the toxin of the control sample, but the radiated sample was definitely attenuated in as short a time as fifteen minutes.

The second series was performed with cooling during the process of radiation. The toxin was placed as a film about 0.5 mm in thickness between two walls of concentric tubes and a chilled fluid was circulated in the inner tube.[1] The type of fluid used for the cooling was found to be of very great importance, because absorption of the electric waves by the central core of cooling fluid conditioned a loss of energy available to affect the toxin. The balanced molecular structure of benzol is such that it has no resultant dipol, its dielectric constant is the same at all frequencies, and therefore no absorption bands should be anticipated. The temperature of the thin toxin layer was determined by the use of a thermocouple at all times during the course of the radiation, and additional experiments were performed to investigate any direct action of the radiation on the thermocouple. Corrections were made accordingly for this latter action. The controls in this series were placed in similar thin films and at the same temperatures. Radiation of the type described above with the benzol cooler was found to be active in producing deterioration of the toxin in thin films at temperatures as low as 15° C. Similar results were obtained with cold air as a cooler.

It is notable at this time that although D'Arsonval had pronounced that a film of toxin was necessary in such work, the toxin in our hands was not in the beginning radiated in a film with any object other than efficient cooling. Later when the means were devised to cool a full column of the toxin, the radiation on such a column was found to be ineffective at temperatures under 18° C. It is remarkable that although the physical conditions of D'Arsonval's work are very remote from those employed here, we still arrived independently at the same conclusion; namely, that a film of the toxin seems to be essential to the greatest action of the radiation.

The method of assay of the results obtained was both by skin tests in guinea-pigs and by tests of the lethality of the toxin as well. In general, so far in the work, the two methods of assay have been found to be in close parallel, with the skin test being a somewhat quicker and possibly more delicate measure. The inactivation of the toxin is not complete, but by fifteen minutes' radiation with the benzol cooler at temperatures never above body temperature, the toxicity is so diminished that twenty-five skin test doses injected intradermally into a guinea-pig give the same reaction as that obtained with one skin test dose of the control toxin. One hour's radiation makes twenty-five skin test doses equal to one half of one skin test dose, and six hours' makes fifty skin test doses (one minimum lethal dose) equal to one half of one skin test dose of the control.

The exact nature of the mechanism of the change is not clear at present, but that the action occurs without heat effect is apparent. The different responses to changes in wave-length and the action on the two other major toxins, botulinus and tetanus, together with development of methods to measure accurately the output between the plates at all times may in the future clarify the problem.

We are further interested in the suggestion of D'Arsonval that the irradiated diphtheria toxin should be investigated with regard to its properties as an immunizing substance.

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[1] The efficiency of such a cooling mechanism was tested by special experiments with the whole device immersed in a water bath at 55° C. The cooler was found to preserve the toxin unchanged for three hours. Such a temperature was never approached in the radiation experiments.

RESULTS OF TOTAL AND PARTIAL ADRENALECTOMY AND ADRENAL TRANSPLANTATION IN THE ALBINO RAT

IT has generally been considered that a large proportion of white rats will survive total extirpation of the adrenal glands, and this has been attributed to the presence of accessory adrenal tissue. In the course of experiments on the survival of adrenalectomized rats at different atmospheric pressures it soon became evident that all the control animals, *i.e.*, adrenalectomized but kept at normal atmospheric pressure, died showing the classic symptoms of adrenalectomy, *viz.*, excessive prostration, muscular weakness, anorexia, etc. On the contrary, those rats in which a small fragment was left *in situ* survived indefinitely. This was so directly opposed to the results of other investigators that we have made it a point of special study.

To date, a total of forty-eight rats has been operated on. In thirty-two of these both adrenals were removed at the same time. All these animals died, the survival period varying from three to twenty days. None of these rats gained in weight after the operation; on the contrary, there was a steady decline. Rat ♂ 2035, typical of all these cases, was at the time of operation seven months old, weighed 358 grams, was in excellent condition; it died eight days after operation and weighed 304 grams; at autopsy

it was found that the fat was entirely depleted, there were no adhesions and the wound was cleanly healed.

In five rats a very small portion of one gland was left *in situ*. All these rats survived; there was never loss in weight throughout the period they were under observation. Rat ♀ GH4126 is typical—at time of operation it weighed 167 grams, and now, 125 days after operation, it weighs 193 grams and is in excellent condition.

This, however, does not rule out the possibility of the presence of accessory adrenal tissue, and that such tissue may have taken over the function of the normal gland. To test this point we transplanted a small fragment of adrenal tissue within the ovarian capsule, inserting it through a small slit. In five successful transplants the animals showed no loss in weight and appeared indistinguishable from normal animals for an indefinite time. Two of these rats are still living and are in perfect condition. Three were observed for approximately a month, at the end of which time the ovary containing the transplant was removed. Had accessory tissue been present, these three rats should have survived, since the second operation did not involve the site of the first, and removal of one ovary does not interfere with the normal condition of the animal. But following the removal of the ovary, in each case, there was the gradual loss in weight characteristic of adrenalectomized animals, and the usual train of symptoms appeared. One died within eight days. The other two died within fifteen days.

This work shows that the rat is no exception to the rule; it does not survive complete adrenalectomy, and that very small fragments left *in situ* or successfully transplanted within the ovarian capsule suffice to keep the animal alive and in good condition.

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THE METABOLISM OF THE BRUCELLA GROUP IN SYNTHETIC MEDIA

MANY important questions concerning the biology of the Brucella group remain unsolved. As a part of an extensive program an effort has been made to analyze the elementary food requirements according to the principles evolved by Braun. After considerable experimentation a series of synthetic media in which 22 strains of the Brucella grow freely have been developed. Doubly distilled water, highly purified chemicals and Pyrex glassware have been used in the experiments. The growth, although moderate in the first generation, has improved after the fifth passage, indicating a gradual adaptation of the strains to the protein- or peptone-free environment. The following facts are worthy of note.

Cystine is one of the essential N-sources. Asparagine in combination with cystine enhances multiplication. Sodium or ammonium citrate serve as carbon and energy sources. Cultures of recently isolated strains are obtained provided sodium carbonate and potassium acid phosphate is mixed in concentrations which will give a pH of 7.0. In all probability the CO_2 thus evolved fulfils the requirements of the sensitive strains. Twenty parts per million of iron either as ferrous or ferric ion has a stimulating effect. Glycerol is not indispensable but in concentration of 2 per cent. enhances the growth of every Brucella type. Physical and not chemical factors are responsible for the growth-promoting properties of this alcohol. The surface tension and physical consistency of the substratum play an important rôle. In liquid synthetic media the growth is sparse but the addition of 0.2 per cent. agar repeatedly washed in distilled and redistilled water accelerated the multiplication many fold. In this connection it is of interest that progressively increasing amounts of agar diminish the growth until it is completely inhibited at 1.5 per cent. concentration. A semisolid synthetic medium with 0.1–0.3 per cent. agar permits the cultivation of recently isolated CO_2 tolerant strains to grow at the normal oxygen tension of air.

The details of the various experiments on the N and S requirements, C and energy sources, essential cations and anions, optimal surface tension, osmotic pressure, utilization of carbohydrates, intramolecular respiration together with growth curves will be published in the *Journal of Infectious Diseases*.

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