conduct research, provide a center of information, hold schools of instruction and demonstration, and organize and direct a practical course in woods industries of two twelve-week terms and one nine-week term.

ERRATUM: Page 148, 2d column, article by Dr. Gregory Shwartzman; the thirteenth line down on the right-hand column reads: "experiments which were all carried out in meat-inferior broth." This should have been "meat infusion broth."

SPECIAL ARTICLES

REVERSIBLE PARALYSIS OF MOTOR FUNCTION IN RATS FROM THE CHRONIC ADMINISTRATION OF DITHIOBIURET

DURING the course of investigations on the antithyroid activity of compounds related to thiourea it was observed that small concentrations of dithiobiuret (NH_2–C(=S)–NH–C(=O)–NH_2) were lethal within seven days.[1] It was soon apparent that death was preceded by paralysis and was presumably due to involvement of the respiratory muscles. This phenomenon was found to be readily reproducible and the effective dosage of dithiobiuret[2] proved to be quite critical. When administered in the drinking water in a concentration of 0.002 per cent. no effect was observed for 2 to 4 days and the animals continued to gain weight; then weakness of the hind quarters became apparent. Soon thereafter the muscles of the entire body with the exception of the muscles of respiration and those of the head and neck became completely paralyzed and the animals lay on their sides breathing with difficulty. If continued in this dosage the compound was uniformly lethal at the end of 5 to 6 days, but if reduced to 0.001 per cent. the animals would survive. By appropriate adjustments in the concentration of the drug between 0.001 per cent. and 0.002 per cent. the animals could readily be maintained in a state of profound muscular paresis for many weeks. With the onset of muscular weakness food and water intakes decreased and the animals began to lose weight; food and water were procured at the expense of great effort. The only constant associated finding was an incrustation about the eyes containing the red porphyrin pigment from the Harderian glands. There later ensued progressive weight loss, muscular wasting and contractures of the paralyzed muscles. The condition was consistently reversible, for despite the presence of extensive paralysis complete recovery followed a few days after withdrawal of the drug; even when atrophy and contractures had become manifest good recovery of muscle function followed within a week or two.

A single subcutaneous or intraperitoneal injection of relatively huge doses of this substance was well tolerated and failed to cause paralysis. A dose of 20 to 50 mg was necessary to kill an adult rat from a single injection, whereas fatal paralysis ensued in 5 days from the daily administration of less than 0.5 mg either in the drinking water or by subcutaneous injection.

The paralysis seemed not to be due to a disturbance of the muscles themselves, of the peripheral nerve or of the myoneural junction, for faradic stimulation of the motor nerves elicited muscular contraction. Nor did there seem to be any disturbance of pain sensation as judged by the responses to painful stimuli in partially paralyzed animals.

The brains, spinal cords and roots, spinal ganglia, sciatic nerves and muscles of animals which had been kept paralyzed for two to three weeks and of control animals were removed and sections were stained for cells, axis cylinders, myelin and fat. Microscopic examination disclosed no visible structural damage, a finding which is in keeping with the reversible nature of the disturbance. Determinations on the content of acetylcholine and of choline esterase in the brains of paralyzed rats yielded normal values.[3]

Attempts to elucidate the phenomenon by the administration of drugs which affect the nervous system and by a search for a remedial agent were unsuccessful. Strychnine failed to induce convulsions in paralyzed animals and double the usual lethal dose was required to produce death, which was accompanied by a slight flexure of the head upon the thorax and cessation of respiration. Pilocarpine and prostigmine did not improve muscular function in doses up to the lethal level. Atropine, epinephrine and ephedrin were likewise ineffective. Large doses of a crude liver extract, thiamin, nicotinic acid, vitamin A, biotin, brewer's yeast or biuret failed to prevent paralysis and caused no improvement when administered to paralyzed animals.

These observations appear to be of interest in that they probably point to the existence of a new mechanism which is essential to the transmission of nervous impulses within the central nervous system. They

[1] E. B. Astwood, *Jour. Pharm. and Exper. Therap.*, 78: 79, 1943.

[2] This compound was made available through the kindness of Dr. R. O. Roblin, of the American Cyanamid Co., Stamford, Conn.

[3] We are indebted to Mr. J. C. Seed for the acetylcholine determinations and to Miss Mary Root for the assays of cholinesterase.

indicate that the chronic administration of this simple chemical compound interferes with a hitherto unrecognized process, perhaps by making unavailable a component of some enzyme system.

