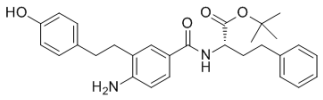
Neoseptin 3

Cat. No.:	HY-U00435
CAS No.:	1622863-21-1
Molecular Formula:	$C_{29}H_{34}N_2O_4$
Molecular Weight:	474.59
Target:	Toll-like Receptor (TLR)
Pathway:	Immunology/Inflammation
Storage:	Please store the product under the recommended conditions in the COA.



BIOLOGICAL ACTIVITY

Description	Neoseptin 3 is a Toll-like receptor 4/myeloid differentiation factor 2 (**mTLR4/MD-2**) agonist with an **EC_{50}** of 18.5 μM.
IC_{50} & Target	EC50: 18.5 μM (mTLR4/MD-2) [1]
In Vitro	Neoseptin 3 is a Toll-like receptor 4/myeloid differentiation factor 2 (mTLR4/MD-2) agonist with an EC_{50} of 18.5 μM. Neoseptin-3 induces TNFα production by macrophages in a concentration-dependent manner. Neoseptin-3 induces phosphorylation of IκB kinases α (IKKα), IKKβ, p38, c-Jun N-terminal kinase (JNK), and ERK, and degradation of IκBα, consistent with activation of MAPK and canonical NF-κB signaling. TANK-binding kinase 1 (TBK1) and IRF3 phosphorylation also increase in response to Neoseptin-3[1].

PROTOCOL

Kinase Assay [1]	**HEK293T cells** are transfected with an NF-κB-dependent luciferase reporter plasmid and clones with stable expression are selected by culture in DMEM containing puromycin. Cells are cotransfected with constructs for mouse or human TLR4 plus mouse or human MD-2, and 2 d later are stimulated with **50 μM Neoseptin-3** or 1 μg/mL LPS **for 6 h**. Cells are lysed, and luciferase activity is measured[1]. MCE has not independently confirmed the accuracy of these methods. They are for reference only.
Cell Assay [1]	Cells are seeded onto 96-well plates at **1×10^5 cells per well** and stimulated with **Neoseptin-3** [**dissolved in DMSO**; final DMSO concentrations (≤0.2%) are kept constant in all experiments] **for 4 h**. Mouse TNFα, IL-6, or IFN-β, or human TNFα in the supernatants are measured by ELISA kits according to the manufacturer's instructions. Unless otherwise indicated, **mouse cells are from wild-type C57BL/6J mice**[1]. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

REFERENCES

[1]. Wang Y, et al. TLR4/MD-2 activation by a synthetic agonist with no similarity to LPS. Proc Natl Acad Sci U S A. 2016 Feb 16;113(7):E884-93.

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

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