

SPECIFICATION SHEET

Product Name: Glycerol Kinase
Catalog # GK01D1-E311H

Product Description: This enzyme is a diagnostic grade reagent from a microorganism host. It is useful for enzymatic determination of glycerol and of triglyceride when coupled with glycerol-3-phosphate oxidase or pyruvate kinase and lactate dehydrogenase, lipoprotein lipase in clinical analysis.

EC 2.7.1.30

Storage and Stability: Product can be stored at 2~8°C for transportation process up to ten days but long-term storage should be at -20°C.

Principle:

ATP: glycerol 3-phosphotransferase

$$\text{ATP} + \text{glycerol} \xrightarrow{\text{Mg}^{2+}} \text{ADP} + \text{glycerol 3-phosphate}$$

Unit Definition	: One unit causes the formation of one micromole of hydrogen peroxide (half a micromole of quinoneimine dye) per minute at pH 7.9 and 37°C.	
Appearance	: White amorphous powder, lyophilized	
Activity	: ≥ 30 U/mg solid	
Molecular Weight	: 56.1 kDa	
Contaminants	: Catalase	$\leq 1.0 \times 10^{-1} \%$
	: NADH oxidase	$\leq 1.0 \times 10^{-3} \%$
	: Adenosine triphosphatase	$\leq 1.0 \times 10^{-3} \%$
Isoelectric Point	: 5.83	
Michaelis Constant	: 6.88×10^{-5} M (Glycerol), 4.88×10^{-5} M (NAD ⁺)	
Inhibitors	: Ag ⁺ , Hg ²⁺ , NEM, SDS, Proclin-300	
Optimum pH	: 7.5 ~ 8.0	(Figure 1)
Optimum Temperature	: 50°C	(Figure 2)
pH Stability	: pH 5.5-10.0 (25°C, 20 hr)	(Figure 3)
Thermal Stability	: Below 65°C (pH 7.5, 30 min)	(Figure 4)
Substrate Specificity	: (Table 1)	
Effect of Various Chemicals	: (Table 2)	

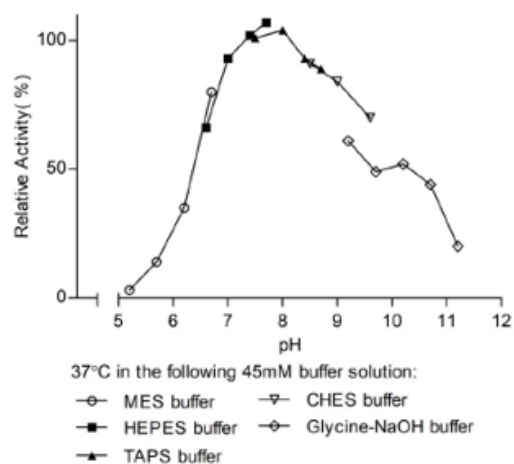
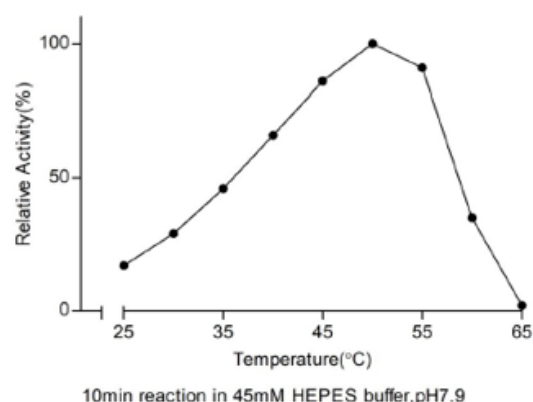
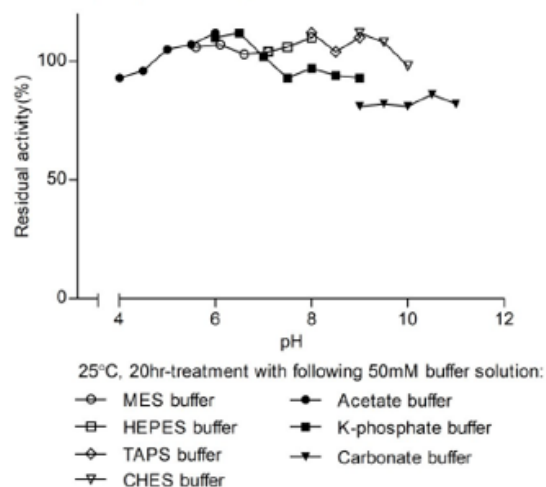
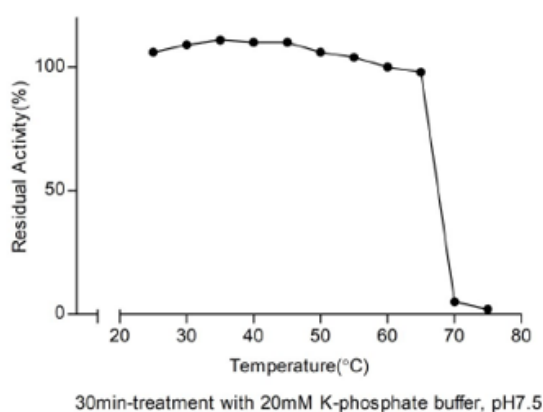
Table 1. Substrate Specificity of Glycerol kinase

Substrate (4.76 mM)	Relative activity (%)	Substrate (4.76 mM)	Relative activity (%)
Glycerol	100	2,3-Butanediol	0.48
Glycerol- α -monochlorohydrin	11	1,4-Butanediol	0.48
Glucose	1.4	D-manitol	0
1,3-Propanediol	1.93	Ethanol	0

Table 2. Effect of Various Chemicals on Glycerol kinase

The enzyme dissolved in 20 mM K-phosphate buffer, pH 7.5 (100 U/mL) was incubated with each chemical at 25°C for 1 hr.

Chemical	Conc. (mM)	Residual activity (%)	Chemical	Conc. (mM)	Residual activity (%)
None	—	100	BME	2	96
CaCl ₂	2	98	Hydroxylamine	2	104
MgSO ₄	2	103	EDTA	5	105
ZnSO ₄	2	93	NaF	20	103
NiCl ₂	2	94	NaN ₃	20	106
CoCl ₂	2	101	Proclin-300	0.045% (v/v)	15
MnCl ₂	2	99	SDS	0.05% (w/v)	1
FeCl ₃	2	94	Na-Cholate	0.1% (w/v)	122
CuSO ₄	2	99	Tween-20	0.1% (v/v)	111
AgNO ₃	2	75	Triton X-100	0.1% (v/v)	117
HgSO ₄	2	1	Span-20	0.1% (v/v)	113
NEM	2	12	Brij-35	0.1% (w/v)	114
IAA	2	104			

Fig.1. pH Activity**Fig.2. Temperature Activity****Fig.3. pH Stability****Fig.4. Thermal Stability**

Version: 1.0