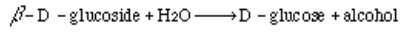
Catalog Number: 100348, 195197

beta-Glucosidase

CAS #: 9001-22-3

beta-Glucosidases catalyze the hydrolysis of glucosides:



Source: *Sweet almonds*

Physical Description: A pale yellow freeze-dried powder

Unit Definition: That amount of enzyme causing the liberation of one microgram of glucose per minute at 35°C

Typical Associated Activity, Contaminants: α Amylase: ~ 0.00007%

Solubility: Dissolves readily at 5 mg/ml in 0.1 M sodium acetate pH 5.0 to give a clear pale yellow solution.

Assay

Method: Salicin is hydrolyzed by beta-glucosidase to yield saligenen and beta-D-glucose. The rate of formation of glucose is measured in a hexokinase/glucose-6-phosphate dehydrogenase system. One unit releases one micromole of glucose per minute at 37°C and pH 5.0 under the specified conditions.

Reagents

- 0.1 M Acetate buffer, pH 5.0
- 1% Salicin solution. Prepare by dissolving 1 gm salicin in 100 ml 0.1 M acetate buffer, pH 5.0. Incubate at 37°C for 6 - 8 minutes before using.
- Glucose reagent system: 0.1 M Tris · HCl buffer pH 7.6, containing hexokinase 1.5 units/ml, ATP: 0.77 *u*mol/ml, NAD: 0.91 *u*mol/ml, and glucose-6-phosphate dehydrogenase 1.9 units/ml.

Enzyme

Dissolve at one mg/ml in reagent grade water. Immediately prior to use dilute to 0.1 - 0.05 mg per ml in reagent grade water.

Procedure

Pipette 1.0 ml of respective enzyme dilutions into a series of numbered test tubes. Include a blank with 1.0 ml reagent grade water. Incubate tubes at 37°C for 6 - 8 minutes to achieve temperature equilibrium. At timed intervals add 4 ml of the salicin solution. Mix well. Incubate each sample exactly 10 minutes then stop reaction at timed intervals by immediately placing each tube in a boiling water bath for at least 5 minutes. Cool in ice bath.

Pipette 3.0 ml Glucose reagent system into cuvettes.

Determine A_{340} of each solution before adding blank or enzyme-salicin reaction mixture. Add 0.1 ml blank and record change in A_{340}. To other cuvettes add 0.1 ml of the heated enzyme-salicin reaction mixture and record change in A_{340}. Record A_{340} until no further change occurs in 3 - 5 minutes. Read final A_{340}.

Calculation

$$\Delta A_{340} = A_{340}(\text{Final}) - A_{340}(\text{Initial})$$

$$\text{Units/mg} = \frac{(\Delta A_{340}\ \text{Sample} - \Delta A_{340}\ \text{Blank}) \times 5 \times 3.1}{6.22 \times 10\ \text{min} \times 0.1 \times \text{mg enzyme in reactions mixture}}$$

Availability:

Catalog Number	Description	Size
100348	β-glucosidase, activity approximately 2500 u/mg material	500 KU 1000 KU 5000 KU
195197	β-glucosidase, activity approximately 1000 u/mg material	5 KU 25 KU 100 KU