Purification and Properties of Alkaline Phosphatase with Protein Phosphatase Activity from Saccharomyces cerevisiae

Danka Galabova*, Borijana Tuleva, Evgenia Vasileva-Tonkova, and Nelly Christova

Department of Microbial Biochemistry, Institute of Microbiology, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., Bl 26, 1113 Sofia, Bulgaria.

Fax: +359 2 700 109. E-mail: dgal@microbio.bas.bg; dgal@bas.bg

* Author for correspondence and reprint requests

Z. Naturforsch. **55 c**, 588–593 (2000); received May 25, 1999/January 21, 2000

Alkaline Phosphatase, Protein Phosphatase Activity, *Saccharomyces cerevisiae*An alkaline phosphatase (ALPase) from *Saccharomyces cerevisiae* strain 257 was purified 345-fold with specific activity of 54 533 nmol \times min⁻¹ \times mg protein⁻¹. It was shown to be a dimeric protein (apparent mol. wt. approx. 130 kDa) with optimum activity at pH 8.6–8.8 and good stability at 50 °C. The ALPase was a non-specific enzyme hydrolyzing a wide variety of monophosphate esters. The enzyme showed protein phosphatase activity and this activity was not Mg²⁺ – dependent in contrast to *p*-nitrophenyl phosphate (*p*NPP) activity. The $K_{\rm m}$ value for pNNP hydrolysis was determined to be 2.2×10^{-5} M. Orthophosphate inhibited the enzyme in a competitive mode with the $K_{\rm i}$ of 2.3×10^{-4} M. Phosphate transfer of the

ALPase is almost zero with all alcohols tested except for Tris.