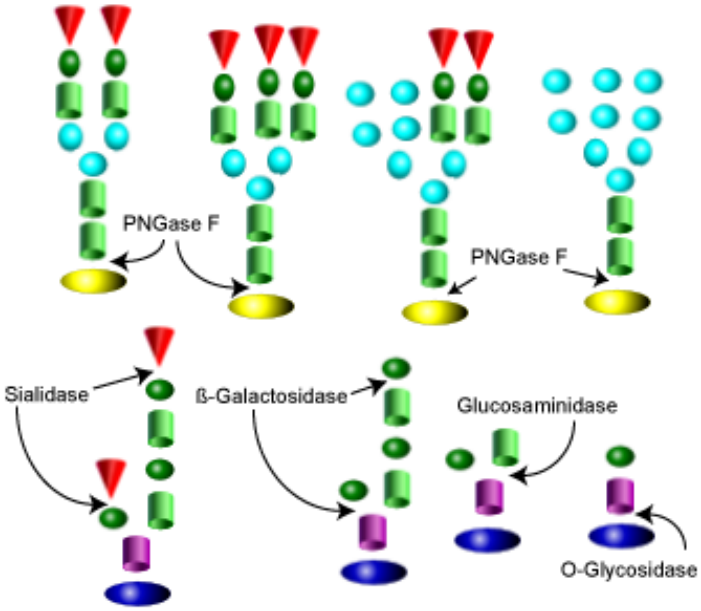

Enzymatic CarboRelease™ Kit

Part Number KE-DG01

Certification of Analysis Lot Number 711.1A

Kit Storage Kits should be stored at 4°C.

Shipping This product should be shipped on frozen packs in an insulated container.

Kit Contents
Kit includes the enzymes, controls, and reagents required to remove all N-linked oligosaccharides and most O-linked sugars. Each kit will deglycosylate more than 2 mg of glycoprotein in 20 reactions.

Enzymes

PNGase F (*E. meningosepticum*)
20 µL - 100 mU
O-Glycosidase (*S. pneumoniae*)
20 µL - 25 mU
Sialidase (*A. ureafaciens*)
20 µL - 100 mU
ß-Galactosidase (*S. pneumoniae*)
20 µL - 60 mU
Glucosaminidase (*S. pneumonia*)
20 µL - 20 mU
refer to enzyme specifications for further details

Other Supplied Reagents

5x Reaction buffer - 200 µL
250 mM sodium phosphate, pH 7
Denaturation Solution - 100 µL
Triton X - 100 µL
Bovine Fetuin (control) - 10 mg/ml

Control
Fetuin is include in this kit as a positive control of the deglycosylation reaction. The concentration of the fetuin is 10 mg/ml .The molecular weight is approximately 48,000 daltons.

Specificity
The Enzymatic CarboRelease Kit will remove all N-linked oligosaccharides and many O-linked oligosaccharides from glycoproteins. N-links (Asn-linked) are removed using the enzyme PNGase F. In addition, all Ser/Thr-linked (O-linked) Gal-(β1-3)-GalNAc-(α1) and all sialic acid substituted Gal-(β1-3)-GalNAc-(α1) will be removed using the combination of Sialidase and O-Glycosidase. The addition of ß-Galactosidase and Glucosaminidase will assist in the deglycosylation of larger O-link structures.

KE-DG01 CarboRelease Kit
Specifications - Protocol

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Directions for Use

1. Mix 10 µl of 5x reaction buffer with up to 100 µg of glycoprotein in 30 µl distilled water in a 1.5 ml tube.
2. Add 2.5 µl denaturation solution. Mix gently and place in boiling water bath for 5 minutes. Chill on ice.
3. Add 2.5 µl of Triton-X.
4. Add 1 µl each of PNGase F, Sialidase, ß-Galactosidase, Glucosaminidase, and O-Glycosidase. Incubate for 3 hours at 37°C.

Note: Denaturation increases the rate of enzyme digestion up to 10 fold. If denaturation is not desired omit step 2-3, add with 5 µl of distilled water and increase incubation time to 24 hours.

The efficiency of deglycosylation can be tested by running a sample on a SDS-PAGE gel.

Warranties and liabilities

QA-Bio warrants that the above product conforms to the attached analytical documents. Should the product fail for reasons other than through misuse QA-Bio will, at its option, replace free of charge or refund the purchase price. This warranty is exclusive and QA-Bio makes no other warrants, expressed or implied, including any implied conditions or warranties of merchantability or fitness for any particular purpose. QA-Bio shall not be liable for any incidental, consequential or contingent damages. This product is intended for *in vitro* research only. *updated 11/24/2017*

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