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Absorption of Lactic Acid from an Isolated Intestinal Segment in the Intact Rat (32882)

MARSHALL D. HELLER AND FRED KERN, JR.

Division of Gastroenterology, Department of Medicine, University of Colorado Medical Center, Denver, Colorado 80220

In certain disorders of digestion and absorption dietary carbohydrates are poorly absorbed and are degraded by intestinal bacteria. Some of the resultant products, principally organic acids, including lactic acid are excreted in the feces. These acids, because of their osmotic properties, have been implicated in the production of symptoms of disaccharide malabsorption (1–3).

Little is known about lactic acid absorption *in vivo*. This study was undertaken to determine the site of lactic acid absorption and the mechanism of its absorption, with special reference to the effect of intraluminal pH.

Materials and Methods. Male albino rats, 150–350 gm (Holtzman) were fasted 16–20 hours, anesthetized with ether, and through a midline incision a 20-cm loop of small intestine or colon was isolated. A glass cannula was sutured in place at each end of the segment. After a 20–ml saline wash, a test solution was perfused at room temperature by means of a pump at the rate of 5 ml for 15 min for 2 hours as described by Biggs and Davis(4). The perfusate consisted of various concentrations of sodium lactate with 1.0 μC of sodium lactate-1-^{14}C (4.5 mC/mmole) (New England Nuclear Corp.) adjusted to the desired pH with HCl and to approximately

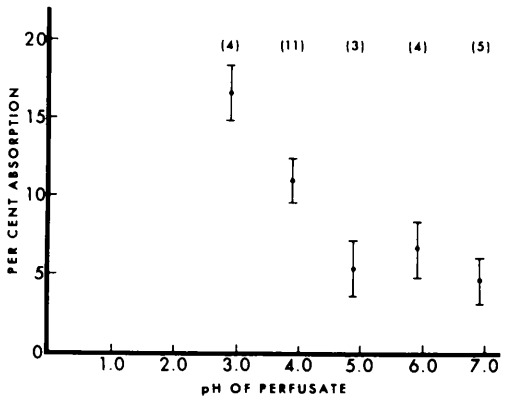

FIG. 1. Absorption 0.11 *M* sodium lactate-1-^{14}C from rat jejunum at various pH's. In this and subsequent illustrations the numbers in parentheses indicate the number of animals. All figures show the mean ± SE.

290 milliosmoles with NaCl. The radio-purity of the labeled sodium lactate was 90–95% as determined by paper chromatography (5).

The aspirate was collected from the distal end of the segment by gravity in 15–min aliquots in tubes immersed in iced water. The volume of each 15-min aliquot was measured; 0.5 ml was added to 10 ml of Bray's solution (6) and the radioactivity was assayed in a liquid scintillation spectrometer. The segment was rinsed with saline at the end of the study and the rinse contained negligible radioactivity.

The amount of lactic acid absorbed was calculated by subtracting the radioactivity collected from the radioactivity administered. In two experiments the intestinal perfusion was performed with the rats in a metabolism chamber (7). Expired air was bubbled through phenethylamine to trap the $^{14}CO_2$ and its radioactivity was determined in the scintillation spectrometer (8). In two experiments a metabolic inhibitor, 2,4-dinitrophenol was administered subcutaneously (2 mg/100 gm of body wt.) 15–30 min prior to the perfusion. Segments of intestine and colon were taken before and after perfusion and fixed for light and electron microscopy.

Results. Lactate absorption was clearly related to pH. It was maximum at pH 2.8, decreased linearly to pH 4.8 and remained constant from pH 4.8 to pH 6.8 (Fig. 1). The pH of the aspirate was 0.3–0.6 of a unit higher than the pH of the perfusate. The volume of the aspirate was within 3.2% of the volume of the perfusate. Absorption was greatest in the proximal jejunum; it was less in the ileum and was least in the colon (Fig. 2). After equilibration, the amount of lactate absorbed in each 15-min period remained fairly constant throughout the experiment (Fig. 3). Figure 3 also shows the cumulative absorption of lactate.

