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32-2298: DUT Recombinant Protein

Alternative Name: Thermostable dUTPase, dUTPase.

Description

Source: Escherichia Coli. Thermostable dUTPase (pyrococcus fruriosus) maximizes the efficiency of high-fidelity PCR (using proofreading DNA polymerases). It removes contaminating dUTP present in PCR reactions and dNTP solutions. The presence of dUTPase in a proofreading DNA polymerase reaction can prevent dUTP misincorporation by maintaining dUTP levels below their inhibitory concentrations despite the constant generation of the molecule by the spontaneous deamination of dCTP. The incorporation of dUTP into PCR products causes mutations within the amplified product, proofreading polymerases to stall and slows down non-proofreading polymerases such as Taq. The dUTPase increase in PCR product yield, length and fidelity enables further down-stream applications. These effects make dUTPase useful in PCR fidelity and yield-sensitive applications such as cloning and subsequent recombinant protein technology, and gene expression analysis (semi-quantitative RT-PCR techniques and real-time PCR analysis), where small differences in product accumulation can have a significant impact on gene expression analysis.

Product Info

Content:

Amount: 1000 IU

Purification: Greater than 97.0% as determined by SDS-PAGE.

dUTPase is specific for dUTP and is critical for the fidelity of DNA replication and repair. dUTPase

hydrolyzes dUTP to dUMP and pyrophosphate, simultaneously reducing dUTP levels and

providing the dUMP for dTTP biosynthesis.

Store at 4°C if entire vial will be used within 2-4 weeks. Store, frozen at -20°C for longer periods

Storage condition: of time. For long term storage it is recommended to add a carrier protein (0.1% HSA or

BSA). Avoid multiple freeze-thaw cycles.

Application Note

(A) Measured by its ability to hydrolyze dUTP to dUMP in reaction buffer 20mM Hepes pH7.5, 150mM KCI, 5mM MgCl2, 10mM dUTP at 85 Centigrade for 1 hour. (B) Enhancing PCR amplification: 50ul of Pfu PCR reaction system with 1-3u of dUTPase (<3.0kb) or 4-6u dUTPase (>3.0kb) to amplify genomic DNA target up to 15-19 kb in length. (High concentrations of dUTPase will inhibit PCR reaction!).

