

# ENZYMES

BY

# SORACHIM

## Glutamate Dehydrogenase (NADP-dependent) from Proteus sp.

### GTD-209

#### SPECIFICATIONS

Product name	L-Glutamate:NADP <sup>+</sup> oxidoreductase (deaminating)
EC	1.4.1.4
Appearance	50mM Tris buffer solution at pH 7.8 with 0.05% NaN <sub>3</sub> and 5mM EDTA
Activity	> 9000 U/ml
Contaminants	NADPH oxidase ≤ 0.01 %, Gluthathione reductase ≤ 0.01 %
Stabilizer	EDTA
Stability	Stable at 2-8 °C for at least 6 months
Molecular weight	Approx. 300,000
Isoelectric point	4.6
Michaelis constants	1.1×10 <sup>-3</sup> M (NH <sub>3</sub> ), 3.4×10 <sup>-4</sup> M (α-Ketoglutarate), 1.5×10 <sup>-5</sup> M (NADP <sup>+</sup> ) 1.2×10 <sup>-3</sup> M (L-Glutamate), 1.4×10 <sup>-5</sup> M (NADPH)
Structure	6 subunits per mol of enzyme
Inhibitors	Heavy metals, PCMB, Pyridine, 4-4'-dithiopyridine, 2-2'-dithiopyridine
Optimum pH	8.5 (α-KG→L-Glu), 9.8 (L-Glu→α-KG)
Optimum temperature	45 °C (α-KG→L-Glu), 45-55 °C (L-Glu→α-KG)
pH stability	6.0 – 8.5 (25 °C, 20hr)
Thermal stability	Below 50 °C (pH 7.4, 10min)

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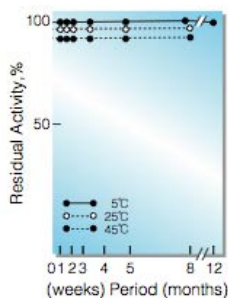


Fig.1. Stability (Solution)

[50% glycerol solution in 25mM Tris-HCl buffer contg. 2.5mM EDTA, pH7.8 enzyme concentration: 5,000U/ml]

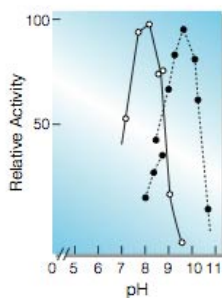


Fig.3. pH-Activity

[○—○,  $\alpha$ -KG  $\rightarrow$  L-Glu; ●—●, L-Glu  $\rightarrow$   $\alpha$ -KG] in 0.1M buffer solution: pH7.4-8.8, [Tris-HCl; pH8.7-10.7, glycine-NaOH]

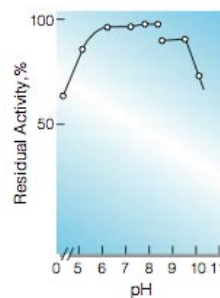


Fig.5. pH-Stability

[25°C, 20hr-treatment with 0.1M buffer solution: pH4.4-6.2, acetate; pH6.2-8.4, phosphate; pH8.8-10.2, glycine-NaOH]

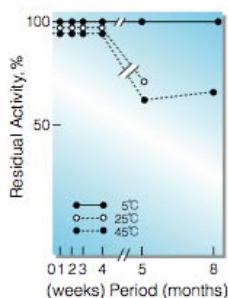


Fig.2. Stability (Suspension)

[3.0M ammonium sulfate suspension in 50mM Tris-HCl buffer containing 5mM EDTA, pH7.8 enzyme concentration: 10,000U/ml]

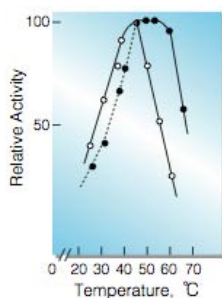


Fig.4. Temperature activity

[○—○,  $\alpha$ -KG  $\rightarrow$  L-Glu; 0.1M Tris-HCl buffer, pH8.3; ●—●, L-Glu  $\rightarrow$   $\alpha$ -KG; 0.1M glycine-NaOH buffer, pH10.0]

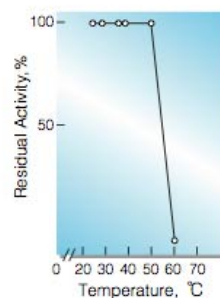


Fig.6. Thermal stability

[10min-treatment with 0.1M K-phosphate buffer, pH7.4]

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