

sufficient to elicit antibody formation although not sufficient to give precipitate formation in the present experimental circumstances. As another possibility, the antibodies against the growth hormone itself might be suspected of also reacting with certain normal serum constituents. Absorption of one immune serum against the growth hormone preparation with a normal human serum by means of intrabasin gel-absorption⁹ did not, however, absorb the antibodies against the growth hormone preparation.

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Increase of Esterase Activity during Intracellular Digestion in a Histophagous Ciliate

THE macrophages of vertebrates and invertebrates¹ as well as some Protozoa² exhibit strong esterase and lipase activity demonstrable by histochemical and biochemical methods. Considering the prominent position of the hydrolytic enzymes in digestive processes, it seems probable that they have also some significance in the intracellular breakdown of particles, a common characteristic of both cell-types in question. In the course of our recent work on the problems of feeding and digestion in a histophagous ciliate, *Tetrahymena corlissi* Thompson, strain W, we obtained some results that may confirm this assumption.

Cells of *T. corlissi*, grown axenically on a tryptone (1 per cent) and yeast extract (0.05 per cent) broth, were washed twice in Prescott solution and kept during the experiments in the same solution. Fresh frozen sections of rat spleen served as food. The animals readily ingested the spleen cells. Smears of unfed and fed (for 1 hr.) animals, and those of animals starved (2-24 hr.) in Prescott solution after feeding, were fixed in cold 10 per cent formalin (1 hr.). The 'Tween' procedure of Gomori³ and the azo-dye procedure of Nachlas and Seligman⁴ were used for the demonstration of esterases. The slides were mounted in glycerine-jelly.

Unfed animals exhibited a slight or moderate activity on the substrates α -naphthyl acetate, 'Tween 60' and '80'. Parallel with the formation of food vacuoles a great increase in esterase activity has been observed. Using α -naphthyl acetate, which gives a distinctly localized reaction product, the food vacuoles could be demonstrated as the site of the increased activity (Fig. 1). The 'Tween' procedure revealed only a general increase in the activity, but did not show its intracellular localization. In animals

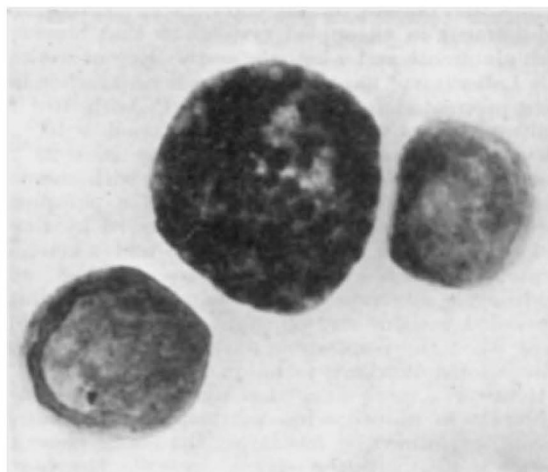


Fig. 1. One fed and two unfed cells of *T. corlissi*. The food vacuoles of the fed individual exhibit a strong esterase activity. Azo-dye method

starved after feeding there is a drop in the activity to the initial level within 24 hr. The digestion is already completed at this time and the animals contain only empty vacuoles as revealed by the control slides stained with hæmatoxylin.

These changes are probably due to an activation of the enzymes and may indicate an intimate correlation of the esterases with the intracellular digestion. Our recent findings do not enable us to determine the type of the enzymes responsible for the increase observed. The splitting of both substrates, α -naphthyl acetate and the unsaturated 'Tween 80', seems to indicate that at least a non-specific esterase and a lipase may be at work. Further studies aiming at a more detailed characterization of the enzymes are in progress.

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A High-Energy Phosphate Requirement for Succinate Oxidation in Uncoupled Rat Liver Mitochondria

It has been reported by Borst and Slater^{1,2} that rat liver mitochondria, depleted of endogenous phosphate by pre-incubation with hexokinase and glucose, showed a marked phosphate requirement for the oxidation of glutamate in the presence of 2,4-dinitrophenol. This phosphate requirement was attributed to the substrate-level phosphorylation linked to the oxidation of α -ketoglutarate which is not uncoupled by dinitrophenol. Evidence has been given that other Krebs-cycle metabolites, such as succinate,