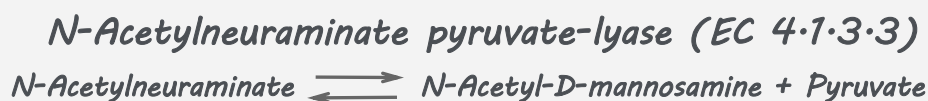


N-ACETYLNEURAMINIC ACID ALDOLASE

from Microorganism



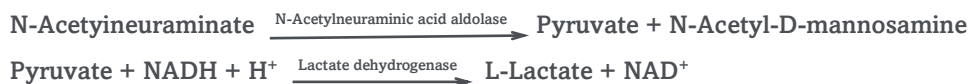
PREPARATION and SPECIFICATION

Appearance	: Slight yellow amorphous powder, lyophilized
Activity	: 15U/mg-solid or more
Contaminants	: Catalase $\leq 1.0\%$; NADH oxidase $\leq 1.0 \times 10^{-3}\%$
Stabilizers	: Mannitol, EDTA

PROPERTIES

Stability	: The product can be stored at 2~8°C for transportation process up to ten days but long-term storage should be at -20°C.	
Molecular weight	: 33.9 kDa	
Isoelectric point	: 6.22	
Michaelis constant	: $2.8 \times 10^{-3} \text{M}$ (N-Acetylneuraminic acid)	
Inhibitors	: Co^{2+} , Cu^{2+} , Ag^{2+} , Hg^{2+} , NEM, Proclin	
Optimum pH	: 7.5~8.0	(Fig.1)
Optimum temperature	: 50°C	(Fig.2)
pH stability	: pH4.5~10.5 (25°C, 25hr)	(Fig.3)
Thermal stability	: below 80°C (pH7.5, 30min)	(Fig.4)
Effect of various chemicals	: (Table 1)	

PRINCIPLE



The disappearance of NADH is measured at 340nm by spectrophotometry.

UNIT DEFINITION

One unit causes the oxidation of one micromole of NADH per minute at pH7.5 and 37°C

APPLICATIONS

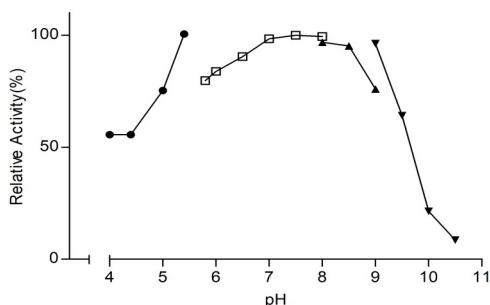
This enzyme is useful for enzymatic determination of N-acetylneuraminic acid and sialic acid when coupled with the related enzyme in clinical analysis.

Table 2. Effect of Various Chemicals on N-Acetylneuraminic acid aldolase

The enzyme dissolved in 50mM K-phosphate buffer, pH7.5 (10U/ml) was incubated with each chemical at 37°C for 2 hr.

Chemical	Concn.(mM)	Residual activity(%)	Chemical	Concn.(mM)	Residual activity(%)
None	—	100	BME	2	98
CaCl ₂	2	93	Hydroxylamine	2	101
MgSO ₄	2	92	EDTA	5	95
ZnSO ₄	2	121	NaF	20	99
NiCl ₂	2	130	NaN ₃	20	97
CoCl ₂	2	64	Proclin-300	0.045% (v/v)	56
MnCl ₂	2	93	SDS	0.05% (w/v)	86
FeCl ₃	2	94	Na-Cholate	0.1% (w/v)	102
CuSO ₄	2	0	Tween-20	0.1% (v/v)	102
AgNO ₃	2	1	Triton X-100	0.1% (v/v)	101
HgSO ₄	2	1	Span-20	0.1% (v/v)	102
NEM	2	2	Brij-35	0.1% (w/v)	102
IAA	2	87			

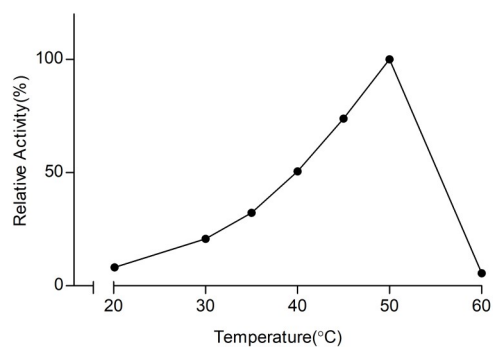
Fig.1. pH-Activity



37°C in the following solution:

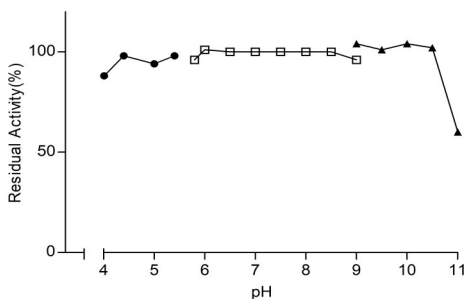
- 50mM Acetate buffer
- 50mM K-phosphate buffer
- ▲ 50mM Tris-HCl buffer
- ▼ 50mM Glycine-NaOH buffer

Fig.2. Temperature Activity



in 50mM K-phosphate buffer, pH7.5

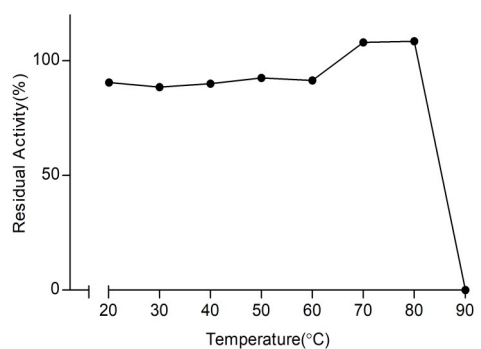
Fig.3. pH Stability



25°C 25hr-treatment with 50mM buffer solution:

- Acetate buffer
- K-phosphate buffer
- ▲ Borate buffer

Fig.4. Thermal Stability



30min- treatment with 50mM K-phosphate buffer, pH7.5