

# Enolase

## Phosphopyruvate hydratase

2-Phospho-D-glycerate hydro-lyase (EC 4.2.1.11)

### from Yeast

#### Reaction Equation



#### Specification

##### Specific Activity

IU/mg protein

##### Contaminants

Phosphoglyceromutase  
Pyruvate kinase

##### Specifications

>40 units

<0.02%

<0.02%

#### Assay Procedure

##### I. Spectrophotometric Method

Wavelength ; 340 nm, Light path length ; 1 cm,  
Temperature ; 25°C

Pipette the following reagents into a cuvette

2.31 mL	Triethanolamine-HCl-NaOH buffer (0.1 mol/L, pH 7.5)
0.15 mL	MgCl <sub>2</sub> (0.1 mol/L)
0.15 mL	KCl (2.0 mol/L)
0.15 mL	ADP (10 mmol/L)
0.09 mL	2,3-Diphospho-D-glycerate (10 mmol/L)
0.15 mL	Glycerate-3-phosphate (0.4 mol/L)
0.05 mL	NADH (10 mg/mL) dissolved in Tris (10 mmol/L)
0.002 mL	PK (5,000 IU/mL)
0.003 mL	LDH (10,000 IU/mL)
0.01 mL	(r) PGAM (770 IU/mL)
0.02 mL	Enolase (about 3 IU/mL)

##### II. Calculation

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

$\Delta A/\text{min}$  = The change in absorbance at 340 nm/minute  
(revise the blank activation of Enolase (-))

V = Total volume of reaction mixture  
(3.085 mL)

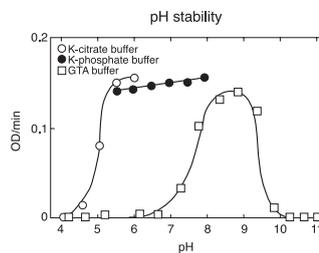
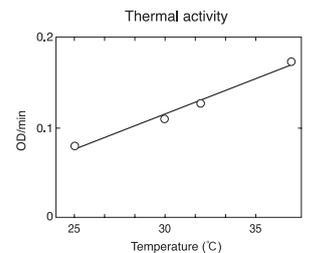
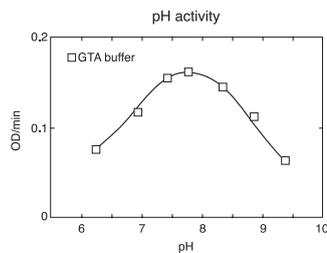
D = Enzyme dilution factor

6.3 = mM extinction coefficient of NADH  
(L·mmol<sup>-1</sup>·cm<sup>-1</sup>)

d = Light path length (1 cm)

v = Volume of enzyme sample (0.02 mL)

#### Reference Data



#### Preparation and storage

Product Code : Enolase-05

50% Glycerol solution..... -25°C ~ -15°C

IU per 1 ml solution is approximately 600 units.

#### OYC No./Package

OYC No.	Package
46450005	1,500 units
46451005	6,000 units
46452005	30,000 units
46449905	Bulk

(Research reagent use only, not for medical use.)



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