The 2,4-dinitrophenol (2 mg/100 gm of body wt.) was injected subcutaneously into two rats 15 min prior to jejunal lactate perfusion (pH 2.8). Each rat absorbed 24% of the lactate, slightly more than the control animals. In these poisoned rats the amount absorbed in each 15-min period was constant, as in the control. Both of these rats died at the conclusion of the experiment. When different concentrations of lactate were perfused, absorption was directly related to concentration. A saturation phenomenon characteristic of active transport kinetics did not exist within the range of concentrations employed (Fig. 4).

To prove absorption as the cause of the disappearance of ^{14}C lactate from the intestinal lumen expired $^{14}CO_2$ was collected during jejunal perfusions in two rats. Thirty-

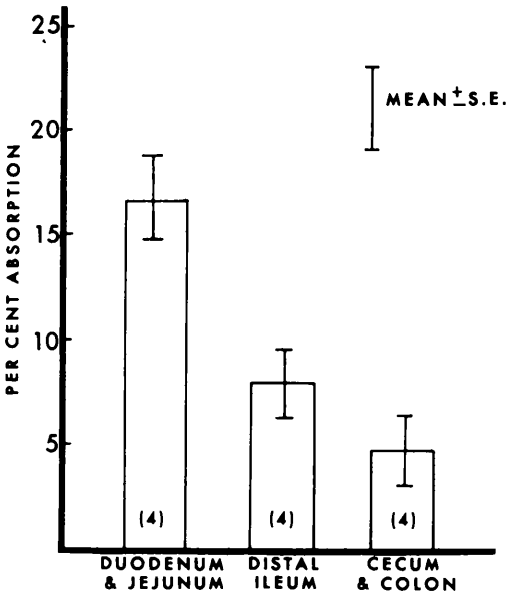

FIG. 2. Absorption of 0.11 *M* sodium-1-^{14}C lacate from different regions of rat intestine, pH 2.8.

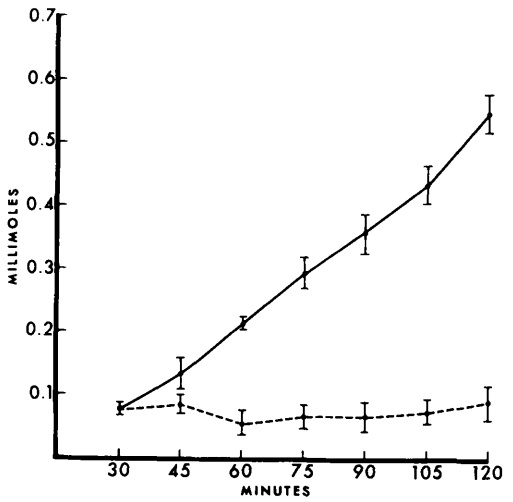

FIG. 3. Cumulative absorption of 0.11 *M* sodium lactate-1-^{14}C by rat jejunum; - - -, rate of absorption in each 15-min period, pH 3.8 (n = 11). The perfusion rate was 0.55 mmoles/15 min.

two and 42% of the labeled lactate that disappeared from the lumen was recovered as $^{14}CO_2$ (Fig. 5). The mucosa of perfused and control segments of small intestine and colon was normal by light and electron microscopy.

Discussion. Only a few investigators have studied lactate absorption. In 1928 Cori and Cori found that 40–57% of sodium lactate fed to rats disappeared from the intestinal tract in 3 hours. Sodium lactate was absorbed much more slowly than either glucose or fructose (9). In 1954, Wilson used everted intestinal sacs to study glucose metabolism and absorption and found that about one-quarter of the absorbed glucose was converted into lactic acid. The total amount of lactate found on the serosal side was 4–10 times that on the mucosal side. Added lactate, however, was not transported against a concentration gradient (10). In 1955, Newly *et al.* found that when lactic acid was added to an *in vitro* intestinal preparation in the absence of glucose, the rate of lactic acid transfer from the mucosal side to the serosal side was greater than in the reverse direction. When the initial concentration of lactic acid was the same on both sides, there was a fall in concentration on the mucosal side, but no corresponding increase on the serosal side. These experiments failed to demonstrate transfer of lactic acid against a gradient (11).

