

of seaweeds will be further investigated, and a detailed report will be published elsewhere.

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Occurrence of D-Acylase in Soil Bacteria

IN work reported earlier¹, it was shown that a strain of *Pseudomonas* sp., *KT83*, can hydrolyse N-benzoyl derivatives of both L- and D-forms of amino-acids such as phenylalanine, tyrosine and alanine.

In a further experiment² carried out with the *KT83* acetone powder, it was found that the N-dichloroacetylated D-isomers of threo- β -phenylserine, threo- β -p-nitrophenylserine and phenylglycine were hydrolysed more easily than the corresponding N-dichloroacetylated L-isomers, suggesting the possible occurrence of D-acylase in this bacterial species. The present communication is concerned with evidence for the occurrence in Nature of D-acylase, which hydrolysed only the D-forms, but not the L-isomers of N-acylated amino-acids.

KT83 was grown in 1 litre of bouillon at 25° C. for two days, and the cells were then harvested by centrifugation and washed with distilled water. The yield of cells, in wet weight, was approximately 14 gm. The cells, after having been ground with alumina, were extracted with 40 ml. of distilled water. Crude extract thus obtained was dialysed against cold distilled water for 15 hr. To 40 ml. of the dialysed crude extract was added 8 ml. of a 1 per cent solution of protamine sulphate, and the precipitate resulting was removed by centrifugation. To the clear supernatant fluid was added 24 ml. of a 1 per cent solution of protamine sulphate. The white precipitate, which contained most of the activity of L-acylase, was separated by centrifuging, and dissolved in 1.5 M sodium chloride solution and dialysed first against 1.5 M sodium chloride solution for 30 hr., and secondly against distilled water for 20 hr. (partially purified L-acylase sample).

At the same time, the clear, straw-coloured supernatant, which contained most of the activity of D-acylase, was fractionated by addition of increasing amounts of saturated ammonium sulphate. The fraction which was precipitated between 50 and 60 per cent saturation of ammonium sulphate and which would be most active was dialysed against distilled water for 10 hr. until free of ammonium sulphate. The inside fluid was then centrifuged to remove inactive protein which was precipitated during the dialysis (partially purified D-acylase sample).

The enzymatic assay was performed according to Grassmann and Heyde's method. Protein was

determined by the phenol method of Lowry *et al.* The acylase-activities of crude extract, partially purified L-acylase and partially purified D-acylase, are shown in Table 1.

On the other hand, the resolution experiment, in which 6 ml. (15 mgm. of protein) of the partially purified D-acylase sample was made to act on 1.35 gm. of benzoyl-DL-phenylalanine at 37° C. for 20 hr., gave the following results: 0.33 gm. (80 per cent) of D-phenylalanine $[\alpha]_D^{25} = +35.0^\circ$ ($c = 2$, H₂O); 0.57 gm. (84 per cent) of benzoyl-L-phenylalanine $[\alpha]_D^{25} = +14.9^\circ$ ($c = 3$, NNaOH) m.p. 139–40°; 0.25 gm. (82 per cent) of benzoic acid m.p. 118–20°.

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¹ Kameda, Y., Toyoura, E., Yamazoe, H., Kimura, Y., and Yasuda, Y., *Nature*, **170**, 888 (1952).

² Kameda, Y., Toyoura, E., Kimura, Y., and Matsui, K., *Yakugaku Zasshi*, **78**, 202 (1958).

Colonization of *Anopheles funestus*

ATTEMPTS to colonize *A. funestus funestus* Giles, the second most important vector of malaria in Africa, commenced in 1954 and continued for three years. During 1955–56 the first successful colony of this mosquito was established from eggs laid by wild-caught females of *A. funestus*, given alternative feeds on human and guinea pig blood. The egg production of this colony was of short duration and small quantity, and the colony eventually died out after a few months.

In August 1957 another colony was established from wild-caught, gravid females maintained on guinea pig blood meals only. This colony, now in its eighth generation, is remarkable for its vigour and egg production. The eggs are laid on filter paper immersed in a dish filled with water; the larvae are maintained in bowls filled with tap water with floating water weed *Elodea canadensis*. Larval food consists of pure dried yeast. Adults are kept in cages 1 cu.ft. capacity in total darkness; anaesthetized guinea pigs are used daily as a source of blood meals for female mosquitoes; males are fed on sugar solution freely available in the cage. The duration of the cycle from eggs to pupae is about 21 days at the temperature 80–85° F. Two egg batches of this colony were sent by air to London where a subsidiary colony was successfully established. Studies on the taxonomy and bionomics of this colony are now in progress.

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Table 1

| Enzyme sample | Specific activity | | Total activity | |
|------------------------------|-----------------------|-------|-----------------------|-------|
| | Benzoyl-phenylalanine | | Benzoyl-phenylalanine | |
| | D- | L- | D- | L- |
| Crude extract | 2.3 | 11.5 | 1,035 | 5,175 |
| Partially purified L-acylase | 1.0 | 23.0 | 120 | 2,760 |
| Partially purified D-acylase | 32.0 | <0.03 | 520 | <0.5 |

Expressed as micromoles hydrolysed per hr. per mgm. protein. The digests consisted of 0.5 ml. enzyme solution, 0.5 ml. of water and 1 ml. of 0.05 M neutralized substrate (pH 8) at 37° C.

The General Relationship between Test Factors and Person Factors; Application to Preference Matrices

THE application of factor analysis to correlations between persons had already become widespread among psychologists when it was made the subject of a special study by Burt and Stephenson shortly before the Second World War. They began in collaboration, but concluded by differing¹ in their