analysis of the feces revealed no dye. This is not surprising since its presence in the stool requires a patent biliary system. In regard to renal excretion, Table II shows that in both species studied, less than 55% of the administered dye appeared in the urine.

Discussion. It is apparent that the results obtained in dogs and rats with regurgitation jaundice differ significantly from similar observations in human subjects(1). The rapidity with which BSP disappears from the serum of these animals suggests that other mechanisms are present for removal of dye in the absence of hepatic excretory function.

Cohn, Levine and Streicher showed that BSP is removed from the serum of the hepatectomized dog(7). Our data indicate that the dye is similarly removed when normal hepatic pathways are blocked. These data, however, are not applicable to BSP removal in man(1,8).

It has been stated that the kidney is able to excrete significant amounts of BSP when the serum concentration remains elevated for prolonged periods(9). In our experiments, 30-50% of the amount of dye given was recovered in the urine. Mechanisms of removal or destruction other than renal, therefore, probably play a role when the dye is not completely excreted through normal channels. In this connection, Brauer and coworkers have demonstrated that BSP may be chemically altered by the liver(10,11).

Summary. In dogs and rats with complete biliary obstruction, the retention of BSP in the serum was of much shorter duration than that previously observed in man. Conclusions concerning mechanisms of BSP removal in man based on observations made in the dog must be questioned in the light of these data.

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Effect of Chloromycetin and Streptomycin on Embryonic Tissue Growth in *In vitro* Tissue Culture. (19384)

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In a recent survey of the literature, it was noted that few reports have been presented concerning the effects of chloromycetin and streptomycin on normal cells in *in vitro* tissue culture. Ikegaki(1) has reported on the depressive effect of streptomycin on tissue growth, while Rodkova(2), Lepine, *et al.*(3) have presented conflicting data concerning chloromycetin.

Experiments were conducted in our laboratories with the following aims: (a) To satisfy increasing interest in the action of antibiotics on normal cells, with a view toward using antibiotics to help maintain sterility in tissue cultures. (b) To attempt to confirm the few existing reports.

Methods and Materials. Chloromycetin: Test solutions of chloromycetin were made up in the Tyrode portion of the usual tissue culture media(4) which was made up to the following concentrations in the final nutrient media: 1200, 900, 600, 480, 360, 240, 120 and

60 g/ml. *Streptomycin*: Test solutions of streptomycin sulfate were made in the same manner previously described in the following concentrations: 120000; 90000; 60000; 45000; 30000; 15000; 6000; 4500; 3000; 1000; 600; 450; 300; 150; and 60 g/ml. *Tissue culture:* Tissue culture studies were conducted by the roller tube technic using a chicken plasma clot. Chicken plasma was obtained commercially (Difco), while the chick embryo juice was prepared in these laboratories. Chick embryo heart explants, cut into pieces about 1 mm square were obtained from 9-day-old chick embryos. The radius of growth about the explant was measured after 48 hours incubation at 37°C. The "average index of growth" listed in the tables is the average radius of new growth about the explants in the indicated groups with their respective standard errors. Duplicate experiments were conducted.

Results. Table I. *Chloromycetin.* Concentrations up to and including 360 g/ml had no significant inhibitory effect. A slight but consistent increase in growth was observed at 240 μg/ml, but this cannot be borne out statistically on the basis of the small number of cultures tested. Growth was reduced but not completely inhibited at concentrations of 480 μg/ml and higher. The latter substantiates the reports of Rodkova and Lepine.

TABLE I. Effect of Chloromycetin on Embryonic Cells in Tissue Culture. Growth index in relationship to chloromycetin free controls.

Conc. of chloromycetin, μg/ml	Avg index of growth* Exp. 1	Exp. 2	No. of explants
Controls	37.6 ± 5.2	64.7 ± 2.3	43
60	35.7 ± 4.2	65.7 ± 4.2	10
120	45.8 ± 8.2	60.6 ± 4.6	14
240	48.3 ± 8.1	72.5 ± 4.6	14
480	41.1 ± 4.4	55 ± 4.1	16
600	29.9 ± 3.8	52.5 ± 3.4	17
900	22.1 ± 1.4	15.7 ± 4.2	14
1200	20.4 ± .04	2 ± .04	13

* Index is average radius of new growth about the explants.

TABLE II. Effect of Streptomycin on Embryonic Cells in Tissue Culture. Growth index in relationship to streptomycin free controls.

Conc. of streptomycin, μg/ml	Avg index of growth* Exp. 1	Exp. 2	No. of explants
Controls	89.9 ± 4.6	46 ± 6.6	31
60	67.8 ± 5.8	45.1 ± 1.8	14
150	69.1 ± 5.4	Not read†	15
300	72.7 ± 6.4	Not read†	10
450	45 ± 6.2	43.3 ± 4.2	12
600	61.6 ± 4.2	47.8 ± 3.6	15
1500	64.4 ± 6	51.1 ± 3.6	16
3000	54.3 ± 3	39.1 ± 4.8	13
4500	58.3 ± 4	27 ± 6.4	13
6000	54 ± 2.6	Not read†	11
15000	17.7 ± 8.2	11.5 ± 3.2	13
30000	12.2 ± 2.1	5 ± 4	12
45000	2.2 ± 1.1	3 ± 2.4	13
60000	Not read†	3.3 ± 1	16
90000	No growth	No growth	17
120000	,,	,,	15

* Index is average radius of new growth about the explants.
† Explants not clotted.

Table II. *Streptomycin.* Growth was markedly reduced by concentrations of 6000 to 15000 μg/ml and was completely inhibited by a concentration of 90000 μg/ml. Generally this is in agreement with the results of Ikegaki.

Summary. Growth of normal embryonic tissue was completely inhibited by concentrations of 90000 and greater μg/ml of streptomycin. Growth was partially inhibited by concentrations of 480 μg/ml through the highest concentration of chloromycetin tested.

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