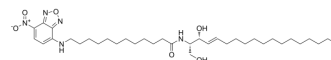


C12-NBD-ceramide

Cat. No.:	HY-141575
CAS No.:	202850-01-9
Molecular Formula:	C ₃₆ H ₆₁ N ₅ O ₆
Molecular Weight:	659.9
Target:	Fluorescent Dye
Pathway:	Others
Storage:	Please store the product under the recommended conditions in the Certificate of Analysis.



BIOLOGICAL ACTIVITY

Description	C12-NBD-ceramide is a fluorescent analogue of ceramide, it can be used as a substrate in ceramidase assays ^{[1][2]} .
In Vitro	<p>Guidelines (Following is our recommended protocol. This protocol only provides a guideline, and should be modified according to your specific needs).</p> <p>In this experiment, C12-NBD-ceramide is used as a fluorescent nCDase substrate and reaction products will be separated by reverse-phase HPLC^[2]:</p> <ol style="list-style-type: none"> 20 μM substrate, 1 nM nCDase in 75 mM NaCl, 25 mM HEPES (pH 8.0), and 0.4% Triton X-100, for 2 h at 37°C in a final volume of 100 μL. Extract samples with 1:1 chloroform-methanol dries under nitrogen gas, and resuspends in 60 μL of HPLC mobile phase B. Separates reaction products by reverse-phase HPLC using a Spectra 3 μm C8SR column (3 μm particle, 3.0 × 150 mm). Mobile phase A contains 0.2% formic acid and 1 mM ammonium formate in HPLC-grade water. Mobile phase B contains 0.2% formic acid (IL) and 1 mM ammonium formate in HPLC-grade methanol. FRET-based assay using 20 μM substrate ES.173.cds, 50 ng of nCDase, 75 mM NaCl, 15 mM sodium phosphate (pH 7.4), and 0.3% Triton X-100 for 3 h in 96 black well plates. Measure resorufin fluorescence (Ex=347 nm, Em=430 and 530 nm). <p>MCE has not independently confirmed the accuracy of these methods. They are for reference only.</p>

REFERENCES

- [1]. Tani M, et al. Specific and sensitive assay for alkaline and neutral ceramidases involving C12-NBD-ceramide. *J Biochem.* 1999 Apr;125(4):746-9.
- [2]. Otsuka Y, et al. Identification of Small-Molecule Inhibitors of Neutral Ceramidase (nCDase) via Target-Based High-Throughput Screening. *SLAS Discov.* 2021 Jan;26(1):113-121.

Caution: Product has not been fully validated for medical applications. For research use only.

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