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Purification of a Streptomyces Chitinase. By C. Jeuniaux\* (introduced by M. Florkin). (Laboratoires de Biochimie, Université de Liège, Belgium)

Different chitinolytic streptomycetes, grown on culture media containing the salts of the Czapek solution with pure chitin as sole source of carbon and nitrogen, produce an exo-chitinase. We succeeded in preparing very active chitinolytic filtrates, using a Streptomyces sp., isolated from soil (strain A1) (Jeuniaux, 1955). We purified the chitinase of this filtrate by acidification, 'mass' adsorption on powdered chitin, followed by washing and enzymic hydrolysis of the chitin, and two consecutive fractionations by ammonium sulphate at 0° and pH 5·2 (Jeuniaux, 1956).

The chitinolytic activity has been concentrated 70 times by this procedure, with a yield of 27 %. The proteolytic and cellulolytic activities, as well as the pigments and the reducing sugars of the crude filtrate are completely removed.

The total proteins and the chitinase of the final solution have the same solubility properties in ethanol solutions, and in concentrated ammonium

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sulphate solutions. Ultracentrifugation (59780 rev. min.) indicates that the final solution is homogeneous; the molecular weight of the chitinase is about 30000.

By electrophoresis in agar-agar plates (Gordon, Keil, Sebesta, Knessl & Šorm, 1950; Uriel & Grabar, 1956), the proteins of the final solution migrate to the cathode at pH 8·2, as three components. A quantitative analysis of the different fractions suggests the presence of three chitinases, having the same specific activity per mg. of protein. When recombined, these different chitinases show a synergistic effect on chitin hydrolysis. There is some evidence that the concentration of non-chitinolytic proteins in the final solution does not exceed 5 %.

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