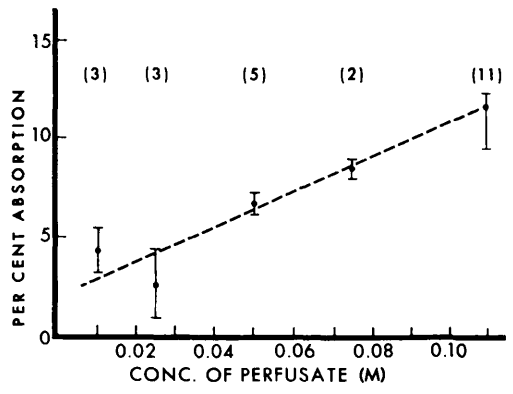

FIG. 4. Absorption of sodium lactate-1-^{14}C from rat jejunum at pH 3.8 at various concentrations. - - - Values were calculated by the method of least squares.

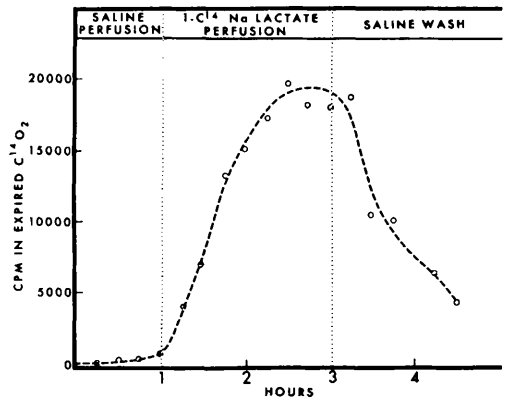

FIG. 5. $^{14}CO_2$ in expired air during perfusion of rat jejunum with 1.0% sodium lactate-1-^{14}C at pH 3.8 (n = 2).

The *in vivo* studies reported in this paper suggest that lactic acid is absorbed by passive diffusion. This is supported by the absence of saturation kinetics, the effect of pH and the lack of an effect of the metabolic inhibitor, 2,4-DNP.

The absorption of lactate was pH dependent as is characteristic of the absorption of weak acids and bases and certain drugs (12–14). Lactate absorption increased as the [H^+] was increased and was maximal at pH 2.8, the pH near its pK(3.08). The unionized species (at low pH) readily penetrate the lipid cell membrane of the intestinal epithelium, whereas the polar ionized form (at higher pH) does not. In the following equilibrium,

$H^+ + Lactate^- \rightleftarrows$ Lactic acid,

as the $[H^+]$ is increased, the equilibrium is shifted to the right favoring the unionized form. At physiologic pH (6–7) the ionized species diffuses more slowly. The poor absorption of lactic acid at the pH of intestinal content is consistent with the conclusion that the osmotic activity of the short chain fatty acids contribute to the watery diarrhea in patients with sugar malabsorption.

It has been suggested that acid solutions damage the mucosa of the small intestine. Several investigators have described villus atrophy, inflammation of the lamina propria, and damaged surface epithelial cells in patients with low intraluminal pH due to Zollinger-Ellison syndrome, gastrojejunostomy, or pancreatic insufficiency (15–17). There were no morphologic changes in the intestine of rats after acid perfusion for 2 hours.

Summary. Lactic acid absorption was studied in anesthetized rats by perfusing isolated segments of small intestine or colon with sodium lactate-1-^{14}C for 2 hours. The concentration and pH of the perfusate were varied. Absorption was maximum at pH 2.8, near the p*K* of lactate, and minimum at physiologic pH. Absorption was directly related to concentration of the lactate perfused and it was not affected by a metabolic inhibitor. The jejunum was the site of maximum absorption. Absorbed lactate was metabolized to $^{14}CO_2$. Poor lactate absorption at physiologic pH and in the distal intestine might lead to an osmotic diarrhea in certain patients with malabsorption.

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Comparative Studies on Pathogenesis of Two Strains of Transplantable Carcinomas of Rabbits, Vx2 and Vx7* (32883)

YOHEI ITO, IKUO KIMURA, AND TAKASHI MIYAKE
Laboratory of Viral Oncology, Research Institute Aichi Cancer Center, Nagoya, Japan

Cutaneous papillomas of rabbits induced by Shope papilloma virus (SPV)(1) frequently progress to carcinomas(2). From these malignancies, Rous and his associates have been able to establish several transplantable strains of carcinomas, the so-called Vx series, and among them Vx2(3) and Vx7(4) have been serially transferred for over 20 years to date.

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