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EFFECTS OF INSULIN ON THE UPTAKE OF D-GALACTOSE BY ISOLATED RAT EPIDIDYMAL FAT CELLS

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Summary

In muscle, insulin stimulates uptake of D-galactose as well as D-glucose and certain other sugar isomers (Kono, T. and Colowick, S.P. (1961) Arch. Biochem. Biophys. 93, 514—519). In fat cells, the hormone also stimulates uptake of D-glucose and certain other monosaccharides. Nonetheless, the hormone does not increase the uptake, as determined by the utilization, of D-galactose by fat cells (Ball, E.G. and Cooper, O. (1960) J. Biol. Chem. 235, 584—588; Kuo, J.F. and Dill, I.K. (1969) Biochim. Biophys. Acta 177, 17—26).

As pointed out by Ball and Cooper, this does not necessarily indicate that insulin has no effect on the membrane transport of D-galactose in fat cells. The possible effect of the hormone on transport may not be seen in the utilization data if the intracellular metabolism is considerably slower than the rate of transport and insensitive to insulin.

In the present study, we examined the above possibility by measuring the uptake of D-galactose with the oil-flotation method originally described by Gliemann et al. [4]. The experiment was carried out as described previously [5] with minor modifications, as noted in Footnote 3 of Ref. 5. Labeled sugar isomers were purchased from New England Nuclear.

The control experiments carried out with 3-*O*-methyl-D-glucose indicated that, in agreement with previous observations [5,6], the uptake of this sugar is stimulated by insulin at the beginning of incubation (Fig.1A). Since the

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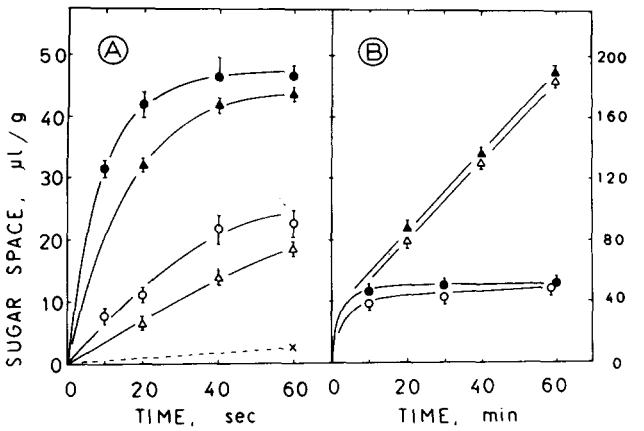

Fig.1. Time course of sugar uptake. Isolated fat cells were incubated with a mixture of 1 mM 3-*O*-methyl-D-[^{14}C]glucose and [^{3}H]inulin or 1 mM D-[^{14}C]galactose and [^{3}H]inulin for the indicated periods. Insulin, 1 nM, when present, was added 10 min before 0 s. Cells were separated from the incubation mixture by the oil-flotation method. The data show the intracellular distribution space of monosaccharides, which is the difference between the total sugar space and the extra cellular inulin space. The latter was between 30 and 40 µl/g cells. Each point and each short vertical bar show the mean value ± S.E. (n = 3). ○, basal uptake of 3-*O*-methyl-D-glucose; ●, plus-insulin uptake of 3-*O*-methyl-D-glucose; △, basal uptake of D-galactose; ▲, plus-insulin uptake of D-galactose; X, apparent utilization of D-galactose.

sugar is non-metabolizable, its uptake was ended at a certain equilibrium point, which was approximately 45 µl/g cells (Fig.1B). This figure was in agreement with the value of the urea space estimated in our previous work [5].

Insulin also stimulated the initial uptake of D-galactose (Fig.1A). The uptake of this sugar continued during the entire 60-min incubation period (Fig.1B). Apparently, the second phase of uptake, which probably corresponded to the utilization, was insulin insensitive. This observation was in agreement with the earlier data reported by Ball and Cooper [2] and Kuo and Dill [3].

The apparent rate of utilization of D-galactose estimated from Fig.1B is 3 µl/g per min. This value is less than 20% of the initial rate of the basal uptake (Fig.1A). We suggest, therefore, that the initial insulin-sensitive uptake represents, for the most part, the membrane transport of D-galactose.

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References

1 Kono, T. and Colowick, S.P. (1961) Arch. Biochem. Biophys. 93, 514—519
2 Ball, E.G. and Cooper, O. (1960) J. Biol. Chem. 235, 584—588.
3 Kuo, J.F. and Dill, I.K. (1969) Biochim. Biophys. Acta 177, 17—26
4 Gliemann, J., Østerlind, K., Vinten, J. and Gammeltoft, S. (1972) Biochim. Biophys. Acta 286, 1—9
5 Kono, T., Robinson, F.W., Sarver, J.A., Vega, F.V. and Pointer, R.H. (1977) J. Biol. Chem. 252, 2226—2233
6 Vinten, J., Gliemann, J. and Østerlind, K. (1976) J. Biol. Chem. 251, 794—800