α-Amylase (α-AL) Activity Assay Kit - Spectrophotometry

Product code: 111968

Product Introduction

Amylase hydrolyzes starch and includes α-amylase and β-amylase. α-Amylase (EC 3.2.1.1) randomly acts on α-1,4-glycosidic bonds in starch, producing glucose, maltose, maltotriose, dextrin, and other reducing sugars while reducing starch viscosity. It is also known as a liquefying enzyme.

Reducing sugars reduce 3,5-dinitrosalicylic acid to form a brown-red substance. β-Amylase is heat-labile and is inactivated at 70℃ for 15 min, enabling determination of α-amylase activity.

Product Packing List

Code	Component	50T	100T	Storage Temperature
111968.1	Reagent I	25 mL	2 x 25 mL	2-8℃, protected from light
111968.2	Reagent II	20 mL	2 x 20 mL	2-8℃
111968.m	Manual	1 copy	1 copy	-

Quality Standards and Safety Information

Raw Material or Packaging Name	Quality Standard	Main Toxicity
Reagent I	--	--
Reagent II	--	--

Shipping and Storage

Shipping: This product is shipped with ice packs.

Storage: Store according to the instructions. Shelf life: 180 days.

Instructions for Use

1. Preparation of Crude Enzyme Extract

Tissue: Weigh approximately 0.1 g of sample, add 1 mL distilled water, and homogenize. After homogenization, let stand at room temperature for extraction for 15 min. Shake once every 5 min to ensure complete extraction. Centrifuge at 3000 g at room temperature for 10 min. Collect the supernatant and add distilled water to a final volume of 10 mL. Mix well. This is the amylase stock solution.

Serum or plasma: Test directly.

2. Assay Procedure

1. Preheat the spectrophotometer for 30 min or more, set the wavelength to 540 nm, and use distilled water to zero the instrument.
2. Prepare tubes and operate according to the table below.

Component or Step	Control Tube	Assay Tube
Amylase stock solution	250 μL	250 μL

Component or Step	Control Tube	Assay Tube
70℃ water bath, then cool	15 min	15 min
Distilled water	250 μL	-
Reagent II	-	250 μL
40℃ constant-temperature water bath	Incubate accurately for 5 min	Incubate accurately for 5 min
Reagent I	500 μL	500 μL
Final reaction	Mix well, heat in a 95℃ water bath for 5 min, then cool	Mix well, heat in a 95℃ water bath for 5 min, then cool
Measurement	Read absorbance at 540 nm and record as A1	Read absorbance at 540 nm and record as A2

α-Amylase Activity Calculation

The regression curve determined under standard conditions is:

y = 3.7215x - 0.1778

Where x is the standard concentration in mg/mL and y is the absorbance value.

Calculated According to Sample Mass

Unit definition: The amount of enzyme that catalyzes the production of 1 mg reducing sugar per minute in the reaction system per g of tissue is defined as 1 enzyme activity unit.

α-Amylase activity (mg/min/g fresh weight) = [(A2 - A1 + 0.1778) ÷ 3.7215 x Vsample] ÷ (W x Vsample ÷ Vtotal sample) ÷ T = 0.537 x (ΔA + 0.1778) ÷ W

Calculated According to Protein Content

Unit definition: The amount of enzyme that catalyzes the production of 1 mg reducing sugar per minute in the reaction system per mg of tissue protein is defined as 1 enzyme activity unit.

α-Amylase activity (mg/min/mg prot) = [(A2 - A1 + 0.1778) ÷ 3.7215 x Vsample] ÷ (Vsample x Cpr) ÷ T = 0.0537 x (ΔA + 0.1778) ÷ Cpr

Calculated According to Serum or Plasma Samples

Unit definition: The amount of enzyme in each mL of serum or plasma that catalyzes the production of 1 mg reducing sugar per minute is defined as 1 enzyme activity unit.

α-Amylase activity (mg/min/mL) = [(A2 - A1 + 0.1778) ÷ 3.7215 x Vsample] ÷ Vsample ÷ T = 0.0537 x (ΔA + 0.1778)

Formula Parameters

- Vsample: sample volume added to the reaction system, 0.25 mL
- Vtotal sample: total volume of extract, 10 mL
- Cpr: sample protein concentration, mg/mL
- W: sample mass, g
- T: reaction time, 5 min
- ΔA: A2 - A1

Precautions

1. If yellow crystals precipitate from Reagent I, heat at 90℃ to dissolve before use.
2. If precipitate forms in Reagent II, heat at 70℃ to dissolve before use.
3. The 50T kit can test 24 samples, and the 100T kit can test 48 samples.
4. This product is intended for scientific research by professionals only. It must not be used for clinical diagnosis or treatment, must not be used in food or drugs, and must not be stored in ordinary residences.

5. For your safety and health, wear a lab coat and disposable gloves when operating.

Visual Reference