suspended in 0.32M-sucrose at a concentration of 1-2mg. of protein/ml., were treated at 0° for 1hr. with 0.5% Triton X-100 and M-KCl, pH 6.5 or 7.5. After centrifugation at 80000g for 20min., almost all of the activity was in the supernatant and was 170% of that originally present. The enzyme remained very soluble after the Triton had been almost completely removed by ammonium sulphate fractionation and dialysis, but the enzymic activity fell to values that approximated closely to the activity originally present in the untreated mitochondria. Triton measurements showed that a residual amount of Triton (approx. 1mg./mg. of protein) remained with the protein in the preparation. Attempts to retain the increased activity by repeating the fractionation procedure in media containing constituents expected to provide stabilizing conditions were unsuccessful. In each case the increased activity in the Triton extract was lost on removal of the Triton. The extracts, notwithstanding the loss of 'Triton-elevated' activity, proved to be quite stable on storage for weeks at -20°. The addition of Triton to the Triton-depleted extracts resulted in a slight but variable reactivation of about 15%, considerably less than the activation in the original Triton extract. The results are relevant to suggestions of part-latency of brain mitochondrial hexokinase.

M.F.T. is the holder of a Medical Research Council Studentship for training in Research Methods. The work was supported by the Research Fund of The Bethlem Royal Hospital and the Maudsley Hospital.

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Peptide Hydrolase Activity of *Klebsiella aerogenes*

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During extensive investigations (Barker, Pardoe, Stacey & Hopton, 1963; Barker, Somers, Stacey & Hopton, 1965; Barker, Hopton, Somers & Repas, 1966) into the ability of *Klebsiella aerogenes* (N.C.I.B. 9479) to synthesize inducible glycoside hydrolases it has become apparent that the micro-organism is able to utilize a variety of peptides as sole source of nitrogen. The present report describes an investigation of the evident peptide hydrolase activity of the micro-organism.

Four cultures of *K. aerogenes* were grown in minimal media (Barker *et al.* 1966) differing only in

the identity of the single nitrogen source. The four nitrogen sources were ammonium sulphate, glycine, glycyglycine and leucylglycine. Towards the end of the exponential phase the cells from each culture were harvested, washed and suspended in cold 0.02M-phosphate buffer, pH 7.0, and disrupted by shaking with glass beads for 45 min. Cell debris was removed by centrifugation and, after exhaustive dialysis of the extracts, peptide hydrolase activity was determined at pH 7.0 and 37° against glycyglycine and leucylglycine. Identical peptide hydrolase activity was exhibited by all four extracts. The cell-free culture fluids exhibited no significant activity. Intracellular constitutive activity was thus indicated.

An extract from ammonium sulphate-grown cells was chromatographed on DEAE-Sephadex equilibrated with 0.02M-phosphate buffer, pH 7.0. Peptide hydrolase activity was eluted between 280mm- and 300mm-NaCl. The active fractions were concentrated by ultrafiltration and re-chromatographed on Sephadex G-100 equilibrated with 0.02M-phosphate buffer, pH 7.0, containing 1.0M-NaCl. Peptide hydrolase activity was eluted as a single, included, peak. The hydrolase exhibited maximum activity at pH 7.2.

In addition to glycyglycine and leucylglycine the enzyme hydrolysed glycy-leucine, phenylalanyl-glycine, glycyphenylalanine, seryl-glycine, glycy-serine, lysyl-glycine, glycy-lysine, triglycine, tetra-glycine, leucylglycyglycine and glycyglycine ethyl ester. Peptides not hydrolysed included prolyl-glycine, glycyproline and the benzyloxycarbonyl derivatives of glycyglycine, triglycine and tetra-glycine. The ability of the enzyme to hydrolyse glycy peptides and glycyglycine ethyl ester but not the benzyloxycarbonyl derivatives of glycyglycine, triglycine and tetraglycine suggests aminopeptidase activity.

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A Soluble Aerobic Reduced Nicotinamide-Adenine Dinucleotide (Phosphate) Azoreductase

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When the lipid-soluble fungicide 3-methyl-4-(2-chlorophenylhydrazono)-isoxazol-5-one (otherwise known as drazoxolon) was incubated