rGlDH(NAD)

recombinant Glutamate dehydrogenase (NAD^+) EC 1.4.1.2

from Bacteria

Reaction Equation

α-Ketoglutarate + NH_3 + NADH + H^+

⇄ L-Glutamate + H_2O + NAD^+

Specification

Specific Activity

U/mg protein	> 350 units

(for reduction of α-Ketoglutarate to L-Glutamate)

Contaminants

NADH oxidase	< 0.01%
Lactate dehydrogenase	< 0.003%
Malate dehydrogenase	< 0.003%
Alcohol dehydrogenase	< 0.003%

Properties

pH stability	: pH 7.5 (37°C 1week)
Thermal stability	: ≦ 60°C (pH 7.8, 10 min)
Optimum pH	: 7.4 - 7.8
Optimum temp.	: 45°C
Km value	: 3.0 × 10^{-2} mol/L (α-Ketoglutarate)
	1.7 × 10^{-4} mol/L (NADH)
	2.0 × 10^{-2} mol/L (Ammonium chloride)
	1.6 × 10^{-2} mol/L (L-Glutamate)
	1.5 × 10^{-4} mol/L (NAD^+)
Molecular weight	: 48 kDa (SDS-PAGE)

Assay Procedure

I Spectrophotometric Method

Wavelength : 340 nm, Light path length : 1 cm

Final volume : 3.02 mL, Temperature : 30°C

Pipette the following reagents into a cuvette

2.60 mL	Tris-HCl buffer (0.1 mol/L, pH 7.8)
0.10 mL	α-Ketoglutarate (0.33 mol/L)
0.10 mL	NADH (7.5 mmol/L)
0.20 mL	Ammonium chloride (2.0 mol/L)
0.02 mL	rGlDH (NAD) (approx. 1.9 U/mL)

II Calculation

$$\frac{\Delta\ A/min \cdot V \cdot D}{6.3 \cdot d \cdot v} = U/mL$$

Δ A/min = The change in absorbance at 340 nm/minute

V = Total volume of reaction mixture (3.02 mL)

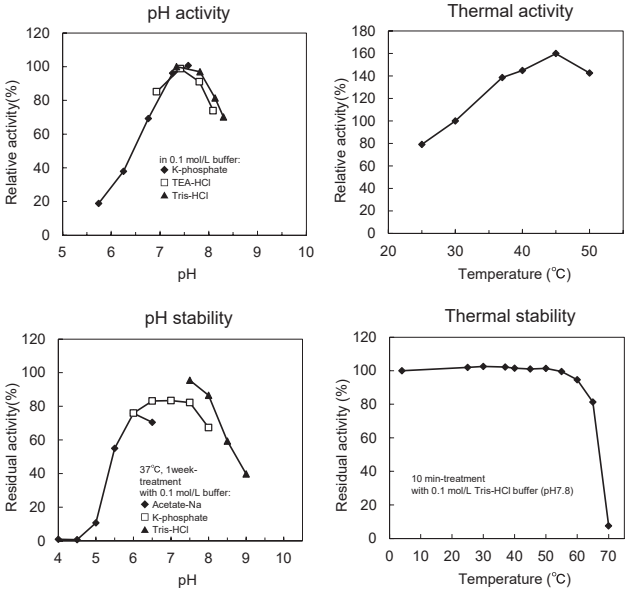
D = Enzyme dilution factor

6.3 = mmol/L extinction coefficient of NADH ($L \cdot mmol^{-1} \cdot cm^{-1}$)

d = Light path length (1 cm)

v = Volume of enzyme sample (0.02 mL)

Reference Data



Preparation and Storage

Lyophilized powder (Ammonium sulfate free)

Store below -20°C

Cat. No./Package

Cat. No.	Package
46874903	Bulk

For in vitro diagnostic or research use only