PRELIMINARY NOTE

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Anaerobic glucose stimulation of amino acid transfer in everted small intestine of the rat

It has been previously proposed that neutral amino acids are absorbed by two possible pathways across the epithelial cell of the rat small intestine[1]. Recently, additional evidence for the use of these two systems has been published for alanine isomers[2]. In an attempt to further characterize the two transport systems, hereafter called "methionine" and "sarcosine", we investigated the response to glucose of these carriers transporting L-, D- and β-alanine under various experimental conditions[3]. In order to obtain a more complete picture of their response to glucose, some experiments were carried out under anaerobic conditions. I report here results which show an unexpected but clearcut difference between these two systems with respect to glucose stimulation.

Experiments were carried out with 17-cm sacs of rat everted intestine as described previously[1]. The only modification consisted in the bubbling of a gaseous mixture of N_2–CO_2 (95:5, v/v) through the mucosal solution which was started 45 min before the experimental period and continued during the 30-min incubation period. The initial 1.0 ml serosal solution was similarly made anaerobic at the start. Experimental amino acid was present at a mucosal concentration of 1.0 mM. When present, glucose was 28 mM (mucosal and serosal), competitive amino acids were 10 mM (mucosal). All the tested substances were labelled with ^{14}C. The results are expressed as mean ± S.E. The number of experiments performed for each experimental condition is 4 or 6.

Water transfers have been measured under the various experimental conditions reported in Fig. 1 and Table I. Glucose has been found to increase very significantly ($P < 0.01$) the water transfer since the mean was raised from 0.17 ± 0.014 ml/sac per 30 min (number of experiments (N) = 29) in its absence to 0.52 ± 0.029 ml/sac per 30 min ($N = 35$) in its presence. Solvent drag seems to be of very little significance since, despite this relatively large increase in water transfer, thiourea is transferred to the same extent, whether glucose is present or not. Thiourea may be considered to cross the intestinal barrier by diffusion only[4] and is, therefore, a good control for the tested amino acids which are of similar molecular weight.

Fig. 1 shows the absence of any stimulation of transfer in the cases of thiourea and β-alanine. For D-alanine, the slight increase is just significant ($P < 0.05$). In contrast, the transfers of L-alanine and L-methionine are stimulated by about 100 %. Thus, the situation is just the opposite of that found in aerobiosis where L-alanine[2] and L-methionine[5] transfers are poorly increased by glucose, whereas D- and β-alanine ones are strongly stimulated[3,2].

The present results are at variance with those of SAUNDERS AND ISSELBACHER[6]

who found that anaerobiosis abolishes L-alanine active uptake in rat intestine, glucose being apparently unable to restore a higher level of accumulation; this information is not explicitly given, however. The reason for the difference between their results and those reported here might be that they did not use sacs but rings and from more distal parts of the intestine than chosen here. That active transport is likely to occur in my conditions is shown by the fact that tissue concentration of L-alanine in the presence of glucose is 2.4 ± 0.02 mM ($N = 4$) and that competition occurs for the transfer.

The effect of this competition is shown in Table I. One can see that the increase in L-alanine transfer brought about by glucose is to a great extent abolished by L-methionine. L-Proline results also confirm the picture previously proposed for amino acid absorption. L-Proline is known to use both systems[1]. It is, therefore, not surprising to find some stimulation of anaerobic transfer by glucose and a stronger competition from L-methionine than from sarcosine, since the part of the transfer stimulated by glucose is that going through the methionine carrier.

The present situation is different from that met in some instances in the literature where there is a stimulation of transport due to anaerobic conditions[7–10]. In my observations, anaerobiosis causes an inhibition of transfer which is partly overcome by glucose. Such a glucose effect is likely to exist in a tissue which is able to sustain a high glycolytic activity[11]. This could well be the case of rat intestine where a Pasteur effect has been reported[12], although some earlier investigators did not find any[13].

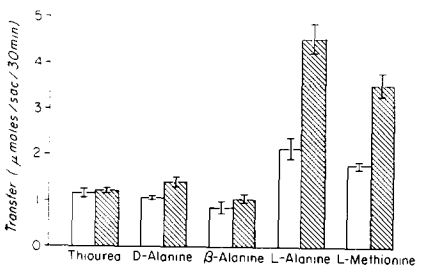

Fig. 1. Effect of glucose on solute transfer by anaerobic small intestine everted sacs of the rat. □, no glucose; ▨, 28 mM mucosal and serosal glucose.

TABLE I

EFFECT OF GLUCOSE ON THE ANAEROBIC TRANSFER, BY RAT SMALL INTESTINE, OF L-ALANINE AND L-PROLINE IN THE PRESENCE OF COMPETITORS

N = number of experiments.

Amino acid	*Glucose (28 mM)*	*Competitor (10 mM)*	*N*	*Transfer (μmoles/sac per 30 min)*
L-Alanine	−	None	4	2.14 ± 0.25
	+	None	4	4.56 ± 0.33
	−	L-Methionine	4	1.05 ± 0.03
	+		6	1.76 ± 0.19
L-Proline	−	L-Methionine	4	0.90 ± 0.07
	+		4	1.70 ± 0.19
	−	Sarcosine	4	1.44 ± 0.13
	+		4	2.62 ± 0.15

That glycolytic activity is not the only energy source for the methionine system is shown by the fact that L-alanine[3] or L-methionine[5] transfer is higher in aerobiosis than in anaerobiosis, even in the presence of glucose. The interpretation which seems best to fit the facts is, therefore, that the methionine system is able to draw energy from both the aerobic and the anaerobic metabolic pathways; thus, it is slightly stimulated by glucose in presence of oxygen and strongly in absence of oxygen. Such a participation of the two pathways in the transfer of L-alanine by frog intestine has already been presented[10] and could be more general than suspected. The sarcosine system obtains its energy from oxidative metabolism alone, being stimulated in aerobiosis but unaffected by glucose under anaerobic conditions where the transfer values obtained are those of diffusion. Whether the amino acid transport is a primary or a secondary event, the present observations are of some interest, especially if the same carrier systems are operating under aerobiosis and anaerobiosis.

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