Disaccharidase Deficiency Associated with the Intestinal Phase of Trichinosis in Guinea Pigs[1] (36455)

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Relatively few investigations have dealt with the pathogenesis of the intestinal phase of trichinosis. This is especially true in human infection which is usually diagnosed only after the disease has progressed into the extraintestinal stage. In guinea pigs administered graded doses of *Trichinella spiralis* larvae the gut phase is accompanied by diarrhea and weight loss, the intensity of which is directly dependent on the size of the worm burden (1).

Interestingly, the small intestine of animals with these symptoms is characterized by histopathology similar to that reported for humans with malabsorption syndromes, and the capacity of the gut to transport glucose actively is greatly reduced (2). Since the transport of glucose is mediated through the brush border of the gut epithelium, it is probable that other physiologic functions associated with this region, including the activity of digestive enzymes such as disaccharidases (3) are adversely affected and may contribute to the clinical picture.

Presently, the physiologic basis of diarrhea in trichinosis is not well understood, although diarrhea as a clinical entity may be caused by various intestinal malfunctions, including deranged absorption and impaired digestive processes (4). Despite the general assumption that the failure of parasitized animals to gain weight at normal rates is related to anorexia, weight loss cannot be attributed entirely to this factor (1). To establish the possibility that helminth-induced diarrhea and weight loss are related, in part, to impaired digestive function, the activity of maltase, sucrase, trehalase, and palatinase in the small intestine of control guinea pigs was compared to that of animals infected with *Trichinella*.

Materials and Methods. The strain of *Trichinella* used was maintained by passage through CF-1 mice (male, 20 to 30 g) infected by oral intubation of viable larvae. Larvae used to infect guinea pigs (Hartley strain, males, 225–300 g) were recovered by pepsin digestion of skeletal muscle from mice infected 4 months previously (5). Treated groups of guinea pigs were given a dose of 10^4 larvae according to a previously described regimen (1).

Disaccharidase activity was studied in mucosal scrapings from isolated segments of small intestine from control and infected guinea pigs. The small intestine was perfused with Krebs–Ringer bicarbonate buffer (pH 7.4), removed from the guinea pig, and divided into quarters according to published methods (2). The duodenal and ileal ends of the gut were designated as the first and fourth quarters, respectively. A 10-cm segment was cut from the distal end of the first and second quarters and everted over a glass rod. The mucosal surface was scraped from the intestine with a glass slide and the scrapings were homogenized in a prechilled Potter–Elvehjem homogenizer in 10 ml of cold 0.85% NaCl. Quantitative determinations of disaccharidase activity were made on the homogenate according to the procedure described by Dahlqvist (6). The glucose oxidase used (160 units/mg; Worthington Biochemical Corp.) was more active than that mentioned in Dahlqvist's methods, so only 0.5 mg/100 ml Tris–glucose oxidase reagent

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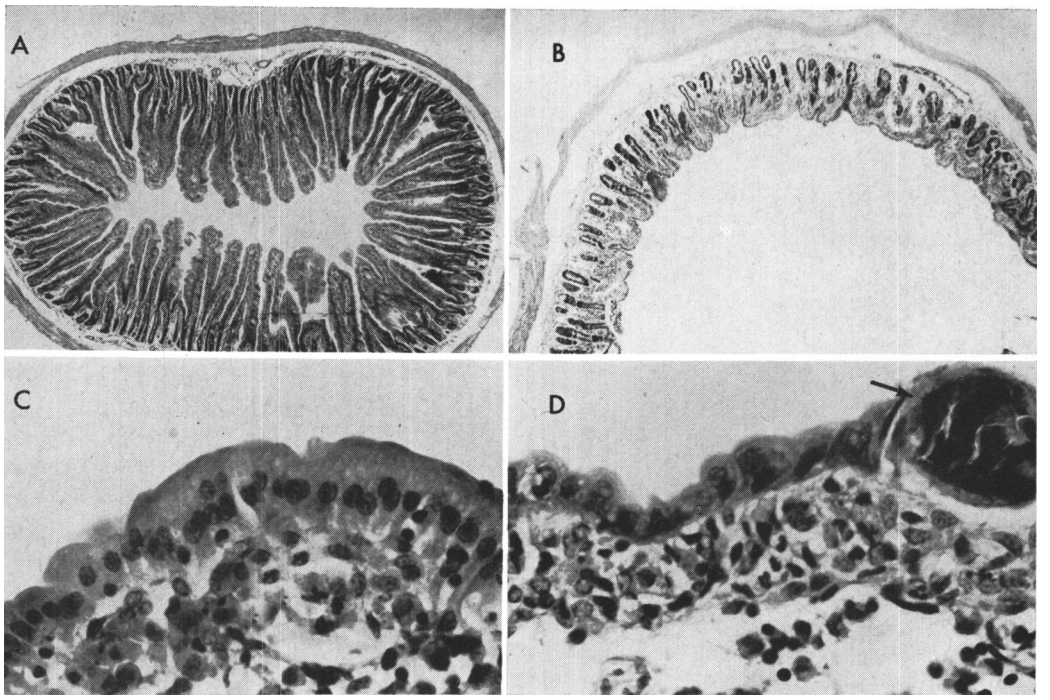

FIG. 1. Histology of the jejunum from uninfected guinea pigs and guinea pigs infected with 10^4 *Trichinella* larvae. (A) Villous pattern of uninfected gut, ×18 (B) Villous pattern of infected gut, ×18. Note the distention. (C) Villous surface of uninfected gut showing columnar epithelial cells, ×325. (D) Gut surface of infected gut showing abnormal epithelial cell morphology, ×325. Note section through parasite at end of arrow. [Photomicrographs and an interpretation of the histopathology shown in this figure have been published in detail elsewhere (2)].

was used instead of the prescribed 2 mg/100 ml. The concentration of all substrates was 28 m*M*. Disaccharidase activity was expressed as mμmoles of substrates hydrolyzed per minute per milligram of protein. Protein concentration was determined by the method of Lowry *et al.* (7).

