

CERTIFICATE OF ANALYSIS

PRODUCT NAME: SIALIDASE S™ (His-tagged, recombinant from *Streptococcus pneumoniae*

expressed in E. coli)

PRODUCT CODE: GK80021

LOT NUMBER: 163 003-1

FORMULATION: 20 mM Tris-HCl, 25 mM NaCl (pH 7.5) upon reconstitution as indicated

RECONSTITUTION: Dissolve the lyophilizate in 726 µl of ultrapure water to obtain the described

formulation.

SUGGESTIONS FOR USE: Up to ~1 nanomole of substrate can be cleaved in a 20 µl reaction volume.

In the standard reaction use: $4 \,\mu l$ of 5x Reaction Buffer B, substrate and water in a combined volume of $14 \,\mu l$, and $2 \,\mu l$ of Sialidase S. Incubate for one hour at 37° C. To cleave more than one nanomole of substrate, increase the reaction volume and enzyme proportionally. Difficult substrates may require optimization to find the correct amount of Sialidase S to add.

STORAGE: -20°C until redissolved. Store redissolved enzyme at 2 – 8°C for up to 2

months.

PACK SIZE: 3 Units (equivalent to 1 Unit of previous product GK80020)

COMPONENTS

Component	Quantity/Pack	Lot No.	Exp. Date
GK80021 Sialidase S (3 Units)	1 each	163 003	Jun 2018*
WS0049 5x Reaction Buffer (1 ml) [250 m <i>M</i> Sodium Phosphate, pH 6.0]	1 each	W160035	Mar 2020

^{*}Extended from prior exp. date based on re-assay

QUALITY CONTROL:

1) Enzyme Specific Activity¹: Pass (Specification: ≥30 U/mg)

2) Protease Assay²: Pass (Specification: "Not Detectable")

3) Contaminants³ Pass (Specification: $\leq 0.001\%$)

Authorized Signature

- 1. One unit is defined as the amount of enzyme required to catalyze the release of 1 μ mole of p-nitrophenol from pNP- α -d-N-acetylneuraminic acid per minute at pH 5.5 and 37°C.
- 2. No protease activity was detectable after incubation of the enzyme with 0.2 mg resorufin-labeled casein for ~18 hours at 37°C based on Schickaneder E, Hösel W, von der Eltz H, Geuß U. Casein-resorufin, a new substrate for a highly sensitive protease assay. Fresenius Z. Anal Chem. 1988 330:360.
- 3. The absence of exoglycosidase contaminants was confirmed by extended incubations with the corresponding pNP-glycosides: α -fucosidase, β -fucosidase, α -mannosidase, β -mannosidase, β -n-acetylhexosaminidase, α -N-acetylgalactosaminidase, α -galactosidase, β -galactosidase, α -glucosidase and β -xylosidase.