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LETTER TO THE EDITOR

RAT EVERTED INTESTINAL SEGMENTS DEMONSTRATE THAT FASTING IS A REGULATOR OF UPTAKE AND METABOLISM OF GLUCOSE

Sir,

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Increased glucose production by the liver through gluconeogenesis is well documented. Does intestine also contribute to this process by provision of substrates for hepatic gluconeogenesis? There exists conflicting reports regarding such a role for the intestine. Mithieux et al suggest that fasting intestine contributes in two ways to maintain blood glucose level, directly by absorbing more glucose and indirectly by providing lactate and alanine (1). However, using a chronically catheterized rat model Kimura et al found that portal vein draining the gut did not show any increase in these substrates of gluconeogenesis (2). Habold et al found an increase in the content of gluconeogenic enzymes and glucose transporter level in the rat intestine after prolonged fasting (3). All these experiments were performed in vivo. Earlier, Gupta and his group observed a number of changes in the intestinal brush border in rats following fasting and these included changes in membrane fluidity along with structural and functional modifications (4,5), alterations in particulate density and lipid concentration (6, 7) and provided some evidence for altered transport of glucose(4). However they did not study lactate production, an important step in gluconeogenesis. To address this hiatus, we used everted intestinal sacs from fed and fasted rats to study glucose transport and lactate production.

Male, albino rats (n=18) from the central animal facility of Kasturba Medical College, Manipal have been used for the study. They were randomly allotted to fed (n=6), 4 day fasted (n=6) or 6 day fasted (n=6) groups. On the day of the experiment, they were sacrificed and the intestine was dissected out. Five centimeter segments of duodenum and jejunum were dissected out and were everted to prepare the sacs, each of which was filled with 2 ml of phosphate Ringer and placed in 10 ml of the same solution contained in a flask. After oxygenation the flasks were incubated for 30 minutes at 37° C. Duodenal segments were used for phosphate uptake and the jejunal segments for glucose uptake. The loss of substrate from the medium was taken as uptake by the sac while gain in lactate was taken as the release of the metabolite by the sac. All the values are expressed as micromoles of the substrate per gram protein. Protein of the sac was determined after homogenization by Lowry's method (8). Enzyme kits (Agappe Diagnostics, Ernakulam) were used for estimation of glucose and lactate.

TABLE I : Body weight (BW) and protein (P) content.

		Fasting groups	
	C (n=6)	*4 days (n=6)*	*6 days (n=6)*
BW before (g)	162 ± 10	146 ± 24	160 ± 38
BW after (g)	169 ± 13	134 ± 29	124 ± 48 *
% Change	4.3	8.3	22.5
Duodenal P (mg/5 cm segment)	253 ± 91	223 ± 109	232 ± 46
Jejunal P (mg/5 cm segment)	66 ± 23	42 ± 9**	35 ± 18**

All values are expressed as means ± SD of six observations in each group. Body weight is in grams while protein content is in milligrams. Unpaired Student't' test was used for statistical analysis. Value marked * is significantly ($P<0.05$) different from the value immediately above it. Values marked ** are significantly ($P<0.05$) different from the control value in the same row.

TABLE II : Uptake of phosphate and of glucose and release of lactate (ìmoles/g protein).

	C	*4 days*	*6 days*
Phosphate Uptake	740 ± 388	1103 ± 903	566 ± 77
Glucose Uptake	206 ± 55	373 ± 120*	428±196
Lactic acid	123 ± 30	210 ± 82 *	102 ± 82

All values are expressed as means ± SD of six observations in each group. Unpaired Students't' test was used for statistical analysis. Values marked *are significantly ($P<0.05$) different from the control.

As shown in Table I, neither the weight gain in fed animals nor the weight loss in 4 day fasted rats were significant. However, the rats fasted for 6 days showed significant loss in body weight. While the protein content of the duodenal segment from the three groups did not differ, the jejunal segments from the fasted rats contained significantly less protein when compared to similar segments of fed rats. Table II indicates that phosphate uptake by the everted duodenal sacs did show any difference between the fasted and fed groups. Glucose uptake and lactate release were significantly elevated in jejunal segments of 4 day fasted rats when compared to similar segments of fed rats. These parameters did not show any change in jejunal segments of 6 day fasted rats.

The results obtained in our experiments clearly indicate that glucose uptake by the everted jejunal sacs of 4 day fasted rats was significantly increased as reported earlier by Gupta and Waheed (4). Lactic acid output was also increased in these segments of the intestine indicating that the glycolytic pathway of metabolism was enhanced. In an intact gut such enhancement in lactate supply may facilitate hepatic gluconeogenesis. Thus our results favour the view that intestine may help in gluconeogenesis that occurs during fasting, by providing higher amounts of lactate. However in 6 day fasted rats such increase in either glucose uptake or lactate release were absent. These results are in contrast to observations of Habold et al, who reported no rise at 4 day but significant change only in rats fasted for more than 6 days (3). In our experiments, while body weights decreased in fasted rats they were significant only at 6 day fasting. Such discrepancy might have resulted from the wide range in the weights of the rats at the beginning of the present set of experiment. But the jejunal weights of all fasted animals decreased significantly when compared to those from the non-fasting controls. This observation indicates the possibility that protein breakdown in the gut occurs earlier than what Habold et al have inferred (3). It also does not appear to interfere with transport of glucose indicating that transporter proteins are spared during this protein breakdown initially. The increases in glucose uptake and lactate release noticed at 4 day fasting disappears at 6 days of fasting. Probably the protein breakdown has begun to affect the proteins involved in transport and metabolism of glucose. Interestingly in our experiments the protein content of duodenum remained unchanged throughout the fasting period and the uptake of phosphate in this segment also remained unchanged. Thinning of the gut during fasting facilitates absorption of nutrients which increase by one and half times (4). These changes may have a role in promotion of survival mechanisms during short term fasting.

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