

ODN 2006 Control (ODN 2137)

Negative control oligonucleotide for human TLR9 ligand ODN 2006 (ODN 7909)

Catalog code: tlr1-2006c-1

<https://www.invivogen.com/odn2006-control>

For research use only

Version 21L21-MM

PRODUCT INFORMATION

Contents

- 1 mg (129.8 nmol) of ODN 2006 Control (ODN 2137) provided lyophilized

Note: ODN 2006 Control (ODN 2137) is sterile filtered prior to lyophilization.

- 1.5 ml endotoxin-free water

ODN 2006 Control (ODN 2137) sequence

5'-tgctgctttgtgctttgtgctt-3' (24 mer)

Note: Bases are phosphorothioate (nuclease resistant).

Molecular weight: 7698 g/mol

Storage and stability

- ODN 2006 Control (ODN 2137) is shipped at room temperature. Upon receipt, store at -20°C.

- Upon resuspension, prepare aliquots of ODN 2006 Control (ODN 2137) 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 effects². Three classes of CpG ODNs have been identified, classes A, B and C, which differ in their immunostimulatory activities³⁻⁴. 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 2006 Control (also known as ODN 2137) contains GpC dinucleotides instead of CpGs and can be used as a negative control together with ODN 2006 (Class B CpG ODN).

Note: In some cell types, ODN 2006 Control may stimulate cell activity, including the production of cytokines²⁻⁴.

1. Krieg A.M. et al., 1995. CpG motifs in bacterial DNA trigger direct B-cell activation. Nature, 374:546-9. 2. Reid G. et al., 2005. CpG stimulation of precursor B-lineage acute lymphoblastic leukemia induces a distinct change in costimulatory molecule expression and shifts allogeneic T cells toward a Th1 response. Blood, 105:3641-7. 3. Dar A. et al., 2008. Attenuated cytokine responses in porcine lymph node cells stimulated with CpG DNA are associated with low frequency of IFN α -producing cells and TLR9 mRNA expression. Vet Immunol Immunopathol, 123:324-36. 4. Voller J. et al., 2002. Highly immunostimulatory CpG-free oligodeoxynucleotides for activation of human leukocytes. Antisense Nucleic Acid Drug Dev, 12:165-75.

TECHNICAL SUPPORT

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METHODS

Preparation of ODN solution (500 μ M)

• Add 260 μ l of endotoxin-free water (provided) to 1 mg vial of ODN 2006 Control (ODN 2137).

• Vortex until completely dissolved. Prepare aliquots and store at -20°C.

TLR9 stimulation

ODN 2006 Control (ODN 2137) can be used as a control ODN to study the stimulatory effect of ODN 2006 (ODN 7909) on TLR9 in HEK-Blue™ TLR9 cells. These cells stably overexpress the TLR9 gene and an NF- κ B-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 2006 Control (ODN 2137) at the same concentration as the CpG-containing ODN 2006.

1. Dispense 20 μ l 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 $\times 10^4$) to each ODN-containing well.
4. Incubate for 6-24 h at 37°C, 5% CO₂.
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™ hTLR9 cells	Human TLR9 reporter cells	hkb-hTLR9
ODN 2006 (ODN 7909)	Stimulatory ODN	tlr1-2006
QUANTI-Blue™ Solution	SEAP detection medium	rep-qbs