

ness show no significant difference. Previous studies³ with cow peas have shown that roots of plants grown with their shoots in the light contain several times as much ascorbic acid as those of plants grown in darkness.

The excised moonflower roots cultured in the light possessed a deep green color due to the presence of well-developed chloroplasts. For this reason a strict comparison can not be made between these light-grown cultures and the colorless roots of intact plants grown in the light. The excised roots grown in darkness may be more nearly comparable to the roots of intact plants, since both lack chloroplasts.

It has been observed⁴ in other experiments with intact plants that roots with a relatively low content of ascorbic acid during a prolonged period of cloudy weather showed a marked increase of this substance following a day of bright sunshine.

Although the evidence is not conclusive, it seems probable that the increased quantity of ascorbic acid in the excised roots cultured in the light is due to the presence of well-developed chloroplasts. If this is true, it seems probable that the colorless roots of intact plants grown in the light do not synthesize vitamin C but receive their supply from the tops. It is planned to continue these studies, using excised roots which do not develop chlorophyll.

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A NEW MENINGOTOXOID

FOLLOWING the successful production of a toxin from gonococcus as well as a potent antitoxin against the same,¹ it was considered of interest to ascertain if similar procedures followed out with meningococcus might not also yield equally promising results. This note is merely to record briefly the method followed and the results obtained to date and is preliminary to a report to be made later.

THE METHOD

Ordinary broth having a pH of 7.7 was used. Two per cent. dextrose was added and the media distributed in quantities of 400 cc to diphtheria toxin flasks. It was seeded with an indigenous strain of meningococcus (No. 64) by planting a pellicle on the surface of the media. The culture was incubated at 37° C for 12 to 15 days until the surface growth or pellicle began to settle to the bottom of the flask. The broth culture was then filtered through Zeiss paper filters K and EK

³ Mary E. Reid, *Amer. Jour. Bot.*, 25: 701-711, 1938.

⁴ Unpublished data.

¹ Gonococcus toxin and antitoxin, *Zentrabl. f. Bakt., Parasitenkun., u. Infektionskr.*, I Abt. Orig. 1939, Bd. 143.

and the filtrate used for the first experiments. This filtrate was found to be so toxic that finally 3 to 4 per cent. of formaldehyde was added to it and then it was incubated at 40° C for 45 days. The toxin was then precipitated by adding 1.5 per cent. alum, the precipitate duly washed and finally dissolved by adding 4 per cent. sodium citrate. The toxoid thus obtained was used for the next series of experiments.

RESULTS

The unmodified toxin when injected into mice intravenously in quantities of 0.2 cc killed immediately; 1.0 cc injected intravenously into guinea-pigs was also lethal at once. The intradermal injection into guinea-pigs of 0.2 cc caused necrosis. This filtrate was thus observed to be of high toxicity. It also gave flocculation and precipitation reactions when tested with anti-meningococcus serum. Its antigenic properties in animals were found to be of a high grade. Repeated injections led to the development of antibodies, as proved by flocculation tests.

The filtrate treated with formaldehyde and then precipitated with alum afforded a much more satisfactory toxoid. This toxoid was much less toxic but retained all its capacity to elicit antibody formation. For instance, one month after the injection of a single dose of 1.5 of the alum-precipitated toxoid into rabbits the presence of antibodies in their sera was demonstrated.

In humans it was shown that consecutive injections of 0.5 cc, 1.0 cc and 1.5 cc of the non-precipitated toxoid or of 1.0 cc of the precipitated toxoid caused the formation of demonstrable antibodies in their sera.

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INCREASED SENSITIVITY OF HYPOPHYSECTOMIZED RATS TO RADIATION

IN a study of the metabolism of Na and K in relation to adrenal cortical physiology, we have administered radioactive isotopes of these elements to variously treated animals. We have been surprised to find that hypophysectomized animals could not tolerate these "tagged atoms" even in "physiological" doses (*i.e.*, 10 microcuries), while intact animals and adrenalectomized animals show no untoward effects from such doses.

Our attention was first called to this phenomenon in an experiment in which 10 hypophysectomized rats received approximately 10 microcuries of radioactive K and four such animals an equivalent dose of radioactive Na made up in a 1 cc isotonic solution and injected intraperitoneally. All the animals were dead within 48 hours. In a large series of normal rats to which the same dose of these radioactive isotopes had been administered, no ill effects have been observed.

