Thymidine kinase from *Tetrahymena thermophila*

Purification and immunological analysis

Peter W. KINYANJUI and Ronald E. PEARLMAN

Department of Biology, York University, North York, Ontario, Canada

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Abstract

Thymidine kinase is an enzyme involved in DNA precursor metabolism and DNA replication. The synthesis of this enzyme is highly regulated during the cell cycle and the activity of the enzyme is also regulated by feedback inhibition. Genes encoding thymidine kinase have been extremely useful as selectable markers for introducing DNA into a number of cells. In order to study cell cycle regulation of thymidine kinase, the gene which encodes this enzyme, as well as aspects of DNA replication in the ciliated protozoan *Tetrahymena thermophila*, we have purified thymidine kinase from *Tetrahymena*. Two forms of thymidine kinase with native molecular masses of 59 kDa and 80 kDa have been identified and purified 6800- and 4600-fold, respectively. The 59-kDa enzyme, a homodimer of 30-kDa subunits, has been purified to near homogeneity and polyclonal antibodies have been raised against the 30-kDa subunit. Serological studies indicate that the two enzymes are antigenically distinct. The antibody against the *Tetrahymena* protein cross-reacts with a polypeptide in Chinese hamster ovary (CHO) cell extracts of 26 kDa which corresponds to the reported size of Chinese hamster thymidine kinase protein.