PROzyme®

NOTICE: ProZyme was purchased by Agilent in July 2018. Documents for products and product lots manufactured before August 2019 will contain references to ProZyme. For more information about these products and support, go to: **www.agilent.com/en/contact-us.**

Agilent

α(1-3,4) FUCOSIDASE

SPECIFICATIONS

Product Code: GKX-5019
Specific Activity: >1.5 U/mg
Shipped on ice pack for next day delivery.
Store at -20°C.
Formulation: Lyophilized from 50 mM sodium acetate, 3 mg/ml bovine serum albumin (pH 5.0).

Note: 2 Units is equal to 1 Unit as previously reported by Glyko, Inc.

Glyko® α(1-3,4) Fucosidase is extracted from almond meal and purified by affinity and other proprietary chromatographic techniques.

Applications:

- Analysis of fucosylated O-linked and N-linked glycans using sequential digestion with exoglycosidases.
- Analysis and modification of blood group oligosaccharides since it is active towards the Lewis X (Le^X) antigen[1].

PRODUCT DESCRIPTION

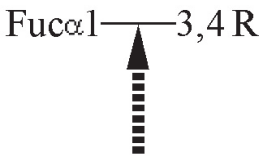
Supplied Reagents (retail packs only)

WS0062 5x Reaction Buffer
(250 mM sodium acetate, pH 5.0)

Purity: No protease activity was detectable after incubation of the enzyme with 0.4% Resorufin-labeled Casein for 18-24 hours at 37°C.

Assays for exoglycosidase contaminants consist of extended incubations with the appropriate substrates. Lot-specific results are reported on the Certificate of Analysis.

Specificity: The enzyme cleaves non-reducing α(1-3 or 1-4)-linked terminal fucose residues[3].



Molecular Weight: ~106,000 daltons, composed of two identical subunits with molecular weight of 54 kD[1] each.

pH Optimum: pH 5.0

Stability: After reconstitution with the incubation buffer supplied with the enzyme, >85% of the original activity is observed after two months at 2–8°C. In the buffer solution at 37°C, the half-life is approximately 80 hours.

Storage: Store lyophilized enzyme at -20°C. Enzyme reconstituted with the provided reaction buffer is stable at 2-8°C for at least two months and may be stored at -20°C for at least six months. Avoid repeated freeze/thaw cycles.

ASSAY

One unit of Glyko® α(1-3,4) Fucosidase is defined as the amount of enzyme which will release one µmole of fucose from lacto-N-fucopentaose II (Cat. No. GKAD-01007) per minute at pH 5.0 and 37°C. The enzyme is not active on pNP-α-fucopyranoside.

Note: Two units is equal to one unit as previously reported by Glyko, Inc.

SUGGESTIONS FOR USE

Before use, briefly centrifuge the vial to ensure all material is at the bottom of the vial. Ensure that reagents, substrates and laboratory-ware are free from contaminants and proteases.

Reconstitute by dissolving the enzyme at an appropriate concentration in a suitable buffer (50 mM sodium acetate, pH 5.0 is recommended as an incubation buffer and is supplied as a 5x concentrate).

The appropriate enzyme concentration depends on the substrate to be digested. Use the enzyme at 0.4 mU/ml for the removal of α(1-3/4) fucose from O-glycans.

Higher enzyme concentrations are required for the removal of α(1-3/4) fucose from complex N-glycans. For example, complete hydrolysis of Fuc-α(1-3)GlcNAc from the outer arms of a di-α(1-3)-fucosylated, di-α(1-2)-fucosylated bi-antennary N-glycan isolated from human parotid gland was obtained at a final substrate concentration of 1 µM and a final enzyme concentration of 6 mU/ml. In general, use the enzyme at 4-8 mU/ml for digestion of N-glycans. Incubate 16-24 hours at 37°C.

REFERENCES

1. Scudder, P. et al. J Biol Chem 265: 16472-16477 (1990).
2. Yoshima, H. et al. Arch Biochem Biophys 194: 394-398 (1979)..

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