

SPECIFICATION SHEET

Product Name: Cholesterol Oxidase
Catalog # CX01D1-E311H

Product Description: Cholesterol oxidase is a recombinant diagnostic grade reagent from a microorganism host. This enzyme is useful for enzymatic determination of cholesterol in serum when coupled with cholesterol in clinical analysis.

EC 1.1.3.6

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:


Unit Definition	: One unit is defined as the amount of enzyme which liberates 1 µmol of Δ^4 -Cholest-3-one per minute at 37°C at pH 7.0.	
Appearance	: Yellowish amorphous powder, lyophilized	
Activity	: ≥12 U/mg solid	
Contaminants	: Catalase	≤1.0 × 10 ⁻¹ %
	: Cholesterol esterase	≤1.0 × 10 ⁻² %
Stabilizers	: BSA, amino acids	
Molecular Weight	: 60 kDa	
Isoelectric Point	: 8.4	
Michaelis Constant	: 1.2 × 10 ⁻⁵ M (Cholesterol)	
Inhibitors	: Cu ²⁺ , Ag ⁺ , Hg ²⁺	
Optimum pH	: 6.5 ~ 7.2	(Figure 1)
Optimum Temperature	: 55°C	(Figure 2)
pH Stability	: pH 5.5~10.5 (25°C, 20 hr)	(Figure 3)
Thermal Stability	: Below 50°C (pH 7.0, 15 min)	(Figure 4)
Substrate Specificity	: (Table 1)	
Effect of Various Chemicals	: (Table 2)	

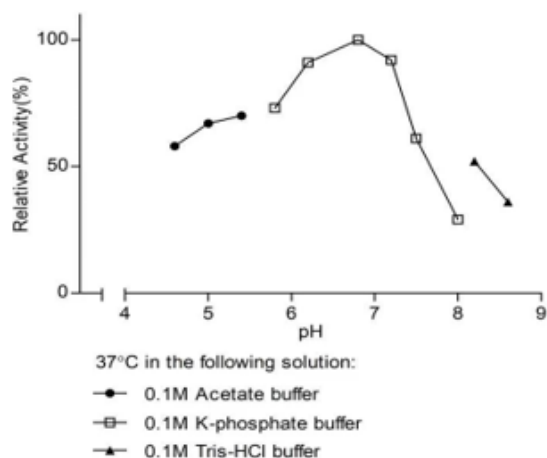
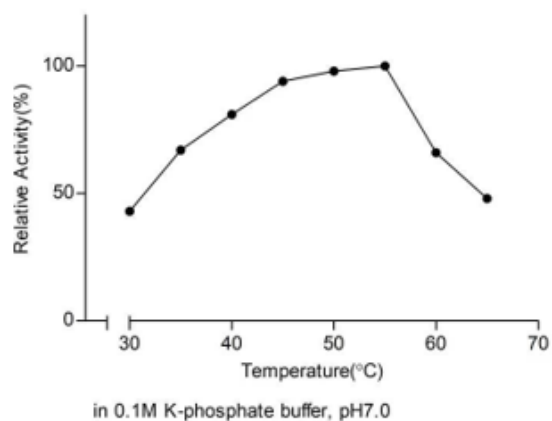
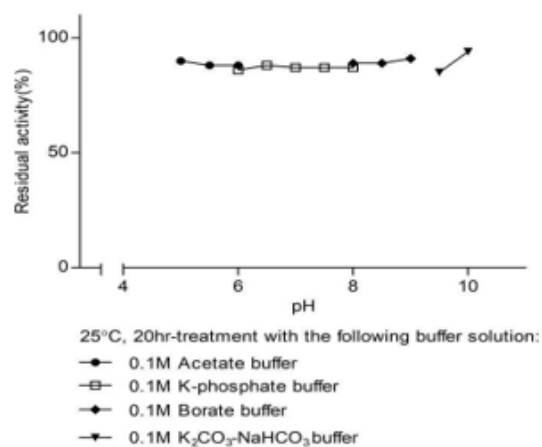
Table 1. Substrate Specificity of Cholesterol Oxidase

Substrate (0.1 mM)	Relative activity (%)	Substrate (0.1 mM)	Relative activity (%)
Cholesterol	100	Ergosterol	32
Pregnenolone	50	Lanosterol	1
B-Cholesterol	61	Testosterone	0
B-Sitosterol	128	Androsterone	2
Stigmasterol	30	Dehydroisoandrosterone	24
5β-Pregnane-3α, 20α-diol	0		

Table 2. Effect of Various Chemicals on Cholesterol oxidase

The enzyme dissolved in 100 mM phosphate buffer, pH 7.0 (1.0 U/mL) and 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	93	Hydroxylamine	2	100
MgSO ₄	2	97	EDTA	5	95
ZnSO ₄	2	95	NaF	20	98
NiCl ₂	2	94	NaN ₃	20	99
CoCl ₂	2	98	Borate	50	100
MnCl ₂	2	100	Proclin-300	0.045% (v/v)	94
FeCl ₃	2	86	SDS	0.05% (w/v)	94
CuSO ₄	2	57	Na-Cholate	0.1% (w/v)	97
AgNO ₃	2	0	Tween-20	0.1% (v/v)	99
HgSO ₄	2	0	Triton X-100	0.1% (v/v)	97
NEM	2	99	Span-20	0.1% (v/v)	89

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