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TECHNICAL INFORMATION

Catalog Number: 100277 Arginase Molecular Weight: 115,000 - 120,000.6 CAS #: 9000-96-8 Synonyms: E.C. 3.5.3.1; L-arginine amidinase; L-arginine amidino-hydrolase Physical Description: Brown powder Source: Beef liver Form: A salt free freeze-dried powder **Composition:** The amino acid content has been reported by Greenberg.⁵ The enzyme does not split into subunits; it binds 4 atoms of Mn2+ which are essential to its activity as well as stability and has a hexose content of 3-5%.6 Unit Definition: That amount of enzyme causing the hydrolysis of one micromole of L-arginine per minute at 37°C and pH 9.5 Optimum pH: 10 (Mn²⁺ activated)⁵ Extinction Coefficient: E^{1%}₂₇₈ = 9.6.6 Solubility: Dissolves readily at 1 mg/ml in 0.05 M manganese maleate pH 7.0 to give a clear pale brown solution, also soluble in distilled water or dilute buffer **Description:** Arginase catalyzes the following reaction: Arginine + H₂O ----> Ornithine + Urea The enzyme participates in the Krebs-Henseleit urea cycle. It is most highly concentrated in mammalian liver. It is also present in the mammary glands.¹² Specificity: Arginase activity requires the free guanidino group and the free carboxyl group of arginine.⁵ Activators:12 Mn2+, Ni2+ or Co2+ Inhibitors:12 Hg2+, Ag2+, Zn2+ Stability: Susceptibility of arginase to proteolytic inactivation is enhanced by increasing activating concentration of Mn^{2+,4} Cysteine, isoleucine, leucine, valine and alanine protect the enzyme against trypsin inactivation.¹² **Assav:**^{1,12} Method: Based upon the colorimetric determination of released urea nitrogen, using 2,3-butanedione reagent. Reagents: A.0.05 M Maleic acid, pH 7.0 with 0.05 M manganous sulfate B.0.713 M L-arginine, pH 9.5 C.Reagent grade water, pH 9.5 (20 mg NaOH per liter). D.0.075%. 2.3-Butanedione in buffered arsenic-sulfuric acid Enzyme: Incubate a one mg/ml solution of the enzyme in maleic-manganous sulfate buffer (Reagent A) at 37°C for 4 hours. Following

activation, dilute to 1-2 micrograms/ml in reagent grade water, pH 9.5 (Reagent C).

Procedure:

Pipette the following into screw capped tubes:

| | | Tube | |
|--|--------|--------|--------|
| | Blank | #1 | #2 |
| Water, pH 9.5 (Reagent C) Diluted, activated enzyme | 0.3 ml | 0.1 ml | 0.2 ml |
| | | 0.2 ml | 0.1 ml |

Incubate in 37°C water bath for 5 minutes to achieve temperature equilibration. At timed intervals start reaction by adding:Arginine (Reagent B)0.2 ml0.2 ml0.2 ml

Incubate for 30 minutes at 37°C. Stop reaction at timed intervals by adding:

4.0 ml

4.0 ml

Cap tubes and develop color by boiling in water bath for 12 minutes. Chill tubes for 3 minutes in an ice bath. Read the color at 490 nm against blank. *Calculation:*

Determine the micromoles of urea released from a standard urea curve in the range of 0.1-1.0 micromoles urea. micromoles urea released

Units/mg = 30 X mg enzyme in reaction mixture

Origin: USA References:

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- Supplier Information.