



Cat. No.: URID-001

# Uricase-D

## (ETERBIO-EZ-URID-001)

Lot No.: \_\_\_\_\_

Expiry Date: yy / mm / dd

Store at -20°C

### Origin

Microorganism

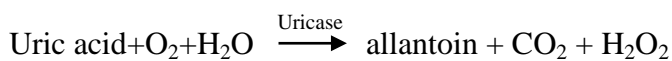
### Specification

<b>Appearance</b>	Lyophilized powder
<b>Activity</b>	$\geq 30$ U/mg protein at 37°C

### Properties

<b>Molecular weight</b>	37 kDa	
<b>Isoelectric point</b>	6.1	
<b>Stability</b>	stored at -20°C for at least 1 year	(Fig.1)
<b>Optimum pH</b>	6.5	(Fig.2)
<b>Optimum temperature</b>	45°C	(Fig.3)
<b>pH stability</b>	7.5-10.0 (4°C, 16hr )	(Fig.4)
<b>Thermal stability</b>	50°C	(Fig.5)

### Assay



The decrease of uric acid is measured at 293nm by spectrophotometry.

### Unit definition

One unit enzyme will cause the oxidation of one micromole of uric acid per minute at 37°C under the standard assay.

### Method

#### **Reagent :**

(A) 20 mM Boric acid, pH9.0



(B) 3.57 mM uric acid

(C) Enzyme solution : Dissolve in 10 mM K-phosphate, pH7.0

**Procedure :**

1. Prepare 3 ml (A) and 0.1ml (B), then put the reaction mixture in a cuvette (d = 1.0cm) and equilibrate at 37°C for about 5 minutes.
2. Add 0.02 ml (C) (dilute the enzyme to 0.3-0.6 U/ml with the K-phosphate buffer), mix well with procedure 1.
3. Record the decrease in optical density at 293 nm for 5 minutes in a spectrophotometer at 37°C, and calculate the  $\Delta OD$  per minute. The value of  $\Delta OD/\text{min}$  becomes in the range of 0.05- 0.10.

**Calculation :**

$$\text{Uricase activity (U/ml)} = \frac{(A_{293\text{nm}}/\text{min Test} - A_{293\text{nm}}/\text{min Blank}) \times V_t \times df}{12.6 \times 1.0 \times V_s}$$

$$\text{Weight activity (U/mg)} = (\text{U/ml}) \times 1/C$$

$V_t$  : Total volume (3.12ml)

$V_s$  : Sample volume (0.02ml)

12.6 : Millimolar extinction coefficient of uric acid at 293nm

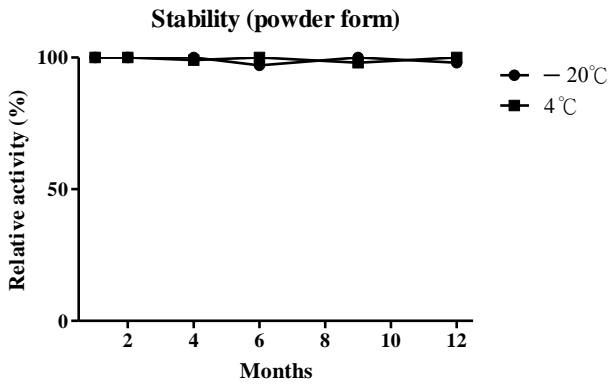
1.0 : Light path length (cm)

df : Dilution factor

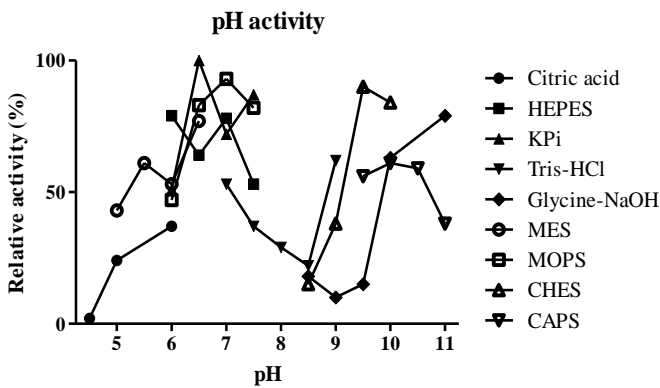
C : Enzyme concentration in dissolution (mg/ml)



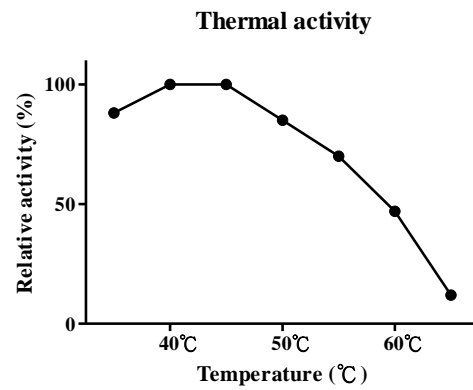
**Fig. 1**



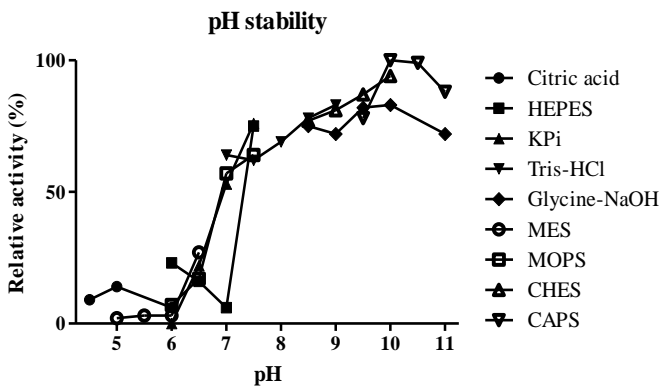
**Fig. 2 (37°C treatment in 50 mM buffer)**



**Fig. 3 (5 min in 20 mM Boric acid, pH9.0)**



**Fig. 4 (25°C 16 hr treatment in 50 mM buffer)**



**Fig. 5 (15 min in 20 mM Boric acid, pH9.0)**

