FT-MS6550

HRP labeled GS-I Lectin

Product Description

Name :	**Pure *Griffonia simplicifolia* lectin (GS-I), HRP conjugated**
Catalog Number :	FP-MS6550 1 mg purified GS-I FITC / 1 ml Buffer
Purification Procedure :	Gel filtration performed after conjugation to remove free FITC
Carbohydrate Specificity :	Melibiose α-D-Galactose.
Inhibitory Carbohydrate :	α-Galactose.
Activity :	20-30 µg/ml is required to agglutinate fresh type B blood cells. Lectin activity against all blood types increases after neuraminidase treatment of the cells.
Buffer :	0.01M Phosphate - 0.15M NaCl containing 0.5 mM CaCl2, pH 7.2 - 7.4

Storage: Store liquid material frozen in aliquots in amber vials or covered with foil. Avoid freeze thaw cycles. Clarify by centrifugation. No preservatives have been added. Sodiumazide will inactivate the enzyme, peroxidase.

Stability: The liquid material is stable for at least 1 year when stored frozen in aliquots.

Directions for use

Chemical Principle

Peroxidase + H_2O_2 → Complex
Complex + AH_2 (donor) → Peroxidase + H_2O + A (colored)

Assays Reagents

BUFFER: 0.01M Sodium phosphate, pH 6.0.
ENZYME: Dilute with Buffer. Acceptable dilution: 1-2 µg/ml.
DYE: 1% o-dianisidine in methanol prepared fresh daily. Store in amber bottle or wrapped in foil.
SUBSTRATE: Prepare 0.3% H_20_2 solution in deionized or distilled water from stock H_20_2 solution Prior to use dilute to a final concentration 0.003% in Buffer.

Procedure

1. Add 0.05 ml of DYE to 6.0 ml of SUBSTRATE. Add 2.9 ml to Reaction test tube and 2.9 ml to Control test tube.
2. At time=0, add 100µl of diluted ENZYME to Reaction tube and 100µl PBS to Control tube. Mix thoroughly.
3. Measure and record optical density at 460nm (OD460) every 15 seconds for 3 minutes, or take the end point reading after 3 minutes by stopping the reaction with 100µl of concentrated NaN_3.
4. Use this value to determine the rate of change in absorbance per minute.

Enzyme Activity Calculations

One unit of peroxidase activity is that amount of enzyme decomposing 1 µmole of peroxide/minute at 25ºC. 11.3 x 10^3 cm^{-1} is the molar absorbance of H_2O_2.

$$OD460/min = \frac{OD460/3\text{min} - OD\ Control/3\text{minutes}}{3\text{minutes}}$$

$$mg\ enzyme/ml\ reactionmixture = \frac{[enzyme\ dilution]}{30}$$

$$units/mg = \frac{OD460/min}{11.3\ x\ mg\ enzyme/ml\ reactionmixture}$$

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Caution

Due to inhibitory sugar present in the conjugates solution, to dilute the Conjugate 50-100 times with buffer before assay.

References

- **Murphy L.** *et al.,* J. Biol. Chem. *252*, 4739-4742 (1977)
- **Judd W.J.** *et a*l, Vox Sang, 33, 246 (1977)
- **Eckhardt A.,** *et al*, Cancer Res., 42, 2977-2979 (1982)
- **Maddox D.,** *et al*, PNAS, 79, 166-170 (1982)

Related / associated products and documents

See BioSciences Innovations catalogue and e-search tool.

- Lectin List (or search conjugated lectins):
- ConA-Biotin, FP-MS9690; -FITC, FP-47496A; -Cy3, FP-WT8680,
- GS-I-FITC; FP-MS9020
- PNA-FITC, FP-BV4181
- WGA-biotin, FP-MS5730; -SR101, FP-MS9540; -FITC, FP-CE8070

Ordering information

Catalog size quantities and prices may be found at www.interchim.com/
Please inquire for higher quantities (availability, shipment conditions).

For any information, please ask : FluoProbes® / Interchim; Hotline : +33(0)4 70 03 73 06

 Rev.K01E-J01O