Maltose, sucrose, trehalose, and palatinose (6-*O*-alpha-D-glucopyranosyl-D-fructose) were obtained from Nutritional Biochemical Co. Samples of all substrates equal to the amount in enzyme assays were checked for glucose contamination by thin-layer chromatography (8).

Data were subjected to analysis by Student's *t* test (9) and differences between means were considered significant when *p* values were less than 0.05.

Results. Clinical signs, including diarrhea and decreased weight gains, and gut histopathology associated with trichinosis in the guinea pig during a 3- to 28-day postinfection period have been described (1, 2). Histologically the small intestine of infected guinea pigs is characterized by a significant loss of the normal villous processes, abnormal morphologic alterations in epithelial cells, and inflammation of the lamina propria (Fig. 1).

In the present study guinea pigs were killed either 4 or 5 days postinfection, and all infected animals showed varying degrees of diarrhea. Disaccharidase activities in the intestines of these animals are shown in Table I and compared to values from uninfected controls. All disaccharide substrates used were hydrolyzed by guinea pig mucosal homogenates. The relative activity and spatial distribution of maltase, sucrase, and trehalase in control animals agrees with data reported by other workers. Malhotra and Philip (10) demonstrated that maltase activity in the guinea pig intestine was about three times

TABLE I. Intestinal Disaccharidase Activity in Control and *Trichinella*-Infected Guinea Pigs.[a]

Enzyme activity	Substrate hydrolyzed mμmoles/min/mg protein[b]	
	Control	Infected[c]
	First quarter	
Maltase	41.8 ± 1.4	3.7 ± 3.1
Sucrase	24.2 ± 1.7	1.1 ± 0.4
Trehalase	22.0 ± 4.4	0.5 ± 0.2
Palatinase	6.8 ± 1.0	0.2 ± 0.1
	Second quarter	
Maltase	43.8 ± 2.1	3.9 ± 3.4
Sucrase	23.0 ± 2.7	1.0 ± 0.5
Trehalase	22.2 ± 2.0	0.5 ± 0.2
Palatinase	6.8 ± 1.1	0.2 ± 0.1

[a] Concentration of all substrates was 28 m*M*.

[b] Specific activities for all enzymes are mean values ± the standard error for assays on four animals.

[c] Values for infected animals were significantly lower than corresponding control values in all comparisons ($p < .05$).

higher than that of sucrase and trehalase, and the distribution of the three enzymes was essentially the same throughout the jejunum. In our study maltase was found to be approximately twice as active as sucrase and trehalase, and the activities of respective enzymes in segments from the first and second quarter of intestine were not significantly different (Table I). Dahlqvist *et al.* (11) pointed out that hydrolysis of palatinose in the human and hog intestines is catalyzed by isomaltase. Palatinase activity was the lowest of the four enzymes assayed in the guinea pig.

The specific activities for all disaccharidases in the infected animals were significantly reduced as compared to those of uninfected controls. The reduction in enzyme activity from both quarters of intestine was similar and varied from an approximate 10-fold decrease in the case of maltase to a 35-fold decrease for palatinase.

Discussion. This study demonstrates that disaccharidase deficiency is a component in the pathogenesis of infection and provides greater insight into the physiologic etiology of symptoms attending trichinosis specifically and other intestinal helminthiases in general.

Diarrhea commonly accompanies intestinal nematode infections, yet the nature and cause of this condition have not been examined experimentally (12). Weight loss in hosts infected with various helminths is well documented. Most workers attribute this to reduced appetite caused by infection. Our results support the contention that disaccharidase deficiency along with malabsorption, which was reported previously (2), contribute to diarrhea. Further, impaired digestion or malabsorption alone or in conjunction with diarrhea would disrupt assimilation of nutrients by the host which would be reflected by weight loss. Such an assumption regarding the underlying cause of these symptoms which characterize various helminth infections is supported by the following reports.

Two of the most common signs accompanying malabsorption in man are diarrhea and weight loss (4). Diarrhea can be induced in humans by the oral administration of poorly absorbed sugars such as xylose and arabinose (13). Guinea pigs administered 10,000 *Trichinella* larvae show a significant reduction in the ability to transport glucose as measured by both *in vitro* and *in vivo* techniques (2). Impaired absorption in these animals is accompanied by diarrhea and weight loss. Similar findings occur in trichinized mice (14). Rats infected with *Trichinella* develop diarrhea (15) and are unable to assimilate calci-

um normally during a 4- to 8-day postinfection period (16).

Rats experimentally infected with *Nippostrongylus brasiliensis* lose weight and are often diarrheic (17). Heavily parasitized regions of the small intestine of these animals show a decreased capacity to digest protein and maltose (18, 19). Spedding (20) reported that a decrease in the ability of sheep infected with *Trichostrongylus axei* to digest protein may account, in part, for the weight loss observed in these animals. It is well known that diarrhea and weight loss are associated with disaccharide intolerance in humans and can be caused by an absence or deficiency of a single hydrolytic enzyme at the level of the intestinal brush border (21, 22). A similar basis may explain the diarrhea observed in parasitized guinea pigs. Our data revealed that the activity of intestinal disaccharidases in diarrheic guinea pigs with trichinosis is reduced 91 to 97% from control values.

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