

SialEXO[®] 23

FOR RESEARCH
USE ONLY
www.genovis.com

STORE AT
-20°C



SmartEnzymes[™]



GENOVIS

INSTRUCTIONS FOR PRODUCT

SialEXO[®]23, 500 units (G1-SD2-005)
Desialylation of up to 0.5 mg glycoprotein

1 Prepare SialEXO[®] 23

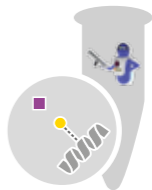
Reconstitute SialEXO 23 in 25 μ l ddH₂O to a concentration of 20 units/ μ l².

**2 Add SialEXO[®] 23**

Add 1 unit SialEXO 23 / 1 μ g glycoprotein or to 5 pmol oligosaccharides.

**3 Desialylation**

Incubate at 37°C for 1 h.



PRODUCT DESCRIPTION

SialEXO 23 is a sialidase for specific removal of α 2-3 linked sialic acids on O- and N-glycans.

SialEXO 23 hydrolyzes sialic acids on glycans under native conditions and displays a high activity in a broad pH range, 7 to 9. The enzyme is derived from *Akkermansia muciniphila* and expressed in *E. coli*. The enzyme contains a His-tag and the molecular weight is 66 kDa.

SialEXO 23 can be used for exoglycosidase sequencing by combining results from a reaction with the sialidase product SialEXO, that efficiently hydrolyzes sialic acids linked with α 2-3, α 2-6 and α 2-8 glycosidic bonds.

Unit Definition

One unit of SialEXO 23 hydrolyzes α 2-3 linked sialic acids from $\geq 90\%$ of 1 μ g glycoprotein (fetalin) when incubated in 20 mM Tris pH 7.5 at 37 °C for 1 h.

Content and Storage

SialEXO 23 is supplied lyophilized in TBS pH 7.6.

SialEXO 23 is shipped at ambient temperature and should be stored at -20°C upon arrival.

After reconstitution, the SialEXO 23 is stable for at least 1 month at +4-8°C.

SialEXO 23 is for R&D use only.

Additional Materials Required

- Reaction buffer¹: 20 mM Tris pH 7.5

Sample Preparation

- Prepare the glycoprotein of interest in reaction buffer to a concentration of 0.1 - 5 mg/ml. Prepare the oligosaccharide of interest in reaction buffer to a concentration of 1 - 5 pmol/ μ l.

Desialylation

1 Prepare SialEXO[®] 23

Reconstitute SialEXO 23 in 25 μ l ddH₂O to a concentration of 20 units/ μ l².

2 Add SialEXO[®] 23

Add 1 unit SialEXO 23 / 1 μ g glycoprotein or to 5 pmol oligosaccharides³.

3 Desialylation

Incubate at 37°C for 1 h⁴.

Optimization of enzyme concentrations and incubation time may be needed for a particular substrate.

Notes

1. *SialEXO 23 displays high activity in buffers at pH 7-9.*
 2. *To prevent microbial contamination, sodium azide can be added to the solution to a final concentration of 0.02 - 0.05% (w/v).*
 3. *A higher enzyme concentration may increase digestion efficiency of individual glycoproteins or oligosaccharides. This requires optimization.*
 4. *Longer incubation times may be required depending on the substrate.*
-

Quality Control

SialEXO 23 is tested to meet specification.

SialEXO 23 is tested for absence of microbial contamination with blood agar plates, Sabouraud dextrose agar plates and fluid thioglycollate medium.

Related Products

Immobilized SialEXO®

Immobilized SialEXO for complete removal of sialic acids with no enzyme in the final preparation

OpeRATOR®

O-glycan specific endoprotease digesting N-terminally of mucin-type O-glycans

GlycOCATCH®

Enrichment of mucin-type O-glycosylated proteins and peptides

OglyZOR®

Hydrolyzes core 1 type O-glycans on native glycoproteins

Legal and Disclaimers

All rights reserved. Genovis products may be covered by one or more patents, trademarks and copyrights owned or controlled by Genovis AB.

For more information about commercial rights, please contact the Genovis team at info@genovis.com.

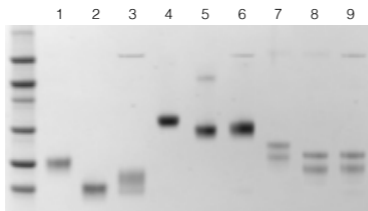
Genovis products are intended for research use only. They are not intended to be used for therapeutic or diagnostic purposes in humans or animals.

OglyZOR®

O-glycosidase Hydrolyzing Core 1 O-glycans

OglyZOR is an O-glycosidase that catalyzes the removal of core 1 and to a limited extent core 3 type O-linked disaccharides from native glycoproteins.

- Hydrolyzes O-glycans
- Specific for core 1 and to a limited extent core 3 type O-glycans



Comparison of the enzymatic activities of OglyZOR and SialEXO to commercially available endoglycosidases and sialidases. All incubations (4 h) were performed according to the manufacturers instructions, and the samples were all separated on SDS-PAGE.

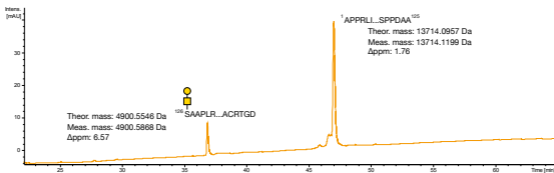
- | | |
|---|---|
| 1. TNF receptor | 6. + Endoglycosidase (<i>E. faecalis</i>) and sialidase (<i>C. perfringens</i>) |
| 2. + SialEXO and OglyZOR | 7. Fetuin |
| 3. + Endoglycosidase (<i>E. faecalis</i>) and sialidase (<i>C. perfringens</i>) | 8. + SialEXO and OglyZOR |
| 4. Etanercept | 9. + Endoglycosidase (<i>E. faecalis</i>) and sialidase (<i>C. perfringens</i>) |
| 5. + SialEXO and OglyZOR | |

OpeRATOR[®]

O-glycan-specific Endoprotease

OpeRATOR is a novel tool for analysis of mucin-type O-glycans on glycoproteins. The protein binds to O-glycans and digests the peptide backbone N-terminally of the S/T glycosylation sites.

- O-glycan-specific, mucin-type
- Requires O-glycans for activity
- Generates glycopeptides with O-glycans and allows for O-glycan profiling and site occupancy determination using mass spectrometry



Erythropoietin (EPO) is a ~30 kDa glycoprotein with one core 1 O-glycan site. The protein was used here as a substrate to demonstrate the specific activity of the OpeRATOR protease. OpeRATOR hydrolyzed the protein N-terminally of the serine O-glycan site, and after reduction of disulfide bridges, the resulting two fragments were separated and intact mass was analyzed by Q-TOF MS using ESI.



US & Canada

Genovis Inc.
245 First Street, Suite 1800
Cambridge, MA 02142
USA

Customer service: 617-444-8421
Order phone (toll free): 855-782-0084
Order fax: 858-524-3006
Email: orders.us@genovis.com

EMEA & Asia

Genovis AB
Box 790
SE-220 07 Lund
Sweden

Customer service: +46 46 10 12 30
Order phone: +46 46 10 12 30
Order fax: +46 46 12 80 20
Email: order@genovis.com