Zymosan

Cell wall from Saccharomyces cerevisiae - TLR2 & Dectin-1 ligand
Catalog # tlrl-zyn

For research use only
Version # 14L17-MM

PRODUCT INFORMATION

Content:

- 100 mg Zymosan
- 10 ml endotoxin-free water

Storage:

- Zymosan is shipped at room temperature and should be stored at 4 $^{\circ}\text{C}.$
- Resuspended zymosan can be stored at 4 $^{\circ}$ C for 6 months or at -20 $^{\circ}$ C for 12 months.

DESCRIPTION

Zymosan, an insoluble preparation of yeast cell, activates macrophages via TLR2. TLR2 cooperates with TLR6 and CD14 in response to zymosan¹. Zymosan is also recognized by Dectin-1, a phagocytic receptor expressed on macrophages and dendritic cells, which collaborates with TLR2 and TLR6 enhancing the immune responses triggered by the recognition of zymosan by each receptor².

1. Ozinsky A. *et al.*, 2000. The repertoire for pattern recognition of pathogens by the innate immune system is defined by cooperation between toll-like receptors. PNAS. 97(25):13766-71. 2. Gantner BN. *et al.*, 2003. Collaborative induction of inflammatory responses by dectin-1 and Toll-like receptor 2. J Exp Med. 197(9):1107-17. 3. Schindler U. & Baichwal VR., 1994. Three NF-κB binding sites in the human E-selectin gene required for maximal tumor necrosis factor alpha-induced expression. Mol Cell Biol, 14(9):5820-5831.

METHODS

Preparation of Zymosan suspension (10 mg/ml)

- \bullet Stimulation of TLR2 can be achieved with 10 $\mu g/ml$ zymosan.
- \bullet Stimulation of Dectin-1 can be achieved with 1 10 $\mu g/ml$ zymosan.
- To rehydrate the product, add 10 ml endotoxin-free water to 100 mg Zymosan.
- Vortex to homogenize.

Note: The solution remains hazy.

Zymosan stimulation of TLR2 or Dectin-1

- Transfect your cell line with an NF- κ B reporter plasmid, i.e. a plasmid carrying a reporter gene such as GFP, SEAP or luciferase, under the control of the NF- κ B-inducible ELAM-1(E-selectin) promoter³.

<u>Note</u>: InvivoGen provides pNiFty, a family of NF-κB-inducible reporter plasmids that can be transfected transiently (pNiFty) or stably (pNiFty2). pNiFty plasmids are available either with the SEAP or luciferase reporter genes (see Related Products).

Zymosan can be used to activate TLR2 or Dectin-1. If your cell line does not naturally express TLR2, cotransfect with a TLR2 expressing plasmid, such as pUNO-TLR2. If your cell line does not naturally express Dectin-1, cotransfect with a Dectin-1 expressing plasmid, such as pUNO-hdectin1b or pUNO-mdectin1. Note: Alternatively, evaluate TLR2 activation using reporter cells, such as InvivoGen's HEK-Blue™ hTLR2 cells which express the human TLR2 and SEAP reporter genes. Dectin-1 activation can be assessed using RAW-Blue™ cells that express the SEAP reporter gene and high levels of endogenous Dectin-1. NF-κB production in these cells can be easily quantified using a SEAP detection medium, such as QUANTI-Blue™ or HEK-Blue™ Detection.

- Twenty-four to forty-eight hours after transfection, stimulate cells with 1 -10 $\mu g/ml$ Zymosan for 6 to 24 hours.
- Determine Zymosan stimulation on TLR2 or Dectin-1 by assessing reporter gene expression using the appropriate detection system.

RELATED PRODUCTS

Product	Catalog Code
HEK-Blue™ hTLR2 Cells pNiFty-Luc (Amp ^R) pNiFty-SEAP (Amp ^R) pNiFty2-Luc (Zeo ^R) pNiFty2-SEAP (Zeo ^R) pUNO1-hTLR2 (human gene) pUNO-mTLR2 (mouse gene) pUNO-hDECTIN1a pUNO-hDECTIN1b pUNO-mDECTIN1 RAW-Blue™ Cells	hkb-htlr2 pnifty-luc pnifty-seap pnifty2-luc pnifty2-seap puno1-htlr2 puno-mtlr2 puno-hdectin1a puno-hdectin1b puno-mdectin1



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