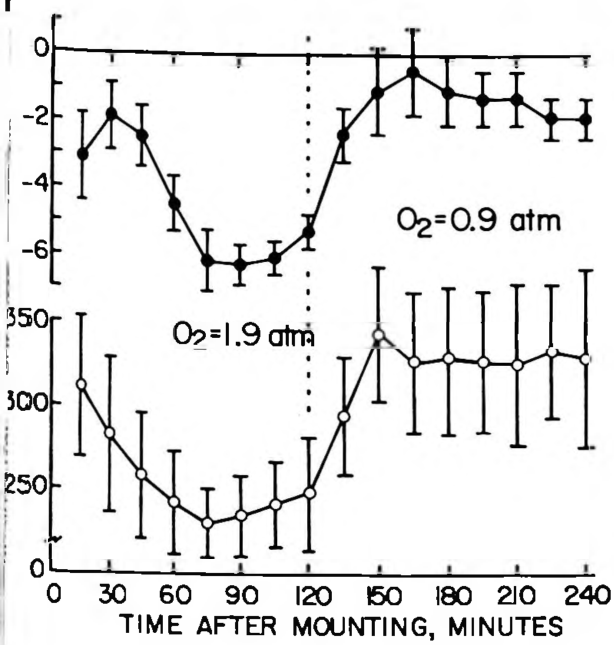
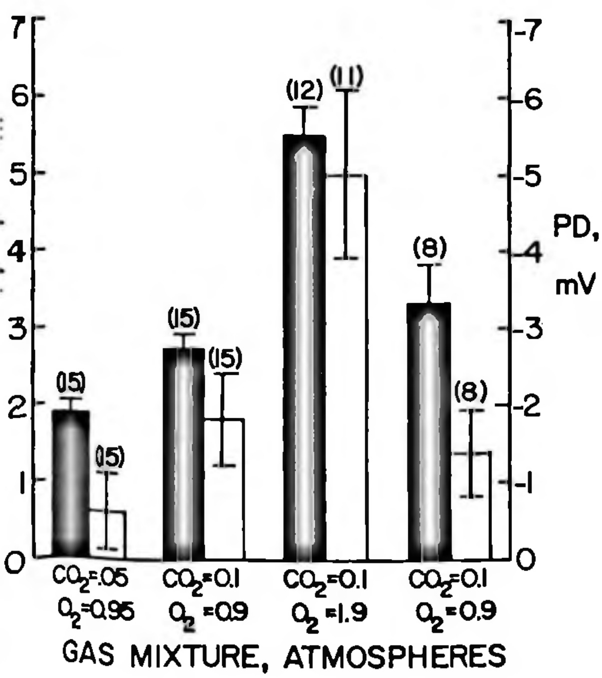
sed on the assumption that in high O_2 the only ions ively transported are H^+ and Cl^-. This is known to be e for the low O_2 condition. If additional ions were nsported in high O_2, they could account for the ntation of the PD. Resolution of these problems must ait further experimentation.

Supported by N.S.F. GB-36278





28 • 1975

The Specificity of the Transport System for D-glactose at the Basal Face of Renal Tubular Cells of the Flounder (*Pseudopleuronectes americanus*)

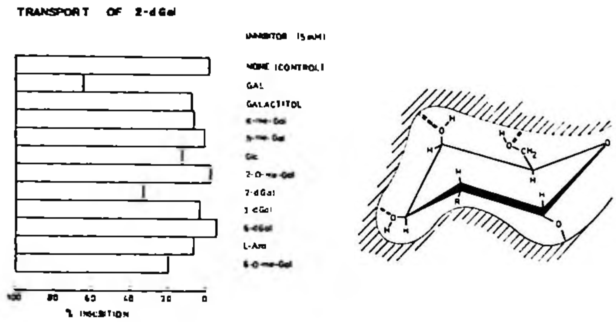
Arnost Kleinzeller, George R. Dubyak and James M. Mullin. University of Pennsylvania School of Medicine, Philadelphia, Pa. 19174.

The structural specificity of the transport of D-galactose and 2-deoxy-D-galactose (0.5 mM) by teased tubules of the winter flounder was investigated. The uptake of sugars by the tubule reflects, preponderantly, transport events at the antiluminal (basal) membrane of the tubular cells. Both sugars compete for the same transport sites, indicating the sharing of a carrier (*Kleinzeller & McAvoy, J. Gen. Physiol. 62, 169, 1973*).

Using previously described experimental techniques, the following structural requirements for the interaction between these hexoses and the carrier were defined on the basis of an inhibition analysis employing structurally analogous sugars (5 mM) 1. A pyranose ring structure is essential, for D-galactitol did not inhibit the cellular uptake of both sugars. 2. A free hydroxl on C_1 is required since both - and -methyl-D-galactosides (-me-Gal), and -methyl-2-deoxy-D-galactoside was not transported into the cells. 3. While 2-deoxy-D-galactose (2-dGal) strongly inhibited the transport of D-galactose (Gal), 2-0-methyl-D-galctose (2-0-me-Gal) was not inhibitory. These observations indicate that a free hydroxal on C_2 prevents the interaction of the sugar with the carrier, suggesting a rather close packing between both molecules in the vicinity of C_2. 4. 3-Deoxy-D-galactose (3-dGal) was not inhibitory, thus demonstrating a requirement for a C_3-OH (in the axial configuration) 5. D-Glucose (Glc) did not inhibit the transport of both sugars. Thus, a hydroxl on C_4 in the axial configuration is mandatory. 6. L-Arabinose (L-Ara) and 6-deoxy-D-galactose (6dGal) did not inhibit the transport of either hexose. On the other hand, 6-0-methyl-D-galactose (6-0-me-Gal) had a marked inhibitory effect. These observations show that an oxygen on C_6 is required for an interaction between galactose or 2-deoxy-galactose and the carrier.

