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pattern for celluloid. For a large microscope piecing may be necessary. Use celloidin, or better, Dupont Duco Cement, permitting the first application to partly dry, apply a second layer of cement, hold in place with weights or pressures for ten minutes and the job is done. (I have such a cone fitted over a Leitz research microscope standing on a Chambers micro-dissection apparatus, all beautifully visible yet dust-proof.) A cone frustum would be better looking but slightly more difficult to make, though it is merely a matter of fitting in the top. It also would eliminate the piecing necessary for the cone.

By folding the celluloid over a wire frame to give better rigidity I have made a celluloid case to cover a Thoma-Jung microtome. Dr. E. P. Bartlett, seeing this, conceived the idea of making dust-proof cases for beam balances. These are folded and cemented like paper boxes.

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DEVELOPMENT OF A PERMANENT BLUE COLOR FOR COLORIMETRIC PHOSPHORUS DETERMINATION

The blue color used as a standard for Dénigés colorimetric method for the determination of phosphorus is very unstable. The color fades rapidly and a new color standard must be prepared rather frequently. By reducing a solution containing 2.5 grams of ammonium molybdate in 100 cc of 10 n. sulfuric acid a permanent blue color can be developed. The solution is reduced by stannous chloride. A dense blue color is formed which can be diluted to the desired intensity by adding 10 n. sulfuric acid. With proper dilutions a series of standards can be prepared which represent definite readings of phosphorus in parts per million.

The blue color developed under the latter condition is of a slightly different hue from the color of the reduced standard phosphorus solution but this slight difference in color is not enough to be objectionable. As a matter of fact, this permanent blue color compares as well to the unknown blue color as to the blue standard phosphate color.

The shades of blue color vary with the higher concentration of both ammonium molybdate and sulfuric acid. With the mentioned amount of ammonium molybdate in a slightly lower concentration than of 10 n. sulfuric acid a bright yellow color is produced upon reduction.

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SPECIAL ARTICLES

AN EFFECT OF SHORT ELECTRIC WAVES ON DIPHTHERIA TOXIN INDEPENDENT OF THE HEAT FACTOR

About thirty-five years ago D'Arsonval and Charrin found that high frequency currents of 200,000 cycles per second diminished the strength of diphtheria toxin. This effect was obtained without elevation of temperature to a level which would by itself affect the toxin. Since that time little has been done to develop this finding. Recent advances in short wave technique have given new impetus to the study of the biological action of these waves. It is readily accepted from many recent papers that such electrical waves may produce tremendous changes through the indirect medium of temperature elevation. Before any result is ascribed to the specific action of short electric waves, heat effect through conductivity and eddy currents must be ruled out. The chief import of this paper is to show that radiation of the type used here is capable of producing changes in biological substances independent of a heat effect.

Throughout, the wave-length was 1.9 meters and the substances to be radiated were placed between two condenser plates of a resonating circuit. The amperage in the resonating circuit was .95 to 1.2, and the frequency was 158,000,000 cycles per second.

From the beginning, and so far in this work—eliminating heat effect—completely negative results were obtained in attempts to sterilize milk and to destroy bacteria both *in vivo* and *in vitro*. Radiation *in vivo*, both as general radiation of the whole animal and local radiation to the site of injection, produced no changes in the course of streptococcus infections in guinea-pigs. In addition, no effects of the radiation could be detected on the precipitin titer of the pneumococcus antisera from rabbits.

The study of diphtheria toxin was made in two series of experiments. First series: One sample of toxin was chilled in ice water to 7° C., then exposed to radiation until the temperature had risen to 38°–40°C. (about four minutes). When such a temperature was attained the sample was taken out of the high frequency field and chilled again in the ice water. This process was repeated until the total time of radiation was fifteen to sixty minutes. A control sample was kept at the identical temperatures with the same rate of heating and cooling by alternate chilling in ice water and immersion in a small heated water bath. The temperature attained did not affect

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 in 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