## THE EFFECT OF URINARY CORTIN-LIKE MATERIAL ON CARBOHYDRATE METABOLISM[1]

PREVIOUSLY we demonstrated that urinary extracts are capable of increasing the resistance of adrenalectomized rats to low environmental temperatures[2] as well as maintaining the lives of adrenalectomized rats.[3] Using glycogen deposition in the liver of the fasting adrenalectomized rat, we have now found that these extracts are able to influence carbohydrate metabolism in a fashion similar to that demonstrated for cortin and certain cortical compounds.[4,5]

The experimental procedure employed was a modification of the technique described by Reinecke and Kendall.[6] Adrenalectomized rats (140–160 gms) were fasted for 24 hours from the third to fourth day after operation. At the end of this period, the urinary extract was administered by stomach tube. Samples of the livers were removed and analyzed for glycogen by the method of Good, Kramer and Somogyi.[7] The urinary extracts used in this study are similar to those already described.[2,3]

In experiment A, the equivalent of 2 liters of urine dissolved in 1 cc of a 10 per cent. ethanol solution was administered once at 0 hours and again at 3.5 hours. The time of the termination of the 24-hour fast was reckoned as zero time. Samples of liver were removed between the seventh and eighth hours. In experiment B, the equivalent of 1.5 liters of urine dissolved in 1 cc of a 10 per cent. ethanol solution was administered at 0, 2.25, 4.5 and 6.75 hours. Samples of liver were removed between the eighth and ninth hours. All controls received 1 cc of a 10 per cent. ethanol solution at the hours indicated.

The results are presented in Table 1, in which the liver glycogen values are expressed in mgs of glycogen per 100 gms of liver (mg per cent.). The mean liver glycogen concentrations of those animals receiving urinary extracts were increased 5 and 10 times, respectively, for experiment A and B as compared with the controls. It is evident, therefore, that these urinary extracts influence carbohydrate metabolism in much the same way as do adrenal cortical extracts.

TABLE 1

THE INFLUENCE OF URINARY EXTRACTS ON THE DEPOSITION OF LIVER GLYCOGEN IN FASTING ADRENALECTOMIZED RATS

| Experiment | Total urinary equivalent administered per rat in equal doses (liters) | Total volume of solvent (10 per cent. ethanol) administered (cc) | Times of administrations after completion of 24 hr. fast (hrs.) | Number of rats | Mean glycogen in liver (range) (mg. per cent.) |
|---|---|---|---|---|---|
| A | 0 | 2.0 | 0 ;3.5 | 5 | 15.5 (13.9–19.0) |
| | 4.0 | 2.0 | 0 ;3.5 | 5 | 75.4 (21.1–128) |
| B | 0 | 4.0 | 0 ;2.25 ; 4.5 ;6.75 | 3 | 16.4 (12.5–19.3) |
| | 6.0 | 4.0 | 0 ;2.25 ; 4.5 ;6.75 | 7 | 178 (60.9–308) |

BENJAMIN N. HORWITT
RALPH I. DORFMAN

# SCIENTIFIC APPARATUS AND LABORATORY METHODS

## NON-VIRULENT FROZEN AND DRIED ANTIGENS FOR COMPLEMENT-FIXATION TESTS WITH CENTRAL NERVOUS SYSTEM VIRUS INFECTIONS

Two steps have already been taken in developing a specific, practical complement-fixation test for diagnosing central nervous system virus infections. The first was the production of antigens from infected brain tissue which, under stated conditions, gave specific and duplicable results;[1] the second was the rendering of the antigens non-virulent in order to make them safe for general use in hospital diagnostic laboratories.[2] We wish now to describe a third step, namely, the successful freezing and drying of small quantities of non-virulent antigens, thus permitting their distribution by central production laboratories and their use over relatively long periods of time.

Standard virulent antigens were prepared[1] by obtaining infected mouse brains, making them into a 10 per cent. suspension with diluent in a mechanical homogenizer, centrifugalizing the material at 2,500 r.p.m. in a horizontal centrifuge, freezing and thawing

[1] From the Brush Foundation and Department of Biochemistry, Western Reserve University School of Medicine, and Department of Medicine, Lakeside Hospital, Cleveland, Ohio. Supported in part by a grant from the Josiah Macy, Jr. Foundation.

[2] R. I. Dorfman, B. N. Horwitt and W. R. Fish, SCIENCE, 96: 496, 1942.

[3] R. I. Dorfman and B. N. Horwitt. In press.

[4] S. W. Britton and H. Silvette, *Am. Jour. Physiol.*, 100: 693, 1932.

[5] C. N. H. Long, B. Katzin and E. G. Fry, *Endocrinology*, 26: 309, 1940.

[6] R. M. Reinecke and E. C. Kendall, *Endocrinology*, 31: 573, 1942.

[7] C. A. Good, H. Kramer and M. Somogyi, *Jour. Biol. Chem.*, 100: 485, 1933.

[1] J. Casals and R. Palacios, SCIENCE, 93: 162–3, 1941; *Jour. Exp. Med.*, 74: 409–26, 1941.

[2] J. Casals, *Proc. Soc. Exp. Biol. and Med.*, 49: 501–4, 1942.