



## DESCRIPTION

<b>Formula</b>	$C_{37}H_{57}N_{10}O_{21}P_2S$
<b>Molecular Weight</b>	1085.93 Da
<b>Formulation</b>	Lyophilized with Tris, at pH 8.0
<b>Storage &amp; Stability</b>	Store the unopened product at $\leq -20$ °C. Use a manual defrost freezer and avoid repeated freeze-thaw cycles. Good for 12 months from date of receipt.

## APPLICATIONS

- Biotinylation of free glycans as well as glycoproteins and glycolipids.
- Detect specific glycan epitopes on glycoproteins as well as cell surface.

## KEY FEATURES & BENEFITS

- Can be directly introduced into glycoproteins and glycolipids via various fucosyltransferases.
- Can be introduced to live cells for imaging.
- Minimum side effect on target molecules.
- Convenient and User-friendly.

### For Details:

[Wu, ZL. et al., \(2020\) Glycobiology 30:970.](#)

## RELATED REAGENTS

### Click Chemistry

- [GDP-Cy5-Fucose \(ES301\)](#)
- [CMP-Cy5-Sialic Acid \(ES302\)](#)
- [GDP-Azido-Fucose \(ES101\)](#)
- [CMP-Azido-Sialic Acid \(ES102\)](#)
- [UDP-Azido-GalNAc \(ES103\)](#)
- [UDP-Azido-GlcNAc \(ES104\)](#)
- [CMP-9-Biotin-Sialic Acid \(ES201\)](#)

### Enzymes and Detection Reagents

- [Various Fucosyltransferases](#)
- [Streptavidin-HRP \(DY998\)](#)

## SAMPLE PROTOCOL

Protocols are guidelines. Parameters need to be optimized by end users.

### OTHER MATERIALS REQUIRED

- Assay Buffer: 25 mM Tris, 10 mM MnCl<sub>2</sub>, pH 7.5
- Sample glycoprotein
- [Recombinant Fucosyltransferase](#)
- Protein Sample Loading Dye
- SDS-PAGE and Western Blot reagents or equivalent
- TBST Buffer: 25 mM Tris, 137 mM NaCl, 0.1% Tween-20, pH 7.5
- Commercial ECL (Enhanced Chemiluminescence) Reagents
- Nitrocellulose Membrane
- 10% fat-free milk
- [Streptavidin-HRP \(R&D Systems, Catalog # DY998\)](#)

### FINAL ASSAY CONDITIONS PER REACTION

- GDP-Biotin-Fucose ([R&D Systems®, Catalog # ES202](#)): 0.25 nmol
- Recombinant fucosyltransferase: 0.5 µg
- Sample glycoprotein: 1-10 µg

**Table 1. Enzyme selection for Glycan Labeling**

Fucosyltransferase	R&D Systems, Catalog #	Substrate	GDP-Biotin-FucoseTolerance
FUT1	-	Terminal Gal in H antigen	Not determined
FUT2	<a href="#">7770-GT</a>	Terminal Gal in H antigen	Yes
FUT3	<a href="#">4950-GT</a>	GlcNAc in type 1 glycan chain	Yes
FUT4	-	GlcNAc in terminal lactosamine	Yes
FUT5	<a href="#">4949-GT</a>	GlcNAc in terminal lactosamine	Yes
FUT6	-	GlcNAc in terminal lactosamine	Yes
FUT7	<a href="#">6409-GT</a>	GlcNAc in sialylated lactosamine	Yes
FUT8	<a href="#">5768-GT</a>	Core GlcNAc in N-glycans	Yes
FUT9	<a href="#">9347-GT</a>	GlcNAc in non-sialylated lactosamine	Yes
FUT10	-	Unknown	Unknown
FUT11	<a href="#">5964-GT</a>	Unknown	Unknown
POFUT1	<a href="#">7409-GT</a>	Notch receptor	Not determined

### ASSAY PROCEDURE

1. Prepare a reaction mixture by combining a sample glycoprotein (ideally between 1-10 µg), 0.25 nmol GDP-Biotin-Fucose, 0.5 µg of a Recombinant Fucosyltransferase, and add Assay Buffer to the final volume to 30 µL.
2. Prepare a negative control by repeating above but omitting the Fucosyltransferases.
3. Incubate all the reactions and controls at 37 °C for 60 minutes.
4. Stop the reactions and controls by adding appropriate volume of Protein Sample Loading Dye to each reaction.
5. Separate the reactions and controls by SDS-PAGE.
6. Blot the gel to a nitrocellulose membrane.
7. Block the blot with 10% fat-free milk for 5 minutes.
8. Thoroughly wash the membrane with TBST Buffer by changing buffer three times for a total of 30 minutes.
9. Incubate the blot with 25 ng/mL Streptavidin-HRP in 30 mL TBST buffer for 30 minutes.
10. Thoroughly wash the membrane with TBST Buffer by changing buffer three times for a total of 45 minutes.
11. Detect with Commercial ECL (Enhanced Chemiluminescence) Reagents.