



Certificate of Analysis

LudgerZyme Acetyl Esterase Kit

Cat. #: LZ-ACASE-KIT

Batch #: B617-04

Size: 1 set of enzyme per kit

This kit conforms to the specifications given in Ludger document # LZ-ACASE-KIT-Guide-v1.0

Each kit contains the following components:

| Quantity per Kit | Cat # | Batch # | Component Name |
|------------------|----------------------|---------|-----------------------------------|
| 1 | LZ-ACASE-50 | B617-02 | LudgerZyme Acetyl Esterase |
| 1 | LZ-ACASE- BUFFX10 | B617-03 | LudgerZyme Acetyl Esterase buffer |

Expiry Date: Nov 2022

Acetyl esterase assay: 1µL of 0.16mg/mL LudgerZyme Acetyl Esterase (LZ-ACASE-50) was incubated with 1µL of 1.18µg/µL Neu5,9Ac2 (B56G-X02), in a 10µL reaction containing 1µL 10X LudgerZyme Acetyl Esterase buffer (LZ-ACASE-BUFFX10). The reaction was incubated for 6 hours at 37°C. The reaction products were dried and DMB labelled (LT-KDMB-A1). DMB labelled products were analysed by hydrophobic-HPLC using R1 column (LudgerSep R1).

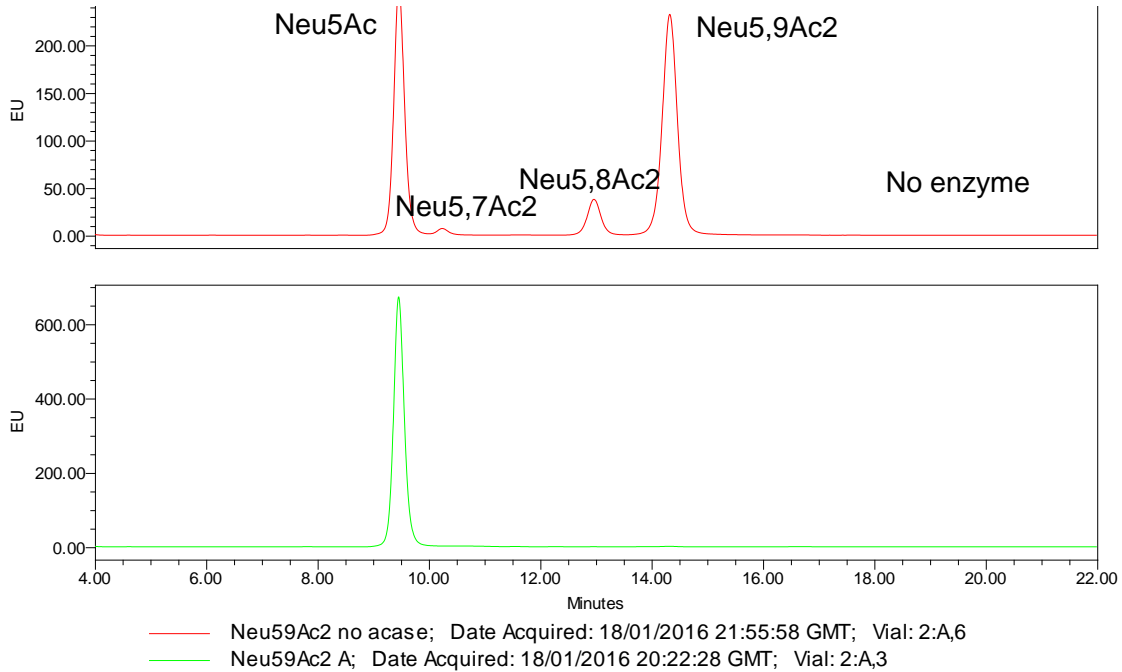


Figure 1: HPLC profile of undigested and DMB labelled Neu5,9Ac2 (Batch: B56G-X02) digested using LZ-ACASE-KIT (Batch: B617-04)

HPLC Running Conditions:

Column: LudgerSep-R1 (LS-R1-4.6x150)

Flow: 0.5 mL/min

Solvent A: Acetonitrile:Methanol:Water (9:7:94) Solvent B: 100 % acetonitrile Temperature: 30 °C.

Gradient: None. Isocratic run.

Detector: Waters 2475 FLR Detector

Excitation wavelength: 373 nm

Emission wavelength: 448 nm

No contaminating exoglycosidase or endoglycosidase activities were detected (ND) with the following substrates:

α-sialidase and endoglycosidase F3:

2AB-FA2G2S2 (CAB-A2F-01) ND

β-galactosidase and α-fucosidase:

2AB-FA2G2 (CAB-NA2F-01) ND

α-mannosidase and endoglycosidase F1 & F2:

2AB-mannose (CAB-MAN5-01, CAB-MAN6-01, CAB-MAN7-01, CAB-MAN8-01 and CAB-MAN9-01) ND