Cholesterol Oxidase from microorganism

COO-122S

SPECIFICATIONS

Product name:	Cholesterol: oxygen oxidoreductase
EC :	1.1.3.6
Appearance:	Yellow to brownish lyophilized powder
Activity:	> 25 U/mg
Contaminants:	Catalase: < 1 % Cholesterol esterase: < 0.1 % Glucose oxidase : < 0.1 %
Stability:	Stable at - 20 °C for at least one year
Stabilizers:	Bovine serum albumin, amino acids
Molecular weight:	Approx. 57,000
Michaelis constants:	4 x 10^{-6} M (Cholesterol)
Inhibitors:	Ionic detergents, Hg^{2+}
Optimum pH:	6.5 - 8.0
Optimum temperature:	50 °C
pH Stability:	pH 4.5 - 10.0 (25°C, 20hr)
Thermal stability:	Below 60 °C (pH 7.0, 15 min)

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Table 1: Substrate specificity of Cholesterol oxidase

Substrate (0.15 mM)	Relative activity	Substrate (0.15 mM)	Relative activity
Cholesterol	100 %	Cholesterol linoleate	1 %
Pregnenolone	85 %	Androsterone	1 %
β–Cholestanol	74 %	Ergosterol	36 %
β-Stigmasterol	44 %	Testosterone	1 %
Lanosterol	2 %	Androsterone	1 %

Table 2. Effect of Various Chemicals on Cholesterol oxidase
[The enzyme (1.0U/m) dissolved in 10mM phosphate buffer, pH 7.0 contg. 0.2% BSA was incubated with each chemical at 25℃ for 1hr.]

Chemical	Concn.(mM)	Residual activity
None	—	100%
Metal salt	2.0	
$MgCl_2$		100
$CaCl_2$		94
$Ba(OAc)_2$		100
$FeCl_3$		83
$CoCl_2$		100
$MnCl_2$		100
$Zn(OAc)_2$		98
$Cd(OAc)_2$		100
$NiCl_2$		95
$CuSO_4$		91
$Pb(OAc)_2$		100
$AgNO_3$		0
$HgCl_2$		0
PCMB	2.0	100
MIA	2.0	100

Chemical	Concn.(mM)	Residual activity
NaF	20	98
NaN_3	20	95
EDTA	5.0	97
o-Phenanthroline	2.0	100
α,α'-Dipyridyl	1.0	100
Borate	50	100
IAA	2.0	98
NEM	2.0	98
Hydroxylamine	2.0	95
2-Mercaptoethanol	2.0	100
Triton X-100	0.10%	100
Tween 20	0.10%	98
Span 20	0.10%	89
Na-cholate	0.10%	100
SDS	0.05%	100
DAC	0.05%	100

Ac, CH_3CO; PCMB, p-Chloromercuribenzoate; MIA, Monoiodoacetate; NEM, N-Ethylmaleimide; IAA, Iodoacetamide;
EDTA, Ethylenediamimetetraacetate;
SDS, Sodium dodecyl sulfate; DAC, Dimethyl-benzyl-alkyl-ammonium-chloride.

ENZYMES BY SORACHIM

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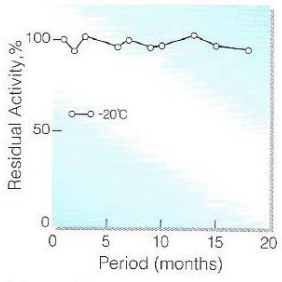

Fig.1. Stability (Powder form)
〔kept under dry conditions〕

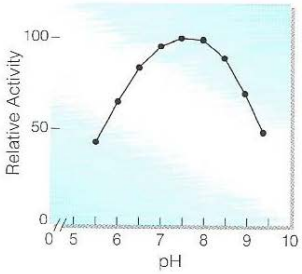

Fig.3. pH-Activity
〔37℃ in 0.1M K-phosphate buffer solution〕

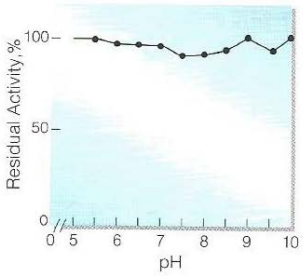

Fig.5. pH-Stability
〔25℃ 20 hr-treatment with 50mM buffer solution:pH5.0-6.0, acetate; pH6.5-8.5, K-phosphate; pH9-10.0,K_2CO_3-$NaHCO_3$〕

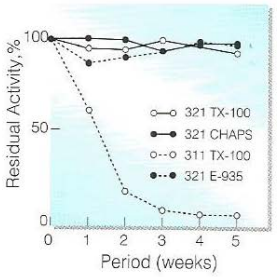

Fig.2. Stability (Liquid form)
〔40℃ in buffer solution, pH7.0〕

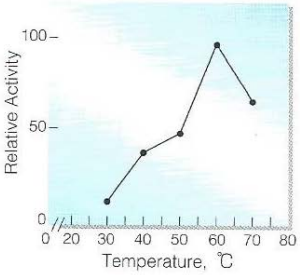

Fig.4. Temperature activity
〔in 0.1M K-phosphate buffer,pH 7.0〕

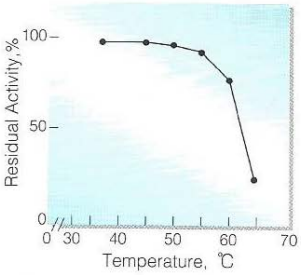

Fig.6. Thermal stability
〔15 min-treatment with 50mM K-phosphate buffer, pH7.0〕