$GlcC_{14}C_{18}$

Glucosyl-6-tetradecyloctadecanoate; Mincle ligand

Catalog code: tlrl-gcc

https://www.invivogen.com/glc-c14c18

For research use only

Version 18J31-MM

PRODUCT INFORMATION

Contents

2 x 1 mg Glucosyl-6-tetradecyloctadecanoate ($GlcC_{14}C_{18}$)

Storage and stability

- $GlcC_{14}C_{18}$ is provided as a translucent film and shipped at room temperature. Upon receipt, store product at -20 °C.
- Store resuspended product in an upright position at -20 °C. Resuspended product is stable for 6 months when properly stored.

Note: To avoid possible leakage or evaporation, we recommend to wrap plastic film around the lid of the vial containing the resuspended product.

Quality control

- Purity: ≥95% (UHPLC)
- The biological activity has been tested using cellular assays.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

DESCRIPTION

$GlcC_{14}C_{18}$ is a C_6-branched glycolipid that activates the macrophage-inducible C-type lectin (Mincle) receptor. This glucose monoester was developed using a rational design to obtain a structurally simple molecule that binds and activates Mincle[1]. $GlcC_{14}C_{18}$ effectively activates both human and murine Mincle. It has activity similar to the extensively studied Mincle agonists trehalose-6,6-dimycolate (TDM, also known as cord factor) and its analog trehalose-6,6-dibehenate (TDB)[1]. Upon $GlcC_{14}C_{18}$ recognition, Mincle interacts with the Fc receptor common γ-chain (FcRγ) triggering signaling through Syk-CARD9-dependent NF-κB activation, ultimately leading to the production of Th1/Th17 polarizing cytokines and chemokines[1-3]. Importantly, $GlcC_{14}C_{18}$ displays less toxicity on human monocytes and monocyte-derived dendritic cells *in vitro* than TDB[1].

1. Decout A. *et al.*, 2017. Rational design of adjuvants targeting the C-type lectin Mincle. PNAS. 114(10):2675-80. **2. Patin EC. *et al.*, 2017.** Macrophage Inducible C-Type Lectin As a Multifunctional Player in Immunity. Front Immunol. 8:861. **3. Williams SJ., 2017.** Sensing Lipids with Mincle: Structure and Function. Front Immunol. 8:1662.

CHEMICAL PROPERTIES

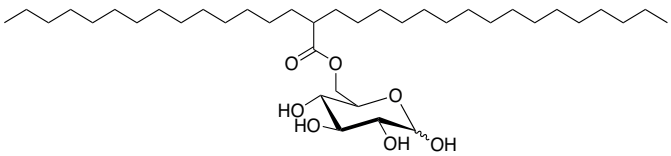
CAS number: 2097365-59-6

Formula: $C_{38}H_{74}O_7$

Molecular weight: 643 g/mol

Solubility: 1 mg/ml isopropanol or 2 mg/ml DSMO

Chemical structure:



METHODS

Preparation of stock suspension (1 mg/ml)

- Add 1 ml of isopropanol (**not provided**) to 1 mg of $GlcC_{14}C_{18}$.
- Heat at 60 °C for 2 minutes, sonicate for 20 seconds and vortex until completely dissolved.
- Use immediately or store at -20 °C.
- Following storage at -20 °C, allow to reach room temperature and vortex before use.
- Prepare dilutions with isopropanol.

Working concentration: 10 ng/ml - 1 µg/ml

Mincle activation using $GlcC_{14}C_{18}$

$GlcC_{14}C_{18}$-induced activation can be studied using HEK-Blue™ Mincle reporter cells, which are HEK293-derived cells stably transfected with the Mincle gene and other genes of the Mincle-NF-κB signaling pathway. They also stably express an NF-κB-inducible secreted embryonic alkaline phosphatase (SEAP) reporter gene. Mincle activation is assessed by measuring SEAP activity using SEAP detection reagents such as QUANTI-Blue™ or HEK-Blue™ Detection.

For more information visit: https://www.invivogen.com/hek-blue-clr

Day 1

1. Dispense 20 µl of the $GlcC_{14}C_{18}$ suspension at various concentrations (10 ng/ml to 1 µg/ml final concentration) per well in a 96-well plate.
2. Ensure that the $GlcC_{14}C_{18}$ suspension is evenly distributed on the surface of the well.
3. Allow to dry for 1 hour at room temperature (15-25 °C).
4. Prepare a cell suspension (~280,000 cells per ml) and add 180 µl of this suspension (~50,000 cells) to each $GlcC_{14}C_{18}$-containing well.
5. Incubate the cells for 20-24 hours at 37 °C and 5% CO_2.

Day 2

1. Prepare QUANTI-Blue™ following the instructions on the data sheet.
2. Add 20 µl of supernatant to each well containing 180 µl of QUANTI-Blue™ Solution.
3. Incubate the plate at 37 °C for 30 minutes to 6 hours.
4. Determine SEAP levels using a spectrophotometer at 620-655 nm.

RELATED PRODUCTS

Product	Catalog Code
HEK-Blue™ hMincle	Inquire
HEK-Blue™ mMincle	hkb-mmcl
HKMT (heat-killed *M. tuberculosis*)	tlrl-hkmt-1
TDB (Trehalose-6,6-dibehenate)	tlrl-tdb
TDM (Trehalose-6,6-dimycolate)	tlrl-tdm
QUANTI-Blue™ Solution	rep-qbs
HEK-Blue™ Detection	hb-det2

TECHNICAL SUPPORT
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