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THE MECHANISM OF ACTION OF ALLOXAN ON BLOOD SUGAR

THE intravenous administration of alloxan induces a triphasic modification of the blood sugar level: (1) hyperglycemia; (2) hypoglycemia; (3) hyperglycemia. We have studied these phenomena in several species, particularly the chlorolosed dog (100 mg of alloxan/kg of body weight) and the toad *Bufo arenarum* Hensel (200 mg/kg).

The initial hyperglycemia did not appear in hepatectomized dogs or toads nor in eviscerated dogs. It was observed in adrenalectomized animals (5 dogs and 6 toads) and in 3 dogs with previous section of the splanchnic nerves (major and minor) so that it can not be attributed to either adrenaline or cortical hormones. It was also observed in recently hypophysectomized toads. If injected in the portal vein, alloxan produces a higher initial hyperglycemia (5 dogs) than if injected in the saphenous vein (8 dogs).

The secondary hypoglycemia is not due to liberation of insulin by the β cells of the islets undergoing destruction. Nine dogs totally depancreatized half an hour before injection of alloxan, showed a marked hypoglycemia beginning 1, 2, 2, 2, 2, 3, 3, 4 and 5 hours after injection; the blood sugar level reaching in 7 cases to 50 and 24 mg per 100 cc. Six of these dogs showed initial hyperglycemia. Pancreatectomized controls with no alloxan only in a few cases showed slight and brief diminution of the blood sugar level half an hour after the operation, followed by a gradual and steady increase from 2 to 6 hours after the operation, reaching at that time 0.149 and 0.180 g per 100 cc of blood.

In 7 dogs depancreatized 24 to 48 hours previous to the injection of alloxan there was no hypoglycemia: on the contrary, the blood sugar level was slightly increased. In only one case was there a moderate decrease (from 0.217 to 0.134 g per cent. between 5 to 6 hours after the injection).

In pancreatectomized toads, alloxan injected immediately after the operation either prevents or decreases the diabetic hyperglycemia during the next 24 hours. If injected 24 hours after pancreatectomy, the existing diabetic hyperglycemia decreases as shown by the blood samples 24 hours after injection. Alloxan also notably decreased the diabetogenic action of the *pars distalis* of the hypophysis when subcutaneously injected to the hypophysectomized and depancreatectomized toad.

The capacity of the pancreas to secrete insulin was investigated by grafting in the neck through vascular anastomosis the duodeno-pancreas of dogs to dogs rendered diabetic through pancreatectomy performed 24 hours before. Normal pancreas decreases the blood sugar to normal level within 3 to 5 hours. Pancreas from dogs injected with alloxan 24 hours (6 dogs) or 48 hours (2 dogs) before extraction and grafting did not secrete insulin in 4 cases, the secretion was very reduced in 3 cases and only in 1 was the secretion normal. It is interesting to note that the pancreas was taken in some cases from animals that were still hypoglycemic. The β cells of the islets showed lesions in all cases (Dr. Di Pietro).

The final rise of the blood sugar reaches sometimes (rats, rabbits and dogs) higher values to those usually observed after pancreatectomy. Values of 0.700 and 1.00 g per 100 cc of blood have been observed. Possibly the liver plays some part in this phenomenon.

Therefore: (1) The liver is essential for the initial hyperglycemia produced by alloxan. Hyperglycemia is observed in adrenalectomized animals and those with section of the splanchnic nerves. It must be attributed principally to a direct action of alloxan on the liver. (2) The secondary hypoglycemia is not due to liberation of insulin, but to an extrapancreatic effect: probably lack of glucose production by the liver. The liver of the animal already in a diabetic condition is generally insensible to this action of alloxan. (3) The final hyperglycemia is mainly due to the destruction of the β cells of the islets of Langerhans, and becomes permanent if the animal survives. (4) The liver plays an important role during the 3 phases of modification of blood sugar level.

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THE PRODUCTION OF ANTI-PENICILLINASE IMMUNE SERA

IT is well known that the injection of an antigen (precipitinogen) parenterally in animals stimulates the production of antibodies (precipitins). In order that a substance may be precipitinogenic, it apparently must contain a soluble protein.[1]

[1] F. P. Gay and Associates, "Agents of Disease and Host Resistance," Charles C Thomas, Baltimore, Md., 1935.