

## New England Biolabs Certificate of Analysis

**Product Name:**  $\alpha$ -N-Acetylgalactosaminidase  
**Catalog Number:** P0734L  
**Concentration:** 20,000 U/ml  
**Unit Definition:** One unit is defined as the amount of enzyme required to cleave > 95% of the terminal  $\alpha$ -D-N-acetylgalactosamine from 1 nmol (GalNAc $\alpha$ 1-3)(Fuc $\alpha$ 1-2)Gal $\beta$ 1-4Glc-7-amino-4-methyl-coumarin (AMC), in 1 hour at 37°C in a total reaction volume of 10  $\mu$ l.  
**Lot Number:** 10037022  
**Expiration Date:** 02/2021  
**Storage Temperature:** -20°C  
**Storage Conditions:** 50 mM NaCl, 20 mM Tris-HCl, 1 mM EDTA, (pH 7.5 @ 25°C)  
**Specification Version:** PS-P0734S/L v1.0

### $\alpha$ -N-Acetylgalactosaminidase Component List

NEB Part Number	Component Description	Lot Number	Individual QC Result
P0734LVIAL	$\alpha$ -N-Acetylgalactosaminidase	10037021	Pass
B9001SVIAL	Purified BSA	10014762	Pass
B1727SVIAL	10X GlycoBuffer 1	10011906	Pass

Assay Name/Specification	Lot # 10037022
<b>Glycosidase Activity (<math>\alpha</math>1-6 Galactosidase)</b> A 10 $\mu$ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled $\alpha$ -Galactosidase substrate (Gal $\alpha$ 1-6Gal $\alpha$ 1-6Glc $\alpha$ 1-2Fru-AMC) and 20 units of $\alpha$ -N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
<b>Glycosidase Activity (<math>\alpha</math>1-6 Mannosidase)</b> A 10 $\mu$ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled $\alpha$ -Mannosidase substrate (Man $\alpha$ 1-6Man $\alpha$ 1-6(Man $\alpha$ 1-3)Man-AMC) and 20 units of $\alpha$ -N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass
<b>Glycosidase Activity (<math>\beta</math>-Mannosidase)</b> A 10 $\mu$ l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled $\beta$ -Mannosidase substrate (Man $\beta$ 1-4Man $\beta$ 1-4Man-AMC) and 20 units of $\alpha$ -N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.	Pass

Assay Name/Specification	Lot # 10037022
<p><b>Glycosidase Activity (<math>\beta</math>-N-Acetylgalactosaminidase)</b> A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\beta</math>-N-Acetylgalactosaminidase substrate (GalNAc<math>\beta</math>1-4Gal<math>\beta</math>1-4Glc-AMC) and 20 units of <math>\alpha</math>-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (<math>\beta</math>-N-Acetylglucosaminidase)</b> A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\beta</math>-N-Acetylglucosaminidase substrate (GlcNAc<math>\beta</math>1-4GlcNAc<math>\beta</math>1-4GlcNAc-AMC) and 20 units of <math>\alpha</math>-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (<math>\beta</math>-Xylosidase)</b> A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\beta</math>-Xylosidase substrate (Xyl<math>\beta</math>1-4Xyl<math>\beta</math>1-4Xyl<math>\beta</math>1-4Xyl-AMC) and 20 units of <math>\alpha</math>-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (<math>\beta</math>1-3 Galactosidase)</b> A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\beta</math>-Galactosidase substrate (Gal<math>\beta</math>1-3GlcNAc<math>\beta</math>1-4Gal<math>\beta</math>1-4Glc-AMC) and 20 units of <math>\alpha</math>-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (<math>\beta</math>1-4 Galactosidase)</b> A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\beta</math>-Galactosidase substrate (Gal<math>\beta</math>1-4GlcNAc<math>\beta</math>1-3Gal<math>\beta</math>1-4Glc-AMC) and 20 units of <math>\alpha</math>-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Protease Activity (SDS-PAGE)</b> A 20 <math>\mu</math>l reaction in 1X Glyco Buffer 1 containing 24 <math>\mu</math>g of a standard mixture of proteins and a minimum of 100 units of <math>\alpha</math>-N-Acetylgalactosaminidase incubated for 20 hours at 37°C, results in no detectable degradation of the protein mixture as determined by SDS-PAGE with Coomassie Blue detection.</p>	<b>Pass</b>
<p><b>Protein Purity Assay (SDS-PAGE)</b> <math>\alpha</math>-N-Acetylgalactosaminidase is <math>\geq</math> 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (<math>\alpha</math>1-3 Mannosidase)</b></p>	<b>Pass</b>

Assay Name/Specification	Lot # 10037022
<p>A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Mannosidase substrate (Manα1-3Manβ1-4GlcNAc-AMC) and 20 units of α-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	
<p><b>Glycosidase Activity (Endo F1, F2, H)</b> A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F1, F2, H substrate (Dansylated invertase high mannose) and 20 units of α-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (Endo F2, F3)</b> A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled Endo F2, F3 substrate (Dansylated fibrinogen biantennary) and 20 units of α-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (PNGase F)</b> A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled PNGase F substrate (Fluoresceinated fetuin triantennary) and 20 units of α-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (α-Glucosidase)</b> A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Glucosidase substrate (Glcα1-6Glcα1-4Glc-AMC) and 20 units of α-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (α-Neuraminidase)</b> A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Neuraminidase substrate (Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-AMC) and 20 units of α-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (α1-2 Fucosidase)</b> A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled α-Fucosidase substrate (Fucα1-2Galβ1-4Glc-AMC) and 20 units of α-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<b>Pass</b>
<p><b>Glycosidase Activity (α1-3 Fucosidase)</b> A 10 µl reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled</p>	<b>Pass</b>

Assay Name/Specification	Lot # 10037022
<p><math>\alpha</math>-Fucosidase substrate (Fuc<math>\alpha</math>1-3Gal<math>\beta</math>1-4GlcNAc<math>\beta</math>1-3Gal<math>\beta</math>1-4Glc-AMC) and 20 units of <math>\alpha</math>-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p> <p><b>Glycosidase Activity (<math>\alpha</math>1-3 Galactosidase)</b> A 10 <math>\mu</math>l reaction in Glyco Buffer 1 containing 1 nM of fluorescently-labeled <math>\alpha</math>-Galactosidase substrate (Gal<math>\alpha</math>1-3Gal<math>\beta</math>1-4GlcNAc-AMC) and 20 units of <math>\alpha</math>-N-Acetylgalactosaminidase incubated for 20 hours at 37°C results in no detectable activity as determined by thin layer chromatography.</p>	<p><b>Pass</b></p>

This product has been tested and shown to be in compliance with all specifications.



Alicia Bielik  
Production Scientist  
22 Jun 2018



Michael Tonello  
Packaging Quality Control Inspector  
13 Mar 2019