0.2 cc of each mixture was injected intracutaneously on the back of a single rabbit. The results are shown in Chart 2.

The remarkable immunological and biological identity between the *Toxoplasma* of animal origin and the first strain of human origin suggests that the same protozoön may operate in all susceptible mammals, a fact which must be considered in the epidemiology of toxoplasmosis. The incidence of toxoplasmosis in animals and human beings remains to be determined, and the existence of clinically inapparent or unrecognized non-fatal cases will very likely be found to play a definite rôle in the dissemination of the infection. These studies also suggest that unless the parasites of birds which resemble *Toxoplasma* morphologically, but are not pathogenic for or do not multiply in mammals, can be shown to possess some immunological relationship to the classical *Toxoplasma,* they should be included in a separate group.

Conclusions. *Toxoplasma* of animal and human origin have been shown to be identical biologically in their pathogenicity for mammals and birds, and immunologically by producing an active immunity against one another and by the fact that a serum against one neutralizes both.

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Hypercalcification, -Calcemia and -Lipemia in Chickens Following Administration of Estrogens.

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Previous studies have shown that pigeons treated with estrogenic substances show a decided rise in blood calcium and a replacement of the marrow cavity by newly-formed endosteal bone.[1] It seemed of interest, therefore, to study the effect of estrogen on the bones and on the blood calcium of the domestic fowl. While our experiments were in progress, Zondek reported that estrogens increase bone calcification

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1 Pfeiffer, C. A., and Gardner, W. U., *Endocrinology,* 1938, **23**, 485.

and raise the blood calcium level of chicks of both sexes.[2] These changes were associated with dwarfism.

Our material consisted of fully-grown and sexually-mature cocks. We used normal and Creeper cocks, the latter belonging to a breed characterized by chondrodystrophy-like disproportionate dwarfism. Except for one cock, which was 9 months of age, the animals were about 6 to 7 months old when the injections were begun. The body weight of the animals ranged from 870 g to 1620 g at the beginning of the experiment. Nine Creeper and 6 normal cocks were treated with daily injections of estradiol benzoate‡; 5 Creeper and 5 normal cocks served for controls. In addition, similar tests were made with 2 capons (one normal, one Creeper), and 2 capons were used for control. The hormone injections were continued for various periods, as little as 3 weeks in some cocks, and up to over 4 months in others. The daily dose of estrogen varied between 1,700 and 30,000 I. U. With one exception, the cocks made normal gains in body weight during the experimental period.

At the beginning of the experiment the cocks still had rather small combs and wattles. During the course of the injections they decreased in size and assumed the typical capon-like appearance. The cock which was 9 months old when the injections were started, though he responded to the treatment in very typical fashion, still had a large, though somewhat mealy-looking comb after 3 weeks when the experiment was concluded. The testes of the treated cocks were very small, with one exception weighing less than one gram. The testes of the 9-months-old cock weighed 11 g when he was killed at the age of 305 days (about half the normal weight). Six of the 15 treated cocks showed some development of Müllerian rudiments.

After 3 weeks of daily treatment with approximately 1,700 I. U. of estradiol benzoate, inspection of the femur and tibia showed a thin layer of newly deposited calcium on the endosteal surface of the bones. With continued treatment the bones showed more extensive ossification. Treatment with 10,000 I. U. of the hormone daily produced within 3 weeks nearly as extreme bone changes as those found after a much longer period of treatment with smaller amounts of hormone. Deposition of new bone occurred uniformly throughout the epiphyses and diaphyses. The walls were uniformly thickened and trabeculae of spongy bone invaded the marrow. In the most extreme cases the marrow spaces of the metaphyseal regions were

[2] Zondek, B., *Folia Clin. orient.*, 1937, **1**, 1.

‡ The estradiol benzoate was generously supplied by Schering Corporation through the courtesy of Dr. E. Schwenk.

nearly obliterated and the marrow cavity was much narrowed near the center of the diaphysis. A definite relationship was found between the daily dose of injected hormone and the extent of new calcification.

The serum calcium of our control cocks varied from 8.96 to 9.61 mg %.§ With one exception, all cocks which received daily injections of 4,600 I. U. or more of estrogen showed increased serum calcium values (11.04 to 78.00 mg %). In 4 cases these values were in excess of 40 mg %. The one exception relates to a cock who had lost weight before he was killed and who was in very poor condition. The extent of the bone changes was related in part to the serum calcium level.

The cocks which had received the highest daily amounts of estrogens had extreme lipemia.[3] The blood serum was a deep canary yellow color, very turbid, and on standing quite a large amount of fat would rise to the surface. The highest figures for lipids were: fatty acids 573.2 m eq., cholesterol 1059.0 mg %, free cholesterol 790.0 mg %, lipid phosphorus 201 mg %, compared to the highest value found in the controls; fatty acids 10.61 m eq., cholesterol 128 mg %, free cholesterol 33 mg %, lipid phosphorus 10.36 mg %.‖

Also the cocks which had been injected with the highest daily amounts of estrogen had light yellow or ochre-colored livers, presumably with a high fat content. Their kidneys were enlarged, pale and had an irregular surface, in some instances with cyst-like protrusions. In three of the treated cocks an aneurysm of the vena iliaca externa was observed.

The normal and Creeper cocks showed no difference in response. The treated capons showed the same blood and skeletal changes observed in the treated roosters.¶

§ Serum calcium methods used described in 1 above.

[3] See also Lorenz, F. W., Chaikoff, I. L., and Entenman, C., *J. Biol. Chem.*, 1938, **126**, 763.

‖ Methods used for lipoid determinations are described in *J. Biol. Chem.*, 1932, **99**, 43; 1933, **101**, 695; 1937, **117**, 183; and 1937, **122**, 77; *Am. J. Physiol.*, 1938, **122**, 73.

¶ Since the conclusion of our experiments Zondek and Marx have reported calcemia and lipemia in young cockerels treated with estrogens. *Arch. Internat. Pharm. Dynam. and Therap.*, **61**, 11.