COLORIMETRIC ASSAY OF GLYCEROL KINASE FROM BAKER'S YEAST

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Glycerol kinase (EC 2.7.1.30; ATP: glycerol 3-phosphotransferase) catalyzes the Mg, ATP-dependent phosphorylation of glycerol to glycerol 3phosphate, which is an intermediate compound of energy production and phospholipid biosynthetic pathways. The enzyme is present in several organisms, from bacteria to human. The present study describes a methodology of dosage of glycerol kinase from baker's yeast proceeding from Mauri Brasil Ind. e Com. Imp. Ltda. The crude cellular extract was prepared as described by Tininis, Master's degree dissertation, Unesp. 2001, and the standardization of the activity of glycerol kinase from baker's yeast was accomplished as described by Huang et al. (*J. Ferment. Bioeng.* 83: 328, 1997). The following reaction mixture was used: glycerol in solution 0.4 M glicine buffer at pH 10.0, 4aminoantipyrine, phenol, ATP, magnesium sulphate, horseradish peroxidase and the diluted enzymatic preparation containing glycerol phosphate oxidase and glycerol kinase. The mixture was incubated at 60°C by 15 minutes and the reaction was stopped by the addition of SDS solution. The pH and temperature stability showed that the enzyme was stable to pH 7.0 - 8.0, and its activity was completely maintained up to 50°C during 1 h. This low cost method doses glycerol kinase in a sequence of reactions and represents great importance to many industries, like food, sugar and alcohol.

Keywords: Colorimetric assay, Glycerol kinase, Baker's yeast

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