ODN 1826 Control (ODN 2138)

Negative control oligonucleotide for mouse TLR9 ligand ODN1826

Catalog code: tlrl-1826c-1

https://www.invivogen.com/odn1826-control

For research use only

Version 21L21-MM

PRODUCT INFORMATION

Contents

- 1 mg (**157.10 nmol**) of ODN 1826 Control (ODN 2138) provided lyophilized

Note: ODN 1826 Control (ODN 2138) is sterile filtered prior to lyophilization.

- 1.5 ml endotoxin-free water

ODN 1826 Control (ODN 2138) sequence

5'- tcc atg agc ttc ctg agc tt -3' (20 mer)

Note: Bases are phosphorothioate (nuclease resistant).

Molecular weight: 6364 g/mol

Storage and stability

- ODN 1826 Control (ODN 2138) is shipped at room temperature. Upon receipt, store at -20°C.
- Upon resuspension, prepare aliquots of ODN 1826 Control (ODN 2138) 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

- The absence of stimulatory activity has been confirmed using HEK-Blue™ TLR9 cells.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

DESCRIPTION

CpG ODNs are synthetic oligonucleotides that contain unmethylated CpG dinucleotides in particular sequence contexts (CpG motifs)[1]. These CpG motifs are present at a 20-fold greater frequency in bacterial DNA compared to mammalian DNA. CpG ODNs are recognized by Toll-like receptor 9 (TLR9) leading to strong immunostimulatory effects[2]. Three classes of CpG ODNs have been identified, classes A, B and C, which differ in their immunestimulatory activities[3-4]. Class B CpG ODNs contain a full phosphorothioate backbone with one or more CpG dinucleotides. They strongly activate B cells but stimulate weakly IFN-α secretion.

ODN 1826 Control (also known as ODN 2138) contains GpC dinucleotides instead of CpGs and can be used as a negative control together with ODN1826 (Class B CpG ODN).

1. Krieg, A. *et al.*, 1995. CpG motifs in bacterial DNA trigger direct B-cell activation. Nature, 374:546-9. **2. Bauer, S. *et al.*, 2001.** Human TLR9 confers responsiveness to bacterial DNA via species-specific CpG motif recognition. PNAS, 98:9237-42. **3. Krug A. *et al.*, 2001.** Identification of CpG oligonucleotide sequences with high induction of IFN-alpha/beta in plasmacytoid dendritic cells. Eur J Immunol, 31:2154-63. **4. Marshall J. *et al.*, 2005.** Superior activity of the type C class of ISS in vitro and in vivo across multiple species. DNA Cell Biol. 24(2):63-72.

METHODS

Preparation of ODN solution (500 µM)

- Add 315 µl of endotoxin-free water (provided) to 1 mg vial of ODN 1826 Control (ODN 2138).
- Vortex until completely dissolved. Prepare aliquots and store at -20°C.

TLR9 stimulation

ODN 1826 Control (ODN 2138) can be used as a control ODN to study the stimulatory effect of ODN 1826 on TLR9 in HEK-Blue™ TLR9 cells. These cells stably overexpress the TLR9 gene and an NF-kB-inducible secreted embryonic alkaline phosphatase (SEAP) reporter gene.

For more information, visit: https://www.invivogen.com/hek-blue-tlr9.

Below is a protocol to study TLR9 stimulation using HEK-Blue™ TLR9 cells in a 96-well plate.

Note: Use ODN 1826 Control (ODN 2138) at the same concentration as the CpG-containing ODN 1826.

1. Dispense 20 µl of stimulatory or control ODN per well of a 96-well plate.
2. Prepare cell suspension of HEK-Blue™ TLR9 cells according to the data sheet.
3. Add HEK-Blue™ TLR9 cells (4-8 x10^4) to each ODN-containing well.
4. Incubate for 6-24 h at 37°C, 5% CO_2.
5. Determine TLR9 stimulation by assessing cytokine expression using ELISA, or SEAP expression using QUANTI-Blue™ Solution, a SEAP detection medium.

RELATED PRODUCTS

Product	Description	Cat. Code
HEK-Blue™ mTLR9 cells	Murine TLR9 reporter cells	hkb-mtlr9
ODN 1826	Stimulatory ODN	tlrl-1826
QUANTI-Blue™ Solution	SEAP detection medium	rep-qbs

TECHNICAL SUPPORT
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