

Cloning and comparison of genes for dTTP synthesizing enzymes from *Geobacillus* bacteria.

Takanori Satoh^{1,2,3}, Takamasa Kageji¹, Ayaka Matsunaga¹, Kazuya Ebara¹, Ken Kurihara¹, Chinami Matsumoto¹, Akihiro Marumo², Masami Toyoshima², and Yousuke Endoh^{1,2}

¹Faculty of Integrated Arts and Sciences, The University of Tokushima, Japan, ²Graduated School of Integrated Arts and Sciences, The University of Tokushima, Japan, ³Institute of Socio-Arts and Sciences, The University of Tokushima, Japan.

Thermophiles have been utilized as the bioresource of useful industrial and medical biomacromolecules. In order to utilize moderately thermophile *Geobacillus* genus bacteria as genetic resource for many useful and thermostable enzymes, we attempted to obtain the genes for dTTP synthesizing enzymes such as Thymidine kinase (TK), Thymidylate synthetase (ThyA), Thymidylate kinase (TMPK), and Nucleoside diphosphate kinase (NDK) from four *Geobacillus* bacteria, *Geobacillus stearothermophilus* ATCC7953 (*Bst*), *Geobacillus kaustrophilus* NBRC102445 (*Gka2*), *Geobacillus thermocatenulatus* NBRC15316 (*Gtc*), and *G.zalihae* NBRC101842 (*Gza*). By PCR on the basis of genomic information of *Geobacillus kaustrophilus* HTA426 (*Gka*), we have cloned 16 genes totally, and analyzed above four kinds of genes from four *Geobacillus* bacteria. It was suggested that the deduced amino acid sequences for all four enzymes (TK:207AA, ThyA:264AA, TMPK:214AA, NDK:148AA) are highly conserved (more than 90%) among them from other *Geobacillus* species, whereas their homology to the corresponding *Bacillus* enzymes are almost 60-70%. On the other hand, it revealed that homology of amino acid sequences among four enzymes might be very low (less than 20%), though they might recognize similar substrate (dT, dUMP, dTMP, dTDP). We will discuss the similarity and differences in amino acid sequences and deduced functional sites of these dTTP synthesizing enzymes.