A LIPASE FROM THE FUNGUS Rhizopus microsporus

STRAIN UZLT-1

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The fungus <u>Rhizopus microsporus</u> was grown in 250-ml Erlenmeyer flasks in a medium of the following composition (%): maize extract 2, cottonseed oil 1, CaCO₃ 1.5.

The present paper gives the results of a study of the lipolytic enzymes (hydrolysis of glycerol esters, E.C. 3.1.1.3) in the culture liquid and in an isopropanol precipitate from the fungus <u>Rhizopus microsporus</u>, strain UzLT-1.

The lipase activity was determined by the method of Ota and Yamada [1] and was expressed in milliliters of 0.1 N alkali solution consumed in the titration of the fatty acids formed from 1 ml of culture liquid under the action of the lipase. The protein was measured by the biuret method, and also by the Warburg-Christian method [2]. An isopropanol powder was obtained by saturating the culture liquid with isopropanol $(1:6 \text{ at } +2^{\circ}\text{C})$. An extract was obtained by triturating 0.1 g of the isopropanol powder in 10 ml of distilled water or buffer. The extract was centrifuged at $15,000 \times \text{g}$ at 2°C for 15 min. The supernatant liquid was investigated for its lipase activity (Table 1).

When the lipase activity was measured in the pH range from 3 to 11, two pH maxima were found. In the case of a 0.1 M phosphate-citrate buffer, the maximum activity appeared at pH 3.5-4.8 and 6-8, and in the case of a glycine buffer at 4-4.5 and 7.8-8.5. Gel filtration on Sephadex G-75 in 0.1 M phosphate-citrate buffer (pH 8.0) showed the presence of two protein components possessing lipase activity (see Fig. 1).

Process	Amount of pro- tein, mg	Activity		Yield (in %) on the		
		spe- cific	total	pro- tein	activ - ity	-
Culture liquid Precipitation with isopropanol (1:6)	8680	565,14	4905068	100	100	
	3741	1106,8	4140538,8	43,8	84,5	

TABLE 1



Fig. 1. Gel filtration on Sephadex G-75 of the protein fraction obtained by precipitation with isopropanol.

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