SHORT COMMUNICATIONS

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The effect of sugars and amino acids on mucosal Na^+ and K^+ concentrations in rabbit ileum

BROWN AND PARSONS[1] have reported that incubation of everted sacs of rat jejunum for 30 min in the presence of various sugars markedly affects the K^+ content of mucosal layers of this tissue. Their results indicate that the mucosal K^+ content is highest when the sacs were incubated in the presence of 27.5 mM D-glucose and lowest when they were incubated in the presence of actively transported sugars that either are not metabolized (3-*O*-methyl-D-glucose) or are poorly metabolized (D-galactose). Intermediate values were observed when the tissue was incubated in the absence of substrate or in the presence of L-sorbose, a sugar that is not subject to active transport by this preparation. Although these authors did not report the effects of these sugars on mucosal Na^+ content, it is probably safe to assume that a decrease in mucosal K^+ content was accompanied by an increase in Na^+ content and that their results reflect the ability of jejunal tissue to maintain normal intracellular Na^+ and K^+ concentrations under various metabolic conditions.

In recent years *in vitro* segments of rabbit ileum have been employed in the study of intestinal transport of various non-metabolized sugars and amino acids and many of these studies were carried out in the absence of added glucose or other substrates. Further, evidence has been presented that the rate of net transmural transport of sugars and amino acids across small intestine may be dependent, at least in part, upon the difference between the intraepithelial Na^+ concentration and the Na^+ concentration in the solution bathing the mucosal surface of the tissue[2,3]. In the light of this evidence, and the findings of BROWN AND PARSONS, it seemed important to evaluate the effect of various sugars and amino acids on the intracellular Na^+ and K^+ concentrations of mucosal strips of rabbit ileum. The conditions of this investigation were designed so that the results would be directly comparable with those of BROWN AND PARSONS.

Mucosal strips of rabbit ileum, which consist of the epithelial cell layer, the underlying lamina propria and a portion of the muscularis mucosa were prepared as described in detail previously[4]. The tissues were incubated for 30 min at 37.5° in an incubation medium that contained 140 mM NaCl, 10 mM $KHCO_3$, 1.2 mM K_2HPO_4, 0.2 mM KH_2PO_4, 1.2 mM $CaCl_2$, 1.2 mM $MgCl_2$ and 20 mM of the organic solutes listed in Table I. Stirring and oxygenation were accomplished by continuous bubbling with a pre-warmed, humidified O_2 and CO_2 gas mixture (95:5, v/v). The pH of the incubation medium was between 7.0 and 7.2. After 30 min the tissues were removed, blotted and extracted in 15 mM $LiSO_4$ solution for 48 h at 4°. The Na^+ and K^+ contents of the tissue were determined using an internal standard flame photometer (Instrumentation Laboratory, model 143). In some of the experiments, the extracellular space, evaluated using [^{3}H]inulin (New England Nuclear Corp.), and the dry

weight to wet weight ratio of the mucosal strips were determined as described previously[4]. In 33 experiments, the [^{3}H]inulin space averaged 0.33 ± 0.01 cm^3/g wet weight and the dry weight averaged 0.25 ± 0.01 g/g wet weight (all errors are expressed as S.E.). These values did not depend upon the particular organic solute included in the medium and were employed for the calculation of the intracellular water content of the mucosal strips. Intracellular concentrations of Na^+ and K^+ were calculated after correction for the contents of the inulin space assuming that the concentrations of Na^+ and K^+ in this space are equal to those in the incubation medium.

The calculated intracellular Na^+ and K^+ concentrations of mucosal strips incubated in the presence of 20 mM mannitol, D-glucose, 3-*O*-methyl-D-glucose, D-galactose, L-alanine and L-lysine are given inTable I. 3-*O*-Methyl-D-glucose[4], L-alanine[4] and L-lysine[5] are actively transported by rabbit ileum but are not subject to significant metabolism. Glucose is actively transported and metabolized[6], and mannitol is neither transported nor metabolized[4,7]. As shown in Table I, the intracellular Na^+ concentrations in the presence of these five solutes do not differ significantly. Further, the intracellular K^+ concentrations following incubation in the presence of alanine, lysine, glucose or 3-*O*-methylglucose do not differ significantly from each other but are significantly lower than that observed after incubation in the presence of mannitol. The explanation for this difference is presently unknown. The intracellular Na^+ and K^+ concentrations in the presence of alanine and 3-*O*-methylglucose are in good agreement with previously reported values obtained under somewhat different experimental conditions[4].

Galactose is actively transported by intestinal tissue and appears to be poorly or incompletely metabolized. Although we have not found any data dealing explicitly with galactose metabolism by rabbit ileum, evidence has been presented that this sugar is phosphorylated by rat jejunum and that the mucosal ATP content of galactose-fed rats is lower than that of rats not fed galactose[8]. As is shown in Table I, there is a significant increase in the intracellular Na^+ concentration and highly significant decrease in the intracellular K^+ concentration following 30-min incubation in the presence of 20 mM galactose. It is of interest that in the experiments reported by BROWN AND PARSONS, the lowest mucosal K^+ content was observed after 30-min incubation in the presence of 27.5 mM galactose, and was approximately one half that observed following incubation in the presence of 27.5 mM glucose.

TABLE I

INTRACELLULAR Na^+ AND K^+ CONCENTRATIONS AFTER 30-min INCUBATION IN THE PRESENCE OF VARIOUS ORGANIC SOLUTES

All errors are S.E.

Solute	*Number of mucosal strips*	*Intracellular concn. (mM)*	
		Na^+	K^+
Mannitol	51	58 ± 2	145 ± 3
Alanine	20	53 ± 4	131 ± 2
Lysine	20	58 ± 3	126 ± 4
Glucose	18	56 ± 4	124 ± 4
3-*O*-Methylglucose	16	59 ± 4	128 ± 4
Galactose	16	70 ± 3	104 ± 4

The data presented in Table I indicate that, in contrast with rat jejunum, rabbit ileum is capable of maintaining a low intracellular Na^+ concentration and a high intracellular K^+ concentration in the absence of substrate and in the presence of actively transported, non-metabolized sugars and amino acids. Further, the presence of glucose does not enhance this ability. Inasmuch as galactose is actively transported and may be phosphorylated at the expense of ATP, the effect of this sugar on intracellular Na^+ and K^+ concentrations may be attributable to a multiple demand on the limited energy supplies of the tissue.

These results and those of BROWN AND PARSONS are consistent with the reported metabolic differences between jejunal and ileal tissue. *In vitro* rat jejunum displays a high rate of glycolytic activity and a small Pasteur effect whereas ileal tissue possesses a much lower rate of glycolytic activity and displays a marked Pasteur effect[9,10]. Further, it appears that glycolysis is the obligatory energy source for ion and water transport across *in vitro* rat jejunum[10,11]. On the other hand, rat[10] and rabbit ileum[12] are capable of sustaining transmural ion and water transport in the absence of added substrate; the requisite energy is apparently derived from endogenous oxidative metabolism.

The present results and those of BROWN AND PARSONS suggest that rabbit ileum, and perhaps ileal tissue in general, is preferable to rat jejunum for the *in vitro* study of the transport of non-metabolized solutes in the absence of glucose. In addition, these investigations indicate that galactose is a poor choice for the study of the transport of non-metabolized sugars and that 3-*O*-methylglucose is to be preferred for this purpose. These conclusions are particularly salient if, as suggested by numerous investigations, the rate of sugar and amino acid transport is influenced by the difference between the intracellular Na^+ concentration and the Na^+ concentration in the mucosal medium.

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