AN EFFECT OF ALLOXAN-DIABETES ON THE ACTIVE TRANSPORT OF SUGARS BY RAT SMALL INTESTINE, IN VITRO*

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On several occasions in the past it has been suggested (Pauls and Drury, 1942; Laszt and Vogel, 1946; Sols et al, 1948) that intestinal absorption of glucose is a more rapid process in the alloxan-diabetic rat than in the normal control. The supporting experiments, however, were carried out *in vivo* under conditions which did not provide enough information to decide whether the observed increases in glucose loss from solutions placed in the intestinal lumen were the result of an augmentation of the specific process of active sugar absorption, the salient feature of which is the transport of sugars against a concentration difference, or whether they reflected more general alterations in the absorptive or metabolic capacities of the small intestine (Crane, 1960). A reinvestigation of the question using a recently developed *in vitro* technique (Crane and Mandelstam, 1960) has provided data which indicate that the specific process of active sugar absorption is, on the average, more than twice as rapid in the alloxan-diabetic rat as in the normal control (Table I). In these experiments, all measurements were made under conditions in which

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Table I. The Active Transport of 6-Deoxy-D-glucose by Strips of Small Intestine from Normal and Alloxan-diabetic Rats.

	Number of animals	Average tissue concentration of 6-deoxyglucose
		μmoles per ml tissue water
Normal	9	2.9 (range: 2.0-4.2)
Alloxan-injected	11	6.9 (range: 4.5-9.7)

Alloxan (20 mg/ 100 g) was given by tail vein to animals weighing 150-200 g following a 48 hr fast. The animals were used 4+ days later. All animals were fasted 24 hr prior to use. The techniques of tissue preparation and incubation have been described by Crane and Mandelstam (1960). 0.4-0.6 g of tissue were incubated for 21 min at 37° in 10 ml of Krebs-Henseleit bicarbonate buffer containing 0.5 μmoles of 6-deoxyglucose per ml. Assay was made by the Dische and Shettles (1948) procedure for methylpentoses.

the test sugar was accumulated within the tissue to concentrations higher than in the medium thus giving assurance that the process of active transport was being observed.

The underlying reason for this apparent increase in active sugar absorption has not yet been found, although a number of possible alternative explanations can be excluded because of the techniques used. In the normal rat the proportion of glucose actively transported to glucose metabolized by *in vitro* preparations is relatively small (Nagler et al, 1960). Thus an alloxan- or diabetes-induced decrease in the rate of glucose metabolism by the intestinal epithelial cells would result in an apparently increased rate of active transport with this sugar. In the present experiments this possibility was excluded through the use of the actively transported but non-metabolized analog, 6-deoxyglucose

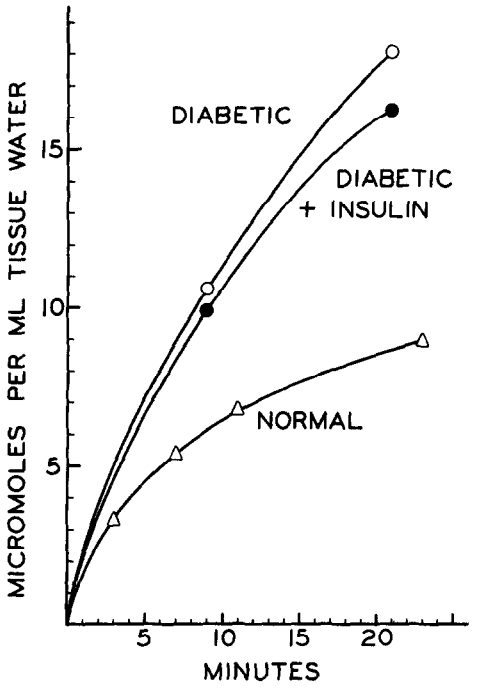

Fig. 1. The Active Transport of D-Galactose by Strips of Small Intestine from Normal and Alloxan-diabetic Rats. The initial medium concentration of galactose was 5 μmoles per ml. Other details as in Table I.

(Crane and Krane, 1956). In other experiments, one of which is shown in Fig. 1, similar results were obtained with D-galactose which is also actively transported but is not so rapidly metabolized as glucose (Crane and Mandelstam, 1960). Also shown by the curves in Fig. 1 is the fact that both the initial rate and the final steady-state level of accumulation are increased in the alloxan-diabetic animal. A number of attempts were made to decrease the diabetic rate of transport by *in vitro* addition or by subcutaneous injection of insulin shortly prior to killing the animal. These attempts were substantially negative. A small and irregular degree of inhibition was noted when 0.4 units per ml of crystalline insulin were present during incubation (e.g., see Fig. 1). There was no noticeable rapid effect of subcutaneous injection.

The data in Table II exclude several additional possibilities

Table II. The Active Transport of 6-Deoxy-D-glucose by Strips of Small Intestine from Normal and Alloxan-injected Rats.

Normal animals

Number of animals	Body weight	Duration of fast	Actual tissue concentrations of 6-deoxyglucose
	g.	hr.	μmoles per ml tissue water
2	150-200	48	3.1, 3.9
2	150-200	120	3.4, 2.9
4	65-70	24	4.6, 4.4, 4.8, 3.7
4	100-110	24	4.1, 4.5, 4.8, 4.7
4	150-160	24	2.8, 3.8, 2.6, 3.6
4	150-160	not fasted	2.2, 2.5, 2.5, 2.5

Alloxan-injected animals

Number of animals	Days after alloxan	Blood sugar (24-hr. fast)	Actual tissue concentrations of 6-deoxyglucose
		mg per cent	μmoles per ml tissue water
2	2	78, 320	3.8, 3.9
2	3	575, 264	6.3, 5.0
3	4	300, 385, 315	8.7, 5.4, 6.1

and further indicate that the increased rate of transport is a functional alteration characteristic of the alloxan-induced diabetic state. The rate of transport did not increase immediately following the injection of alloxan but rose over a several day period. Because of the retardation of growth in diabetic animals, a series of animals of different body sizes (ages) was studied and found not to differ significantly from the normal rat in the 150-200 g weight range.

In some respects, the present results are markedly similar to to the influence of alloxan-diabetes on the *in vitro* carbohydrate metabolism of rat liver (Renold et al, 1953, 1955). However, the effect of alloxan-diabetes on active sugar absorption, unlike its

effect on liver glucose 6-phosphatase levels (Ashmore et al, 1954), is not mimicked by fasting even of extensive duration.

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