Tribioscience

Glutamic Oxaloacetic Transaminase, Enzyme Activity

Catalog	Unit
TBP0069-1KU	1000 U
TBP0069-5KU	5000 U

Product Details

Form: Ammonium Sulfate

Solubility: Distilled water or dilute buffer

Stability: Store at 4° C (39° F); Do not freeze

Activity: 200 U/mg protein

Protein: 10 mg/ml

Contaminants: Glutamate dehydrogenase <0.01%; Glutamate pyruvate transaminase <0.01%; Lactate dehydrogenase <0.01%; Malate dehydrogenase <0.01%; Oxaloacetate decarboxylase <0.05%

Unit Definition

The amount of enzyme which will catalyze the transamination of one micromole of L-aspartate per minute at 25°C and pH 7.5.

Assay Method

The assay is based on a coupled reaction where the oxaloacetate formed is reduced to malate in the presence of NADH and malate dehydrogenase. The decrease in the absorbance at 340 nm caused by the oxidation of NADH is proportional to the catalytic activity of GOT.

Applications

(L-Aspartate: a-Ketoglutarate amino transferase, EC 2.6.1.1) Glutamic oxaloacetic transaminase (GOT), also known as aspartate aminotransferase, catalyzes the following reaction:

GOT L-Asp + a-Ketoglutarate ------> Oxaloacetate + L-Glu

The enzyme is widely distributed in plants and animals, but it occurs in concentrated form in mammalian heart and liver. GOT requires pyridoxal phosphate as a coenzyme for its activity. It exists as two isozyme forms, the mitochondrial form (M-GOT) and the cytosol form (S-GOT). GOT from porcine heart has a molecular weight in the range of 91,000-94,000. Serum GOT levels in healthy subjects are low, but the levels are significantly elevated in a number of clinical conditions such as acute and chronic hepatitis, obstructive jaundice, carcinoma of liver, myocardial infraction and muscular dystrophy. Therefore, determination of serum GOT level has great clinical and diagnostic significance. In addition, GOT has applications in coupled enzyme reactions for measurement of metabolite levels in biological fluids.

Reagents

1. 0.1 M Potassium phosphate buffer, pH 7.5.
2. 0.008 M NADH, disodium salt (5 mg/ml) in buffer. Prepare fresh.
3. 0.3 M L-Aspartate-monopotassium salt (51.4 mg/ml) in buffer.
4. 0.60 M a-Ketoglutarate (87 mg/ml) in buffer.
5. Malate dehydrogenase (MDH). Using phosphate buffer, prepare a solution to yield a concentration of 200-250 U/ml. Prepare fresh prior to assay.
6. Glutamate-oxaloacetate transaminase (GOT) enzyme solution. Using phosphate buffer, prepare a GOT solution with a concentration in the range of 0.2-0.4 U/ml. Must be prepared fresh prior to the assay.

Calculation

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

For research use only