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7158 P

Absence of Effect of Antuitrin-S Injections on the Immature Ovary.*

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In a report¹ dealing with the effect of Antuitrin-S on the mature human ovary, it was pointed out that the response was not similar to that obtained in the rodent. It was therefore planned to inject a series of immature human females and examine their pelvic organs. Naturally such an opportunity is limited, for the cases must be selected from patients who are suffering from diseases that are of themselves fatal.

We have had opportunity to study 2 cases, one an infant of 10 months and one of 18 months. Both were suffering from tuberculous meningitis, proved by bacteriological examination of the spinal fluid and later confirmed at autopsy.

In the first case a 10 months infant received a total of 2,500 rat units of Antuitrin-S† in 5 divided doses over a period of 96 hours. The injections were given intramuscularly in the buttocks. The last injection was given 12 hours before death. At autopsy the ovaries both macroscopically and microscopically presented no evidence of follicular maturation or luteinization. The uterine mucosa showed no evidence of hypertrophy or cystic glandular hyperplasia. The second case, an infant of 18 months, also received a total of 2,500 rat units of Antuitrin-S. The last injection in this case was 16 hours before death. At autopsy the ovaries both macroscopically and microscopically presented no evidence of follicle maturation or luteinization. One ovary showed a tiny pinpoint area of

*I wish to thank Doctor Frank Spielman for his assistance.

¹ *Am. J. Obstet. and Gynecol.*, 1933, **26**, 588.

† The material was supplied by Parke-Davis & Co., whom we wish to thank for their cooperation and interest.

hemorrhage on the surface. The uterine mucosa did not show any variation from the normal.

Several patients desperately ill with gastro-intestinal and brain tumors received similar or greater amounts, but recovered from their illnesses.

The examination of a complete series of cases up to the age of puberty will of necessity require much time.

Conclusions. 1. Antuitrin-S intramuscularly and intravenously apparently produces no by-effects. 2. Antuitrin-S in 2 cases failed to produce follicular maturation or luteinization in the immature human ovary or changes in the uterine mucosa.

7159 C

A Method of Obtaining Undiluted Tissue Juice for the Study of Tissue Antibody Titer.

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The problem of obtaining suitable material for the determination of the antibody content of tissues or organs has usually been met by grinding the tissues with sand and extracting in a solvent, either physiological saline alone or equal parts of the saline solution and glycerine. The method is tedious and the material obtained has already undergone dilution which is not always desirable. The ease with which tissue juices are obtained by pressure in certain experiments carried out by Dr. Foster of the Department of Biochemistry suggested to us the possibility of extracting organs by this procedure for use in immunological experiments. The method consists in placing the tissue in an hydraulic press and extracting its fluid content under a pressure of approximately 16,000 pounds per square inch. The method may be used for any organ but is particularly serviceable in extracting skin and subcutaneous tissue where the large amount of connective tissue makes other methods of extraction difficult.

The tissue to be extracted is cut into pieces and mixed with a large volume of portions of filter paper about 1 cm. square. The whole mixture is cut up further until it consists of a grumous mass of filter paper which has absorbed all the bits of tissue to it. The

filter paper offers traction and prevents the tissues from sliding out of the press. This mixture of filter paper and tissue is then wrapped in a piece of cheese cloth and is ready to be pressed for the extraction of its fluid content. The press used is an hydraulic press manufactured by Fred S. Carver of New York City, capable of exerting a pressure of 20,000 pounds per square inch. As the press is too large to extract the small amounts of tissue used in our experiments, a small press to fit inside the large one was constructed. This consists of a solid steel cylinder 4.7 cm. in diameter at the base. Midway from the base the cylinder is turned down to a diameter of 4 cm. The whole is enclosed in a brass jacket which extends 1.2 cm. above the steel cylinder and which contains a spout to drain the space between cylinder and jacket at the point where the cylinder is narrowed. Another solid steel cylinder 4 cm. in diameter and 3 cm. high is used as a plunger to exert the pressure from above.* The accompanying diagram illustrates the construction of this press. The packet of tissue to be extracted is placed in this small press and

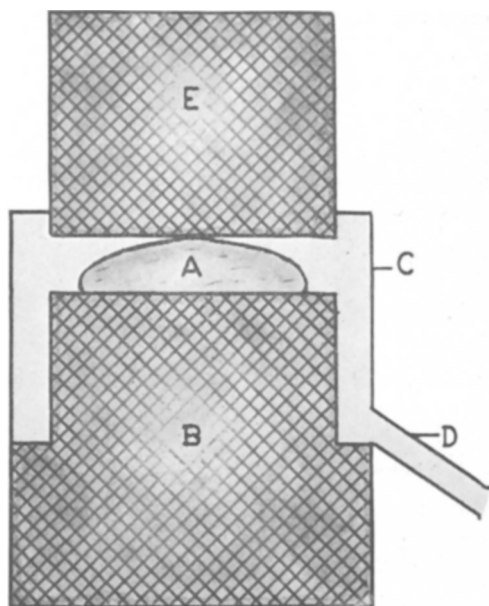


DIAGRAM I. Cross section of small press which is placed inside large press to extract fluid content of tissues. A, Packet of cut up tissue and filter paper. B, Solid steel cylinder. C, Brass jacket. D, Spout to collect fluid. E, Solid steel plunger used to exert the pressure from above.

* We are indebted to Mr. Rosebury of the Department of Biochemistry for constructing this small press.

the whole is placed in the large press under an average of 16,000 pounds per square inch of pressure. When small pieces of tissue are extracted, about 15% to 20% by weight is recovered as fluid. The juice thus obtained contains considerable fat and is therefore allowed to stand in the icebox overnight. The following morning it is centrifuged in the cold centrifuge† and the clear fluid which lies underneath the fat is removed with a capillary pipette. This extracted fluid is suitable for carrying out either precipitin or agglutinin determinations. When it is to be used for precipitin determinations, repeated centrifuging in the cold centrifuge is usually advisable. Since these organ extracts sometimes precipitate spontaneously when incubated at 37°C., all tests are put in the icebox immediately and are read in 24 hours and again in 48 hours, according to the method of Heidelberger, Kendall and Soo Hoo.¹ The end titers obtained in the icebox are the same as those obtained by a preliminary incubation at 37°C. followed by icebox treatment.

The pressure exerted by this method of extraction is apparently sufficient to break up body cells, for stained films of the residue of dry filter paper and tissue have failed to show any intact cells.

The agglutinin titer of tissue juice obtained by this method has been compared with that obtained by the method of Cary² of grinding with sand and extracting with equal parts of physiological saline and glycerine. The titers obtained by the pressure extraction are equal to or higher than those obtained by the method of Cary.

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Local Organ Hypersensitiveness. VII. Demonstration of Agglutinins in Tissues of the Rabbit Eye Following Immunization with Eb. Typhi Vaccine.

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The injection of a foreign protein into the anterior chamber of the rabbit's eye results in a local sensitization of this organ. Weeks

† Manufactured by the International Equipment Company, Boston, Mass.

¹ Heidelberger, M., Kendall, F., and Soo Hoo, C. M., *J. Exp. Med.*, 1933, **58**, 137.

² Cary, W. E., *J. Med. Research*, 1922, **43**, 399.