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**Influence of Sera on Development of the Chick Embryo.**

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This report deals with conclusions arrived at following studies of embryonal tissues when subjected to the influence of sera, normal and pathological. The author, in collaboration with Barratt,<sup>1</sup> reported the inhibiting effect of embryonal tissues on the growth of implanted cancer in mice. Since then the influence of organ extracts and sera on the development of foetal structures and cell formations has been continuously studied by the writer. During these experiments, it was revealed that cancer serum and serum from pregnant and menopausal women had a marked growth-promoting effect on embryonal tissues.

The fertile hen's egg proved the most satisfactory medium. We began by injecting fertile eggs with sera chosen at random. Later, when abundant sera, from subjects with malignant affections, became available from an authoritative source, systematic examinations were undertaken to discover any selective action of sera on embryonal tissues.

Hundreds of sera were used. In all cases of cancer and sarcoma the clinical diagnosis was confirmed histologically from material obtained by biopsy or surgical procedure.

To determine the optimal conditions, sera, active and inactivated, whether fresh or previously in the refrigerator from 3 to 30 days, in varied dosages, were employed. Injections into the fertile eggs were made at various stages of development of the chick embryo. Before using, the sera were kept in the incubator for 24 hours to test sterility. Control sera were obtained from numerous individuals who exhibited a wide range of pathological conditions.

*Technic.* The large end of the egg is painted with tincture of iodine, then punctured with a small sterile hand drill. A fine needle is inserted into the center of the egg, and the serum slowly injected. The minute opening is then sealed with a drop of sodium silicate and the egg is replaced in the incubator (38°C) until the end of the hatching period, usually 21 days.

Data ascertained. 1. The optimal time for injection into the fertile egg is the 4th or 5th day of incubation, when, on transillumination, the blood vessels of the developing embryo and the extra-

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<sup>1</sup> Barratt, J. O. W., and Gelarie, A. J., *Z. f. Krebsforsch.*, 1913, **13**, 1.

embryonic vascular area can be clearly seen. 2. The quantity of serum required for successful results is from 0.5 to 1.0 cc, preferably 1.0 cc. 3. The age of the sterile serum makes no difference in the results.

More than 200 cases of cancer and sarcoma, in different stages and grades, were examined. The controls included sera from more than 100 cases, of varied pathological conditions involving practically all organs including acute and chronic diseases. Sera from pregnant and menopausal women were also used for comparative tests. The results of these tests were remarkably consistent.

*Conclusions.* In 118 out of 122 cases where there were no indications of tumors, injection of the serum was followed by complete arrest of development of the chick embryo, the fertile egg substance undergoing degeneration, usually after the eleventh day of incubation. Confirmation, by waiting until the end of the hatching period, is advisable. The 4 negative results occurred in 2 male cases; one with mitral insufficiency plus migrating phlebitis, and the other with erythema multiforme and trigeminus neuralgia. The other 2 cases were women, one in menopause with atrophic arthritis, and tuberculous ulcers of both corneae; the other with a chronic parenchymatous nephritis.

In marked contrast with these results, when serum from a case of malignant tumor or from a pregnant woman was used, the development of the embryo proceeded normally until a full term chick was hatched. In some cases, the growth was even accelerated, shortening the incubation period, the chicks being noticeably larger and more precocious. Positive results were obtained in 216 cases out of 220. In this group the sera which gave a negative result were 2 with squamous carcinoma of the larynx, one with spindle cell sarcoma of the scapula, and one with melanoma of the face.

The causes of the isolated failures cannot be predicated at the present time. It is to be noted that on the 11th or 12th day, chemical examination of the egg contents for carbohydrates shows marked quantitative differences between eggs injected with cancer serum and those injected with non-cancerous serum. Whatever the explanation, the fact has been established that non-cancerous sera inhibit the development of the chick, while cancerous sera sustain or even accelerate it.