# **Zymosan Depleted**

# Hot alkali treated Zymosan; Dectin-1 ligand

Catalog # tlrl-zyd

http://www.invivogen.com/zymosan-depleted

# For research use only

Version # 16L05-MM

# PRODUCT INFORMATION

#### **Content:**

- 10 mg Zymosan Depleted
- 10 ml sterile endotoxin-free water

#### **Storage:**

- Zymosan Depleted is shipped at room temperature and should be stored at -20  $^{\circ}\text{C}.$
- Resuspended Zymosan Depleted may be stored at -20  $^{\circ}\mathrm{C}$  for 1 month.

#### **Quality control:**

- The Dectin-1 activity has been tested using HEK-Blue<sup>™</sup> hDectin-1b cells.
- The absence of bacterial contamination (e.g. lipoproteins & endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

### DESCRIPTION

Zymosan Depleted was obtained by treating zymosan (an insoluble preparation of *Saccharomyces cerevisiae* cell wall) with hot alkali to remove its Toll-like receptor (TLR)-stimulating properties. Hence, Zymosan Depleted activates the C-type lectin receptor Dectin-1 but not TLR2. The use of hot alkali or organic solvents to abrogate the TLR2-dependent response of zymosan whilst preserving the Dectin-1 activity has been described previously<sup>1, 2</sup>.

Dectin-1, a phagocytic receptor expressed on macrophages and dendritic cells, is a specific receptor for  $\beta$ -glucans<sup>3</sup>, the glucose polymers found in the cell walls of fungi such as *C. albicans* and *S. cerevisiae*. More precisely, this receptor binds and internalizes the  $\beta$ -glucans leading to the production of reactive oxygen species, the activation of NF- $\kappa$ B and the subsequent secretion of proinflammatory cytokines.

1. 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. 2. Ikeda Y. et al., 2008. Dissociation of Toll-like receptor 2-mediated innate immune response to Zymosan by organic solvent-treatment without loss of Dectin-1 reactivity. Biol Pharm Bull.31(1):13-8. 3. Brown GD. et al., 2003. Dectin-1 mediates the biological effects of beta-glucans. J Exp Med. 197(9):1119-24.

# **METHODS**

# Preparation of sterile solution (5 mg/ml)

Stimulation of Dectin-1 can be achieved with 10-100  $\mu g/ml$  of Zymosan Depleted.

- Add 2 ml of sterile endotoxin-free water to 10 mg of Zymosan Depleted.
- Vortex to homogenize.

Note: The solution remains hazy.

#### **Dectin-1 activation using Zymosan Depleted**

Zymosan Depleted can be used to activate Dectin-1 in cells expressing this receptor, such as HEK-Blue™ hDectin-1 cells. These cells stably express the human Dectin-1b gene and genes of the Dectin-1 signaling pathway leading to NF-κB activation. In addition, they express an NF-κB-inducible secreted embryonic alkaline phosphatase (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 it is secreted by the cells.

For more information visit: www.invivogen.com/hek-blue-hdectin1b

- Add 20  $\mu l$  of Zymosan Depleted at 10-100  $\mu g/ml$  per well of a 96-well plate.
- Prepare a cell suspension (~280,000 cells per ml) in HEK-Blue $^{\!\scriptscriptstyle \rm M}$  Detection medium and add 180  $\mu l$  of the cell suspension (~50,000 cells) to each Zymosan Depleted-containing well.
- Incubate the plate for 6-24 h at 37 °C, 5% CO<sub>2</sub>.
- Determine SEAP levels using a spectrophotometer at 620-655 nm.

# RELATED PRODUCTS

Product	Catalog Code
Beta-glucan peptide Curdlan AL HEK-Blue™ hDectin-1b HKCA (Heat-killed <i>C. albicans</i> ) HKSC (Heat-killed <i>S. cerevisiae</i> ) Schizophyllan (β-glucan from <i>S. commune</i> )	tlrl-bgp tlrl-cura hkb-hdect1b tlrl-hkca tlrl-hksc tlrl-spg
HKSC (Heat-killed S. cerevisiae)	tlrl-hksc

