HKCA

Heat-killed preparation of *Candida albicans*; Dectin-1 ligand

Catalog code: tlrl-hkca

https://www.invivogen.com/hkca

For research use only

Version 20J02-MM

PRODUCT INFORMATION

Contents

- 10^9 freeze-dried cells of heat-killed preparation of *Candida albicans* (HKCA).
- 1.5 ml of sterile endotoxin-free water

Storage and stability

- HKCA is shipped at room temperature and should be stored at 4°C. Freeze-dried HKCA can be stored at 4°C for at least 1 year.
- Resuspended HKCA can be stored at 4°C for 6 months.

Quality control:

- The biological activity has been validated using HEK-Blue™ hDectin-1b cells.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

DESCRIPTION

HKCA is a heat-killed preparation of *C. albicans*. HKCA derives from the strain ATCC 10231. HKCA activates the β-glucan specific dectin-1 receptor, which is expressed on phagocytes[1]. β-Glucans are glucose polymers found in the cell walls of fungi, such as zymosan (a cell wall preparation of *Saccharomyces cerevisiae*) and *Candida albicans*. Dectin-1 binds and internalizes β-glucans and mediates the production of reactive oxygen species (ROS), activation of NF-κB and subsequent secretion of proinflammatory cytokines. However, it is now clear that its β-glucan moiety triggers NF-κB activation only through Dectin-1 as treatment with hot alkali or organic solvents abrogates the TLR2-dependent response[2,3]. RAW-Blue™ cells express high levels of endogenous Dectin-1 and therefore can be used as a Dectin-1 reporter cell line. Stimulation of RAW-Blue™ cells with depleted zymosan or heat-killed preparations of yeast induces the activation of NF-κB in a Dectin-dependent manner. NF-κB activation can be readily monitored as RAW-Blue™ cells stably express an NF-κB-inducible SEAP reporter gene.

1. Brown GD. et al., 2003. Dectin-1 mediates the biological effects of beta-glucans. J Exp Med. 197: 1119- 24. **2. Gantner BN. et al., 2003.** Collaborative induction of inflammatory responses by dectin-1 and Toll- likereceptor 2. J Exp Med. 197: 1107- 17. **3. 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: 13-8. **4. 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, 14:5820-5831.

METHODS

Preparation of sterile suspension (10^9 cells/ml)

Stimulation of Dectin-1 can be achieved with HKCA 10^8 cells/ml.

1. Add 1 ml of sterile endotoxin-free water (provided) to rehydrate the pellet.
2. Vortex for 10 seconds to homogenize.

Note: Resuspended HKCA results in a milky solution.

Detection of HKCA-induced Dectin-1 activation

HKCA can be used to activate Dectin-1 in cells expressing this receptor, such as the HEK-Blue™ hDectin-1b cells. These HEK293 cells were transfected with the human Dectin-1b gene and other genes from the Dectin-1 signaling pathway. These cells also stably express a secreted embryonic alkaline phosphatase (SEAP) reporter gene.

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

1. Add 20 µl of HKCA (10^8 cells/ml final concentration) in a well of a 96-well plate.
2. Add 180 µl of HEK-Blue™ hDectin-1b cells (5 x 10^5 cells) per well.
3. Incubate cells for 16-24 h at 37 °C, 5% CO_2.
4. Determine of Dectin-1a activation by assessing SEAP expression using a SEAP detection medium, such as QUANTI-Blue™.

RELATED PRODUCTS

Product	Description	Cat. Code
Beta-glucan peptide	Dectin-1 agonist	tlrl-bgp
Curdlan	Dectin-1 agonist	tlrl-curd
HEK-Blue™ hDectin-1b cells	Reporter cells	hkb-hdect1b
RAW-Blue™ cells	Reporter cells	raw-sp
Zymosan	TLR2 & Dectin-1 agonist	tlrl-zyn
Zymosan depleted	Dectin-1 agonist	tlrl-dzn

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