

# Apo D-Amino Acid Oxidase, Enzyme Activity

Catalog	Unit
TBP0056-100U	100 U
TBP0056-500U	500 U

### **Product Details**

Form: Freeze-dried powder

Solubility: Distilled water or dilute buffer

Stability: Store at -20° C (-4° F)

Activity: 25-30 U/mg protein RCHNH2COOH + O2 + H2O -----> RCOCOOH + NH3 + H2O2

Protein: 90%

Contaminants: Free of FAD and Alkaline Phosphatase

#### **Unit Definition**

The amount of enzyme that will deaminate by oxidation one micromole of D-alanine to pyruvate per minute at pH 8.3, at 37°C in the presence of catalase.

## **Assav Method**

The assay is based on the method described by Bergmeyer. The decrease in the absorbance at 340 nm, due to the oxidation of NADH, is a measure of D-amino acid oxidase activity.

### **Applications**

The D isomers of alanine, methionine, valine, isoleucine, phenylalanine and proline serve as good substrates while the L isomers do not react at all. The enzyme is a flavoprotein. D-amino acid oxidase from porcine kidney has been extensively studied. It has a monomeric molecular weight of 38,000-39,000.

D-Amino acid oxidase (EC 1.4.3.3) has several possible applications such as the determination of D-amino acids, the separation of natural L- amino acid isomers from a racemic mixture and in the preparation of keto acids. The usefulness and application of D-amino acid oxidase can be significantly increased if it is available in an immobilized form.

#### Reagents

- 1. 0.2 M Tris-HCl buffer, pH 8.3.
- 2. 0.02 M D-Alanine (17.8 mg/ml) in buffer.
- 3. 0.008 M NADH disodium salt (5 mg/ml) in buffer.
- 4. Catalase (200 U/ml) in buffer. Prepare fresh.
- 5. Lactate dehydrogenase (LDH) (200 U/ml) in buffer. Prepare fresh.
- 6. FAD (Prepare1 mg/ml solution)
- 7. D-Amino acid oxidase solution. Dilute in buffer to give a concentration of 0.1-0.5 U/ml. Must be prepared fresh prior to assay.

### **Calculation**

Activity (U/mg) = 
$$\frac{(\Delta E_{340\text{nm/min}})(\text{Total Vol.})(\text{Enz. Diln.})}{(6.22)(\text{Enz. Vol.})(\text{mg Enz./ml})}$$

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Tribioscience, Inc.; 365 San Aleso Ave, Sunnyvale, CA 94085 Phone: 408-498-0197

info@tribioscience.com; www.tribiosciences.com