ACTIVE TRANSPORT OF LYSINE, ORNITHINE, ARGININE AND CYSTINE BY THE INTESTINE

Hiroshi Hagihira, Edmund C. C. Lin, A. H. Samiy, and T. Hastings Wilson

Departments of Physiology and Biological Chemistry, Harvard Medical School, Boston, Massachusetts.

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The renal tubule possesses an active transport system for the reabsorption of the basic amino acids, L-lysine, L-ornithine and L-arginine (Beyer et al., 1947; Kamin and Handler, 1951; Brown et al., 1961; Webber et al., 1961). L-cystine appears to share the same absorption mechanism as suggested by studies of the human genetic defect, cystinuria (Robson and Rose, 1957; Dent and Rose, 1951). Individuals with this condition excrete in their urine abnormally large amounts of L-cystine plus the three basic amino acids, the absorption of the other amino acids being unaffected.

The question of whether a similar transport mechanism exists in the epithelial cells of the small intestine has not yet been satisfactorily answered. Previous *in vitro* studies of the intestine indicated that while L-cystine was actively transported against a concentration gradient (Neil, 1959), L-lysine and L-ornithine were not thus transported (Wiseman, 1955). On the other hand, L-lysine was absorbed more rapidly than its D-enantiomorph under *in vivo* conditions (Gibson and Wiseman, 1951). It was also shown that intravenous injection of pyridoxine stimulated L-lysine absorption in vitamin B_6 deficient rats (Akedo et al., 1960). These later results suggest that the absorption of at least one of the basic amino acids might be mediated by a special carrier.

An important recent observation was made by Milne, Asatoor and Loughbridge (1961) who reported that patients with cystinuria showed a

TABLE I

ACTIVE TRANSPORT OF "BASIC" AMINO ACIDS BY THE INTESTINE

Amino Acid	Initial Conc. mM	No. of Expts.	Final Conc. Serosal / Mucosal	Net Transport μmoles/100mg. tissue/hr.
L-Lysine-C^{14}	1.0	3	5.1	0.77
L-Arginine*	1.0	5	1.5	0.21
DL-Ornithine-C^{14}	1.0	3	2.7	0.51
"	2.0	5	2.0	0.62

Everted sacs of intestine were incubated for 1 hr. at 37° under 95% O_2 and 5% CO_2. Initial mucosal volume = 5.0 ml ; initial serosal volume = 1.0 ml.
*Rat instead of hamster was used as the intestine of the former species contained less arginase. Arginine was determined by the method of Sakaguchi (1925).

TABLE II

SEPARATE TRANSPORT FOR NEUTRAL AND "BASIC" AMINO ACIDS

Inhibitor	Conc. of Inhibitor mM	% Inhibition of Tissue Accumulation of Glycine 1 mM	L-Lysine 1 mM
L-Arginine	2.0	16	89
L-Cystine	0.8	0	45
L-Lysine	1.0	14	-
Glycine	1.0	-	0
L-Methionine	1.0	73	32

Hamster intestine was sectioned into 100-150 rings and thoroughly mixed in a beaker. An aliquot of 10-15 segments was weighed and incubated in 5 ml of Krebs-Henseleit solution according to the method of Agar et al. (1956). The test compounds glycine and L-lysine were labeled with C^{14}. Following 20 minutes incubation of 37° the segments were homogenized and the deproteinized filtrate was counted. Each value in the table represents the average of 3 experiments.

defect in the intestinal absorption of L-lysine and L-ornithine. From this observation it would appear that the epithelial cells of the intestine may

possess the same carrier system for the "basic" amino acids which is present in the proximal tubules of the kidney, and that the same mutation affects both systems. We were thus prompted to re-investigate the nature of the carrier system for the intestinal absorption of the four amino acids with experimental animals under in vitro conditions.

Everted sacs of hamster intestine (Wilson and Wiseman, 1954) were incubated in Krebs-Henseleit bicarbonate-saline containing a C^{14}-labeled amino acid on both sides of the intestinal wall. Table I shows the active transport of the three basic amino acids against concentration gradients. It might be noted that in all cases the transport was against an electrical gradient as the serosal side of the in vitro intestine is positive with respect to the mucosal side (Schachter and Britten, 1961). The maximal rates of transport for these amino acids were 1/10 to 1/20 the rates for the transport of some of the neutral amino acids, e.g. glycine, L-alanine and L-proline (Wiseman, 1955). The small capacity for the transport of the basic amino acids explains the difficulty in obtaining significant net transport with high initial concentrations (Wiseman, 1955).

Table II shows that L-cystine and L-arginine inhibited the transport of L-lysine much more than that of glycine. Conversely, L-methionine was a more effective inhibitor of glycine than of L-lysine. Previous studies have shown that the three basic amino acids have little or no inhibitory effect on the transport of the neutral amino acids L-histidine and mono-iodo-L-tyrosine (Wiseman, 1955; Agar et al., 1956; Nathans et al., 1960). In addition L-lysine did not inhibit the transport of L-isoleucine or L-methionine (Finch and Hird, 1960; Wiseman, 1955).

It is concluded that the intestinal epithelial cells possess a "basic" amino acid transport system similar to that found in the proximal tubules of the kidney.

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ERRATUM

In the paper "Steady State Equilibria of Some DPN-Linked Reactions and the Oxidation/Reduction State of the DPN/DPNH System in the Cytoplasmatic Compartment of Liver Cells in Vivo" published in Biochem. Biophys. Research Communs. 4, 159 (1961), the correct equations on page 159 are:

$$1)\ \{L/P\} : \{G/D\} = 1.6 \simeq K_{glyc.}/K_{lact.}$$

$$1a)\ \{M/O\} : \{G/D\} = 12.5 \simeq K_{glyc.}/K_{mal.}$$

In the paper "The Oxidation/Reduction State of the Extramitochondrial DPN/DPNH System in Rat Liver and the Hormonal Control of Substrate Levels in Vivo" published in Biochem. Biophys. Research Communs. 4, 163 (1961), the legend of Fig. 1 on page 165 may be completed by:

Levels in mμmoles/g fresh weight.