# HK (GRADE-I)

## Hexokinase

ATP: D-hexose 6-phosphotransferase (EC 2.7.1.1)

## from Yeast

#### Reaction Equation

ATP + D-Hexose = ADP + D-Hexose 6-phosphate

#### Specification

Specific Activity	Specifications
IU/mg protein	>140 units
Contaminants	
Phosphoglucose isomerase	< 0.1%
Glutathione reductase	< 0.05%
Myokinase	< 0.01%
Phosphogluconate dehydrogenase	< 0.005%
Phosphoglucomutase	< 0.01%
Glucose-6-phosphate dehydrogenase	< 0.01%
Creatine kinase	< 0.005%
ATPase	< 0.003%

#### Assay Procedure

#### I. Spectrophotometric Method

Wavelength;  $340\ \mathrm{nm}$ , Light path length;  $1\ \mathrm{cm}$ ,

Temperature ;  $25^{\circ}$ C

### Pipette the following reagents into a cuvette

2.40 mL	Triethanolamine-HCl-NaOH buffer
	(- · · · · · · · · · · · · · · · · · · ·

(0.1 mol/L, pH 7.5)

containing Glucose (50 mg/mL)

0.30 mL MgCl<sub>2</sub> (0.1 mol/L) 0.15 mL ATP (10 mmol/L) 0.15 mL NADP<sup>+</sup> (10 mmol/L) 0.01 mL G-6-PDH (500 IU/mL) 0.02 mL HK (about 3 IU/mL)

#### I. Calculation

$$\frac{\Delta A/\min \cdot V \cdot D}{6.2 \cdot d.m} = IU/mL$$

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

V = Total volume of reaction mixture (3.03 mL)

D = Enzyme dilution factor

6.2 = mM extinction coefficient of NADPH

 $(L \cdot mmol^{-1} \cdot cm^{-1})$ 

d = Light path length (1 cm)

v = Volume of enzyme sample (0.02 mL)

#### Preparation and storage

Product Code: HK-93

Lyophilized powder (contains no ammonium sulfate)

⋯⋯below -20°C

IU per 1 mg powder is approximately 100 units.

#### OYC No./Package

#### Lyophilized

OYC No.	Package
46560023	500 units
46561023	2,500 units
46562023	10,000 units
46563993	Bulk

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