Figure 1 shows the results of the inhibition analysis of galactose transport at the antiluminal face of the flounder renal cells. Qualitatively identical results have been obtained using 2-dGal as model sugar. The model given in Figure 1 expresses the possible interaction between the sugar molecule and the carrier. In analogy to observations made in rabbit (*Kleinzeller, Proc. VIth Internat. Congr. Nephrology, Florence, 1975*) and flounder renal tissue (*Kleinzeller, Dubyak and Mullin, Bull. MDIBL #29 this issue*), it is assumed here that hydrogen bonds between the oxygen atoms at C_3, C_4, C_6 and the ring oxygen are involved in such interaction. the possibility that at C_1-OH the sugar molecule interacts with a phosphoryl group at the carrier is discussed elsewhere (*Dubyak and Kleinzeller, Bull. MDIBL #11 this issue*). This investigation was supported in part by

USPHS Grant AM-12619. The authors gratefully acknowledge generous gifts of 3-deoxy-D-galactose (Dr. J.E.G. Barnett, Nottingham, England) and 2-D-methyl-D-galactose (Dr. J. Chittenden, Utrecht, Netherlands.).



29 • 1975

The Specificity of the Transport Systems for Glucose in the renal tubular cells of the Flounder (*Pseudopleuronectes americanus*)

Arnost Kleinzeller, George R. Dubyak and James F. Mullin, University of Pennsylvania School of Medicine, Philadelphia, Pa.

Data on the specificity of both transport pathways for D-glucose found at the antiluminal (basal) face of the renal tubular cells of the winter flounder (*pseudopleuronectes americanus*) have been presented previously (*Kleinzeller, and McAvoy, J. Gen. Physiol. 62:169, 1973; Kleinzeller, Rittmaster, Griffin and McAvoy, Bull., Mt. Desert Island Biological Lab. 14:60, 1974*). These studies have been extended using a broader selection of structural analogs of D-glucose. As in previous investigations, an inhibition analysis of model sugars by teased renal tubules served as the principal technique.

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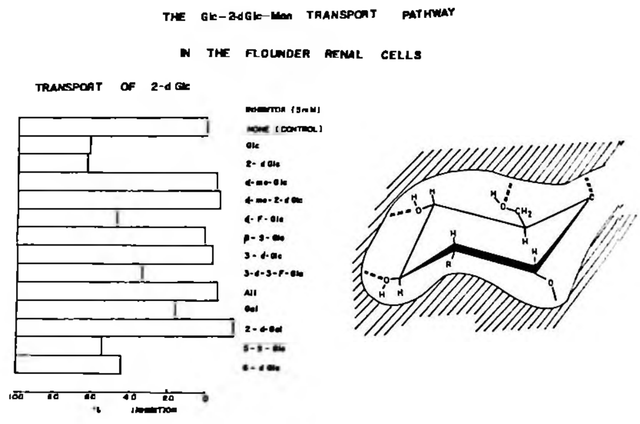
Using 0.5 mM methyl-x-D-glucoside-^{14}C as a model substrate for the transport system shared by D-glucose and both methyl-D-glucosides, the following newly tested sugars (5 mM) were found to be potent inhibitors: -Fluoro-D-glucose, 3-deoxy-3-fluoro-D-glucose. The sugars -thio-D-glucose and 3-deoxy-D-glucose had no effect. These data, taken in conjunction with previously recorded evidence, indicate that an interaction between the transported sugars and the carrier takes place at the following points:hydrogen bridges may be established between the oxygens at C_1, C_3 (in the axial configuration) and C_4 (in the equatorial configuration), a pyranose ring structure appears to be essential; finally, a bond of a firmer nature (covalent ?) may be formed at C_2-OH (in the equatorial configuration).

The specificity of the transport pathway shared by D-glucose, 2-deoxy-D-glucose and D-mannose was studied using the last two sugars as models. Figure 1 summarizes data obtained in previous and the present studies by an inhibition analysis of the tissue uptake of 2-deoxy-D-glucose. The inhibition pattern was identical with that for D-mannose and D-glucose (details not given here). Of the newly tested analogs of D-glucose, only -F-glucose, 3-deoxy-F-glucose, 5-thio-glucose and 6-deoxy-glucose were inhibitory. The scheme given in Figure 1 summarizes the possible points of interaction between the carrier and the transported substrates as follows:

A bond of a relatively firm nature (covalent link to a phosphoryl group at the carrier? See Dubyak, Mullin and Kleinzeller, Bull. Mt. Desert Island Biological Lab 15:000, 1975) is established at C_1-OH; fluoride can replace the hydroxl. Hydrogen bonding at C_3-OH in the axial configuration is indicated by the fact that 3-deoxy-D-glucose, D-allose and D-altrose were not inhibitory, whereas 3-deoxy-3-F-glucose was inhibitory. C_4-OH in the equatorial configuration is essential. A pyranose ring structure is mandatory but no hydrogen bond appears to be formed between the ring oxygen and the carrier in the light of the inhibitory effect of 5-thio-D-glucose.

This investigation was supported in part by USPHS Grant AM-12619.



30 • 1975

Configuration of the Skate (*Raja Erincea*) Nephron and Ultrastructure of Two Segments of the Proximal Tubule

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The micropuncture study of skate kidney (*Stolte et al., MDIBL Bulletin, #42, this issue*) presented evidence that the principal site of Mg, Phosphate and Sulphate secretion is the proximal tubular segment II (PTS II) which is located primarily on the ventral surface of the kidney. Since this is the first time that a specific site for secretion of these divalent ions has been localized, it was of interest to study the ultrastructure of this and other segments of the skate renal nephron. The nephron and