

Murabutide

Synthetic derivative of muramylpeptide; NOD2 ligand

Catalog code: tlrl-mbt

<https://www.invivogen.com/murabutide>

For research use only

Version 23J16-MM

PRODUCT INFORMATION

Contents

- 5 mg Murabutide
- 1.5 ml endotoxin-free water

Storage and stability

- Murabutide is shipped at room temperature. Upon receipt, store at -20°C.
- Upon resuspension, prepare aliquots of Murabutide and store at -20°C. Resuspended product is stable for 6 months at -20°C when properly stored. Avoid repeated freeze-thaw cycles.

Quality control

- Activation of NOD2 has been confirmed using HEK-Blue™ NOD2 cells.
- The absence of NOD1 activity has been confirmed using HEK-Blue™ NOD1 cells.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

DESCRIPTION

Murabutide is a safe synthetic immunomodulator derived from muramyl dipeptide (MDP), the smallest bioactive unit of bacterial peptidoglycan. It was developed as a safe alternative to MDP for use as an immunomodulator after MDP was found to be too toxic to be used as an adjuvant in humans. Murabutide possesses all of the immunomodulatory properties of its parent molecule, MDP, without the associated toxicity¹⁻⁴.

Murabutide exerts its effect through the activation of NOD2. Upon agonist binding, NOD2 oligomerizes and signals via RIP2 kinase. Once activated, RIP2 mediates ubiquitination of NEMO/IKK γ leading to the activation of NF- κ B and the production of inflammatory cytokines. Furthermore, poly-ubiquitinated RIP2 recruits TAK1, which leads to IKK complex activation and the activation of MAPKs. This signaling involves the adaptor protein CARD9. It has been reported that murabutide does not transduce the signal through TLR2 or TLR4⁵.

Murabutide induces the activation of NF- κ B in HEK-Blue™ NOD2 cells, but no NF- κ B was detectable in HEK-Blue™ TLR cells.

1. Rubino S.J. *et al.*, 2013. Identification of a synthetic muramyl peptide derivative with enhanced Nod2 stimulatory capacity. *Innate Immun.* 19(5):493-503. 2. Jakopin Z., 2013. Murabutide revisited: a review of its pleiotropic biological effects. *Curr Med Chem.* 20(16):2068-79. 3. Chedid L.A. *et al.*, 1982. Biological activity of a new synthetic muramyl peptide adjuvant devoid of pyrogenicity. *Infect Immun.* 35(2):417-24. 4. Krueger J.M. *et al.*, 1984. Muramyl peptides. Variation of somnogenic activity with structure. *J Exp Med.* 159(1):68-76. 5. Vidal V.F. *et al.*, 2001. Macrophage stimulation with Murabutide, an HIV-suppressive muramyl peptide derivative, selectively activates extracellular signal-regulated kinases 1 and 2, C/EBP β and STAT1: role of CD14 and Toll-like receptors 2 and 4. *Eur J Immunol.* 31(7):1962-71.

PRODUCT PROPERTIES

Synonym: N-Acetyl-muramyl-L-Alanyl-D-Glutamin-n-butyl-ester

CAS number: 74817-61-1

Formula: C₂₃H₄₀N₄O₁₁

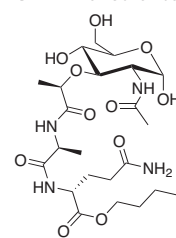
Molecular weight: 548.58 g/mol

Specificity: NOD2

Solubility: 5 mg/ml in water

Working concentration: 10 ng/ml-10 μ g/ml

Chemical structure:



METHODS

Preparation of stock solution (5 mg/ml)

1. Add 1 ml of endotoxin-free water (provided) to the vial containing 5 mg of Murabutide and vortex to solubilize.

NOD2 activation using MDP

MDP can be used to activate NOD2 in cells expressing this receptor, such as HEK-Blue™ NOD2 cells. These cells express the human or murine NOD2 gene and an NF- κ B inducible SEAP reporter gene. Levels of SEAP can be easily determined using HEK-Blue™ Detection, a cell culture medium that allows the detection of SEAP as the reporter protein is secreted by the cells. For more information, visit: <https://www.invivogen.com/hek-blue-nod>.

1. Dispense 20 μ l of MDP at various concentrations (10 ng/ml-10 μ g/ml) per well of a 96-well plate.
2. Prepare a cell suspension ~280,000 cells per ml in HEK-Blue™ Detection.
3. Add 180 μ l of the cell suspension (~50,000 cells) to each MDP-containing well.
4. Incubate the plate for 6-24 h at 37°C, 5% CO₂.
5. Determine SEAP levels using a spectrophotometer at 620-655 nm.

RELATED PRODUCTS

Product	Description	Cat. Code
HEK-Blue™ Detection	SEAP Detection reagent	hb-det2
HEK-Blue™ hNOD2 Cells	Human NOD2 reporter cells	hkb-hnod2
HEK-Blue™ mNOD2 Cells	Murine NOD2 reporter cells	hkb-mnod2
MDP	NOD2 Agonist	tlrl-mdp
MDP Control	L-L isomer, negative control	tlrl-mdpcl

TECHNICAL SUPPORT

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