

ODN 2243 (ODN 2216 Control)

Negative control oligonucleotide for human TLR9 agonist ODN 2216

Catalog code: tlr1-2243-1

<https://www.invivogen.com/odn2216-control>

For research use only

Version 21L21-MM

PRODUCT INFORMATION

Contents

- 1 mg (154.5 nmol) of ODN 2243 (ODN 2216 Control) provided lyophilized
- Note: ODN 2243 (ODN 2216 Control) is sterile filtered prior to lyophilization.
- 1.5 ml endotoxin-free water

ODN 2243 (ODN 2216 Control) sequence

5'- ggG GGA GCA TGC TGg ggg gg -3' (20 mer)

Note: Bases shown in capital letters are phosphodiester, and those in lower case are phosphorothioate (nuclease resistant).

Molecular weight: 6472 g/mol

Storage and stability

- ODN 2243 (ODN 2216 Control) is shipped at room temperature. Upon receipt, store at -20°C.
- Upon resuspension, prepare aliquots of ODN 2243 (ODN 2216 Control) 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

ODN 2243 (also known as ODN 2216 Control) is designed as a negative control for the TLR9 agonist ODN 2216. ODN 2216 is an A-class CpG ODN with a preference for human TLR9. A-class CpG ODNs are characterized by a phosphodiester central CpG-containing palindromic motif and a phosphorothioate 3' poly-G string. They induce high IFN- α production from plasmacytoid dendritic cells (pDC) but are weak stimulators of TLR9-dependent NF- κ B signaling¹⁻³. ODN 2243 (ODN 2216 Control) contains GpC dinucleotides instead of CpGs and does not induce TLR9 activity³⁻⁵.

1. Krug A. *et al.*, 2001. Identification of CpG oligonucleotide sequences with high induction of IFN- α /beta in plasmacytoid dendritic cells. *Eur. J. Immunol.* 31:2154-63. 2. Ballas Z. *et al.*, 1996. Induction of NK activity in murine and human cells by CpG motifs in oligodeoxynucleotides and bacterial DNA. *J. Immunol.* 157:1840-5. 3. Vollmer J. *et al.*, 2004. Characterization of three CpG oligodeoxynucleotide classes with distinct immunostimulatory activities. *Eur. J. Immunol.* 34: 251-62. 4. Rothenfusser S. *et al.*, 2004. CpG-A and CpG-B oligonucleotides differentially enhance human peptide-specific primary and memory CD8+ T cell responses in vitro. *Blood* 103:2162-9. 5. Ayash-Rashkovsky M. *et al.*, 2005. Enhanced HIV-1 specific immune response by CpG ODN and HIV-1 immunogen-pulsed dendritic cells confers protection in the TrimerA murine model of HIV-1 infection. *FASEB J.* 19:1152-4.

TECHNICAL SUPPORT

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METHODS

Preparation of ODN solution (500 μ M)

- Add 310 μ l of endotoxin-free water (provided) to 1 mg vial of ODN 2243 (ODN 2216 Control).
- Vortex until completely dissolved. Prepare aliquots and store at -20°C.

TLR9 stimulation

ODN 2243 (ODN 2216 Control) can be used as a control ODN to study the stimulatory effect of ODN 2216 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 2243 (ODN 2216 Control) at the same concentration as the CpG-containing ODN 2216.

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 2216	Stimulatory ODN	tlr1-2216
QUANTI-Blue™ Solution	SEAP detection medium	rep-qbs