FUJIFILM

Wako

Code No. 012-29241 (2,000,000 units)
018-29243 (20,000,000 units)

Achromopeptidase™, Crude, Lytic Enzyme
(TBL-1)

アクロモペプチダーゼ™, 粗製品, 溶菌酵素
(TBL-1)

Bacteriolytic enzymes are hydrolases that act on bacterial cell wall and cause cell-lysis. The most investigated bacteriolytic enzyme has been lysozyme. Lysozyme is now produced on an industrial scale and has found many applications. Lysozyme, however, is bactericidal only against some Gram-positive organisms (e. g., *Micrococci, Bacilli, Sarcinae, Pediococci, Kurthiae*) and not against pathological organisms such as *Staphylococci, Streptococci* and *Clostridia*. Many new bacteriolytic enzymes have been described and several originate from microorganisms. The soil bacterium first isolated in 1966 has been found to produce a highly active bacteriolytic enzyme with a broad specificity. Like lysozyme, the enzyme causes lysis of *Micrococci, Bacilli* and *Sarcinate* but unlike lysozyme will also cause lysis of *Staphylococci, Streptococci, Clostridia* and *Leuconostoc* and, in addition, is active against some Gram-negative organisms. FUJIFILM Wako has investigated the properties of the bacteriolytic enzyme and has succeeded in producing it on a large scale. This bacteriolytic enzyme is now available as Achromopeptidase (TBL-1) .

Source : *Lysobacter enzymogenes*
Activity : indicated on the label
Molecular weight : about 20,000
Stability at various pH values : Most stable at pH 6.0 and virtually stable at pH 5.5 ～ 10
Appearance : Pale yellowish brown ～ brown, powder
Heat stability : incubation in 50 mmol/L phosphate buffer at pH 6.0 for 10 minutes

Temperature	Activity
Less than 40℃	Stable
50℃	95% remaining
60℃	75% remaining
70℃	Lost

Optimal pH : 7.5 ～ 8.5
Optimal ionic strength : 10 mmol/L sodium chloride (in 10 mmol/L Tris-HCl buffer solution (pH 8.0))

Bacteriolytic Spectrum

Microorganism	Degree of lysis*	
	TBL-1 (300 U/mL)**	Egg lysoxyme (1,000 U/mL)
Micrococcus caseolyticus	+ + +	−
Micrococcus luteus	+ + +	−
Pediococcus acidilactici	+ + +	+
Staphylococcus aureus	+ + +	±
Microbacterium arborescens	+ +	+
Bacillus alvei	+ + +	±
Clostridium acetobutylicum	+ + +	−
Brevibacterium leucinophagum	+	±
Lactobacillus sakei	+ + +	+
Enterococcus faecalis	+ + +	±
Leuconostoc dextranicum	+ +	±
Achromobacter liquidum	+	−
Nocardioides simplex	+	+
Beijerinckia indica	+	±
Mycobacterium diernhoferi	±	±
Kurthia zopfii	+ +	+

*determined by decrease of the optical density at 600 nm in each microorganism suspension under the standard assay conditions incubated for indicated times.
+ + + ; ≧ 80% decrease within 10 min and + + ; ≧ 80%, + ; 80-40, ± ; 40-10%, − ; 10-0% decrease within 60 min.
**The activity value of the conventional product (014-09661).

Assay Method

1) Reagents
A : 0.01 mol/L Tris-HCl + 0.01 mol/L NaCl buffer solution (pH 8.0)
B : Substrate
Suspend 31-32 mg of lyophilized *Micrococcus lysodeikticus* IFO 3333 in Reagent A to bring the volume up to 30 mL. Transfer 5 mL of the solution and dilute with Reagent A to bring the volume up to 25 mL. Place it on ice for 1 hour.
C : Enzyme solution
Dissolve 10 mg of TBL-1 with Reagent A to bring the volume up to 20 mL. Place it on ice for 30 minutes with stirring occasionally.

Procedure :

Reagent	Test	Blank
B	3.0 mL	3.0 mL
	Pre-incubate in 10 mm absorption cell (37℃ 5 min.) Trans * 10mm 吸収セルに入れ、37℃で5分間予備加温する。	
C	0.05 mL	−
A	−	0.05 mL
Immediately, record the absorbance of 600 nm at 37℃ for 1 ～ 6 minutes after addition of Reagent C and Reagent A with deionized water as a reference. Trans * 直ちに、37℃で波長 600nm における吸光度を水を対照液として試液 C 及び試液 A 添加後 1 ～ 6 分間自記記録する。		

Enzymatic activity
One unit of enzyme is defined as that which when present in 1 mL of reaction solution causes turbidity to decrease at the rate of 0.001 per minute under the above conditions.

Calculation formula for determining activity.

$$\text{Units/mg} = \frac{\Delta E - \Delta E_0}{0.001} \times 3.05 \times \frac{1}{S} \times \frac{20}{0.05} \times f$$

ΔE : The change of absorbance of Test for 1 minute.
ΔE_0 : The change of absorbance of Blank for 1 minute.
S : Amount of TBL-1 (mg)
f : 0.9 (correction factor)

Trans *
活性単位の計算式

$$\text{Units/mg} = \frac{\Delta E - \Delta E_0}{0.001} \times 3.05 \times \frac{1}{S} \times \frac{20}{0.05} \times f$$

ΔE : 本試験の 1 分間当りの吸光度変化
ΔE_0 : 空試験の 1 分間当りの吸光度変化
S : 量りとった本品の質量 (mg)
f : 0.9 (補正値係数)

Cautions in Use
(1) The optimal pH value for TBL-1 is between 7.5 and 8.5. It should be used in this range.
(2) TBL-1 is greatly influenced by ionic strength and its activity is markedly decreased with increasing ionic strength. In 10 mmol/L Tris-HCl buffer solution (pH 8.0) containing 10 mmol/L sodium chloride the enzyme is optimally active.
(3) Aqueous solutions of TBL-1 at concentrations up to 0.4% (w/v) remain transparent. At higher concentrations, aqueous solutions of TBL-1 become turbid due to impurities. The turbidity can be cleared by centrifugation. If large amounts of insoluble substances are noted, do not use the enzyme as it may have deteriorated.
(4) TBL-1 is not purified to homogeneity and therefore may contain enzymes other than TBL-1 and this should be taken into consideration when this product is used. Purified TBL-1 (Achromopeptidase®, Purified, Lytic Enzyme (TBL-1), 019-28531) is available for use when required.

【Storage】
2 ～ 10℃

【Package】

Code No.	Packaging
012-29241	2,000,000 units
018-29243	20,000,000 units

【References】
1) Isono, M., Takahashi, K. and Yamazaki, Y., : Patent Notice, Sho 46-42953.
2) Nakamura, K., Okazawa, Y., Soejima, M. and Masaki, T. : *Agri. Biol. Chem.*, **37**, 2667 (1973).
3) Horinouchi, S., Uozumi, T., Beppu, T. and Arima, K. : *ibid.*, **41**, 2487 (1977).
4) Kotani, S. : *Protein, Nucleic Acid, Enzyme*, **13**, 1136 (1968).
5) Kotani, S. : *Protein, Nucleic Acid, Enzyme*, **14**, 38 (1969).

＊ : **Trans** **is the Japanese translation.**

FUJIFILM Wako Pure Chemical Corporation

1-2, Doshomachi 3-Chome, Chuo-Ku, Osaka 540-8605, Japan
Telephone : +81-6-6203-3741
Facsimile : +81-6-6201-5964
http://ffwk.fujifilm.co.jp

Group Companies **Distributors**

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