We next selected ten animals which had been hypo-

physectomized thirty days previously and which were in satisfactory condition. Five of these rats were injected intraperitoneally with 10 microcuries of radioactive P in the form of 1 cc (15 mg) of an isotonic solution of Na_2HPO_4 . The remaining five were given an equivalent dose of ordinary Na_2HPO_4 . Within 48 hours after the injection of the radioactive P all five of the animals were dead, while the condition of the hypophysectomized controls remained unchanged.

The hypersensitivity of hypophysectomized rats to the injection of radioactive isotopes is in marked contrast to the response of adrenalectomized animals to similar doses of radioactive material. The major part of our experimental work in adrenal cortical physiology has been on adrenalectomized animals. We have injected radioactive Na and K into approximately 200 such animals and in no instance seen evidence of hypersensitivity to these substances.

Hypophysectomized rats are also more sensitive than normal animals to x-ray irradiation. Nine rats hypo-

physectomized three weeks previously were given 250 roentgens; this dose is well under the lethal dose for normal animals, which is approximately 700r.*; all the animals succumbed, but the median survival period was 10.6 days, as contrasted with a survival period of less than 2 days following the injection of radioactive isotopes.

It would seem established that hypophysectomized rats are abnormally sensitive to radiation, particularly when injected with radioactive isotopes, and it is to be noted that adrenalectomized rats tolerate radioactive isotopes at dose levels invariably fatal to hypophysectomized animals.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

A SIMPLE MECHANICAL THERMO-REGULATOR

THE regulation of temperature in a bath by controlled cooling, rather than by the usual method of controlled heating, is not a new principle,¹ but simple apparatus designed for precise temperature control has not been available hitherto. Advantages claimed for this regulator over the customary electrical type are comparable sensitivity at lower cost, with greater dependability and simplicity.

Since a small stream of cooling water is constantly introduced into the bath, an overflow or syphon is provided to maintain the level. If desired, a small coil of copper tubing may be used to provide the necessary heat transfer between the cold stream and the bath liquid, thus permitting the use of non-aqueous bath media.

A heater is used continuously, and may be of any type: immersion, lamp, steam coil or even a gas burner placed beneath the bath, provided it furnishes a steady heat input. Customary precautions should be taken to insure adequate stirring and insulation of the bath.

If variation in the pressure of the tap water (cooling water) prohibits the maintenance of a constant, steady stream to the regulator, a simple constant-pressure reservoir should be employed, furnishing water at one to three feet hydrostatic pressure.

The operation of the thermoregulator is self-evident from the sketch of Type 1. After bringing the bath

to the desired temperature and removing excess mercury from the stem with a micropipette, if necessary, several minutes are allowed for equilibration of temperature within the regulator. Then with the mercury level below the orifice, D, in the stem, a steady stream of water is introduced into the top of the stem, its magnitude (controlled by screw-clamp, A) such that outlet C will easily handle it without overloading. Mercury is introduced into the stem from a micropipette until the orifice, D, begins to be occluded and water starts to flow into the bath through B. If the heat input to the bath is of the proper value, a steady stream of drops (or at most a thin stream of water) will hold the temperature stationary. Water must at all times be flowing simultaneously from both B and C when the bath is in equilibrium. The vent, E, prevents syphoning at C. Adjustment of the temperature to the last few hundredths of a degree may be done by varying screw-clamp A. When the bath temperature begins to fall, the regulator decreases the amount of water flowing into the bath, and *vice versa*. In permanent installations, screw-clamp A may be profitably replaced by a stopcock.

By reversing the connections at B and C, a supply of hot water entering at A will regulate the temperature of the bath through controlled heating.

Three types of bulb to be attached to the control

¹ D. F. Othmer, *Ind. and Eng. Chem., Anal. Ed.*, 3: 139, 1931; L. C. Beadle and F. A. Booth, *Nature*, 140: 279, 1937.

* The x-rays were generated by a 220 KV Maximar unit, which has been installed in the Crocker Radiation Laboratory through the generosity of the General Electric X-ray Corporation.