived more than 2 days.
† Day when 50% of embryos died (calculated).
‡ Mean prolongation of life. (DD_{50} for each dose minus DD_{50} for unprotected controls).

‡ Supplied in sterile vials by the Lederle Laboratories.

§ Furnished by the Department of Clinical Investigation, Parke, Davis and Co.

[6] Reed, L. J., and Muench, H., *Am. J. Hyg.*, 1938, **27**, 493.

‖ This method was chosen in preference to the usual calculation for "mean day of death" in order to avoid the error introduced by the embryos which survived at the end of the experiment.

TABLE II.
Protective Dose.

Days after infection	PD_{50} (mg)		LD_{50} of infecting dose*
	Aureomycin	Chloromycetin	
7	.09	0.36	1.8×10^4
8	.20	0.61	8.5×10^4
9	.31	>1	3.1×10^5
10	.42	>1	6.2×10^5
11	.51	>1	9.6×10^5
12	.62	>1	$>10^6$

* The number of LD_{50} contained in the infecting dose will vary according to the time of observation; and the figures in this column indicate the apparent number of LD_{50} on the days indicated in the first column.

The infected and unprotected control embryos all died between the 4th and 6th days. The DD_{50} for this group was 4.6 days. In the treated groups, the DD_{50} varied directly with the dose.

The beneficial effects of the 2 antibiotics may be compared on the basis of the mean prolongation of life (MPL) which, for convenience may be considered as the difference between the DD_{50} of any treated group and that of the control group. The MPL for the aureomycin-treated eggs was greater than that of the chloromycetin-treated groups at each dosage level. The MPL for the groups of embryos receiving doses of 0.5 and 0.25 mg of aureomycin was 3.3 and 3.0 days longer, respectively, than for those receiving the corresponding doses of chloromycetin. At each of the other dosage levels the MPL was about 2 days longer in the aureomycin treated groups.

The greater protective effect of aureomycin as compared with chloromycetin is also strikingly apparent from the percent of survivors among the groups of embryos treated with various doses of antibiotics. This was calculated for each dose at different times after infection and the figures for the 8th, 10th and 12th days are shown in Table I.

A more quantitative estimation of the relative effectiveness of the 2 drugs may be arrived at by comparing the dose required to protect 50% of the embryos (PD_{50}) for any given number of days after infection. The result is shown in Table II for days 7 to 12, inclusive. In the case of aureomycin, it is seen that the PD_{50} increased at a steady rate of approximately 0.1 mg per day during this period. The increase for chloromycetin was at a much more rapid rate between the 7th and 9th days, after which time the PD_{50} of this agent exceeded 1.0 mg, the largest dose used in these experiments. The PD_{50} of aureomycin on days 7, 8 and 9 was 0.09, 0.20 and 0.31 mg, respectively, as compared with 0.36, 0.61 and more than 1.0 mg for chloromycetin on the corresponding days. Thus on these 3 days, aureomycin was from 3 to 4 times as active as chloromycetin, weight for weight, in protecting the embryos against the infecting dose of virus used. A similar advantage in favor of aureomycin was noted after the 9th day, but this was not quantitated since the PD_{50} for chloromycetin was greater than 1.0 mg, the largest dose used. [The infecting dose on the basis of the LD_{50} calculated for each of these days, is also shown in Table II.]

Discussion. The results indicate that the life of chick embryos infected with the 6BC strain of psittacosis virus was prolonged for an average of from 2 to more than 3 days longer by aureomycin than they were by the same dose of chloromycetin. Also, the dose of chloromycetin required to protect 50% of infected embryos for 7 to 9 days was 3 to 4 times as great as the dose of aureomycin required to produce the same effect. Since the molecular weight of aureomycin is 508[7] as compared with 323 for chloromycetin,[8] the PD_{50} of the former is more than 5 times that of the latter on a molecular basis.

Statistically, the observed differences were highly significant.

[7] Broschard, R. W., *et al.*, *Science*, 1949, **109**, 199.

[8] Controulis, J., Rebstock, M. C., and Crooks, H. M., Jr., *J. Am. Chem. Soc.*, 1949, in press.

These findings were rather unexpected in view of the rapid deterioration of aureomycin *in vitro* at neutral or alkaline pH, and at the temperature employed.[9] Other factors in the embryo, however may affect the stability of the two agents and some of these factors are now under study.

Conclusions. Both aureomycin and chloromycetin prolong the life of chick embryos infected with the 6BC strain of psittacosis virus. There is a direct relation between the dose of antibiotic and the prolongation of life of the embryos, the infecting dose being kept constant. With equal doses of the antibiotics, aureomycin prolonged the life of the embryos for an average of from 2 to more than 3 days longer than chloromycetin, depending on the dose used.

On a weight basis, aureomycin was more than 3 times as effective in protecting chick embryos against the dose of virus that was used in this study. On a molecular basis the aureomycin was more than 5 times as effective as chloromycetin in this infection.

9 Paine, T. F., Jr., Collins, H. S., and Finland, M., *J. Bact.*, 1948, **56**, 489.

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Hypotension Associated with Nutritive Failure.* (17434)

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During the 12 years we have been studying persons with nutritive failure in a large Nutrition Clinic, we have learned that there is a great variation in blood pressure among such persons. Readings range from those which are too high to be obtained on an ordinary sphygmomanometer to those which are extremely low. In general, however, we have found that in the person with nutritive failure the blood pressure usually is below normal and that it tends to rise slowly as the nutritive failure is corrected. Even in patients with both hypertension and nutritive failure, we have observed that the blood pressure tends to increase slightly when the nutritive failure is corrected and that they lead a more energetic life.

It occurred to us that it would be worthwhile to determine the effect of desoxycorticosterone acetate on blood pressure. Since it has been known for some time that the development of edema, hypertension, and cardiac failure are the chief dangers associated with overdosage of desoxycorticosterone acetate,[1-4] we selected for study 3 persons who had been under observation in our Clinic for many years and who had every evidence of a properly functioning cardiovascular system except that the blood pressure was below normal. Each morning for a month these patients came to the Clinic early for special consideration and study. After they became accustomed to this routine, their blood pressures were taken under resting conditions each morning at 20 minute intervals over a period of 2 hours for 2 days. Then they were given intramuscular injections of sterile saline and the blood pressure readings were taken as before. This procedure was continued for 4 days. When no systolic reading above 100 and no diastolic reading above 65 was obtained, the patients were considered

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† Clayton Foundation Fellow.

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