

splenectomized group, there was no correlation between properdin level and age of individual, age at splenectomy, and years post-splenectomy. Properdin titers were significantly lower in splenectomized females, but the sex difference was minimal in comparison to the difference between total splenectomized and control groups. Suggestive evidence is presented that low properdin levels in splenectomized persons are related in part to absence of spleen.

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Trypsin, Invertase and Amylase Content of Feces of Germfree Rats.* (25174)

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As measured with specific methods the trypsin and chymotrypsin concentration of intestinal content decreases with length of the intestine, decreasing to zero in feces(1). This fact indicates that intestinal enzymes are autodigested or are inactivated by intestinal microorganisms. To elucidate this problem the enzyme content in feces of germfree and conventional animals was measured.

Material and methods. Germfree rats reared according to Gustafsson(2-3) were used in these studies. Two germfree male adult rats of the 4th generation were fed the semisynthetic diet D₆(4) and kept individually in metabolism cages. Feces and urine were collected each 24 hours. Two conventional rats were fed the same autoclaved diet and kept in metabolism cages in the animal room. The feces were collected for 8 consecutive days. On the 8th day the germfree rats were infected with cecal content of a conventional animal heated to 90°C for 5 min. Thereafter the feces were collected for an-

other 12 days during which time sporeforming organisms were cultivated from the feces. Then one of the animals was killed for assessment of the effect of the sporeformers on the cecum enlargement. The remaining germfree animal was on the same day infected in the apparatus with a suspension of feces from the conventional rats. The feces were homogenized with 10 ml ice cold saline and centrifuged. Aliquots of the clear supernatant solution were used for enzyme determinations. Trypsin was determined spectrophotometrically using benzoyl-arginine ethyl ester as substrate(5). Invertase activity was assayed according to a modification of the method of Sumner(6), and expressed as mg of invert sugar produced in one hour by 1 ml of the sample at 25°C. Amylase activity was determined by the method of Meyer *et al.*(7) and expressed as mg of maltose liberated in 3 minutes by 1 ml of the sample at 25°C.

Results. The feces of the conventional animals did not contain measurable amounts of trypsin or invertase but held 40-100 units of amylase (Fig. 1). On the other hand, feces of the germfree rats contained 1-6 mg trypsin daily and 12-25 units of invertase per day.

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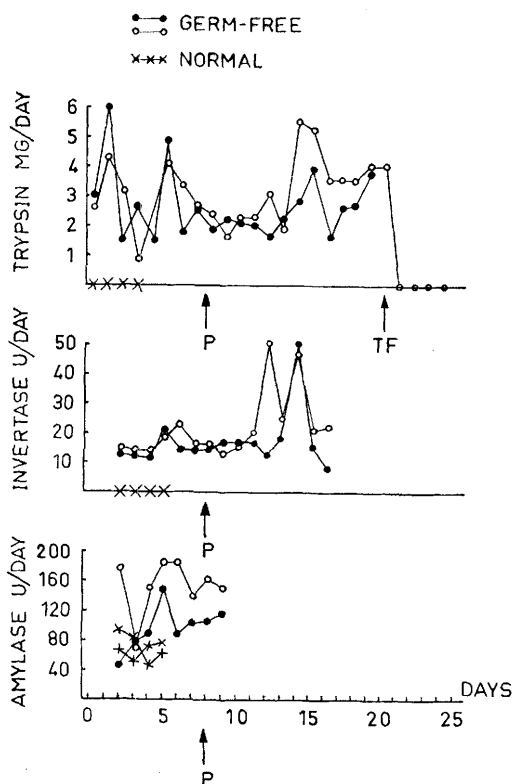


FIG. 1. Trypsin, invertase and amylase content of feces of germfree, ex-germfree and conventional rats. At P germfree animals were infected with sporeforming bacteria from the cecum of a conventional animal and at TF with suspension of feces from conventional rats.

Amylase content of the feces in the germfree rats was the same or slightly higher than that of the conventional rats. The occurrence of invertase in the feces of germfree animals agrees with the finding of carbohydrases in intestinal extracts from germfree rats(8).

After infection of the germfree animals with sporeforming bacteria from the cecum content of a conventional animal, no decrease occurred in the enzymatic contents of the feces but rather sharp fluctuations were noticed in invertase and trypsin activity. This might be connected with the diarrhea that is a constant symptom in infection of germfree animals with certain intestinal bacteria. After superinfection with full intestinal flora of the conventional animals, trypsin content of the feces decreased to the zero values of the conventional rats within 24 hours.

Conclusions. Results indicate that autodigestion is not the main pathway for normally occurring inactivation of trypsin and invertase. The results infer that one or several of the normal microbiological inhabitants of the intestines are responsible for normally occurring inactivation of the digestive enzymes of intestinal contents.

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Effect of Dietary Fat on Excretion and Deposition of C¹⁴ from Cholesterol-4-C¹⁴ in Rats.* (25175)

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man and lower mammals for many years. Recently it has become important to ascertain

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