

# Validation data for Nigericin

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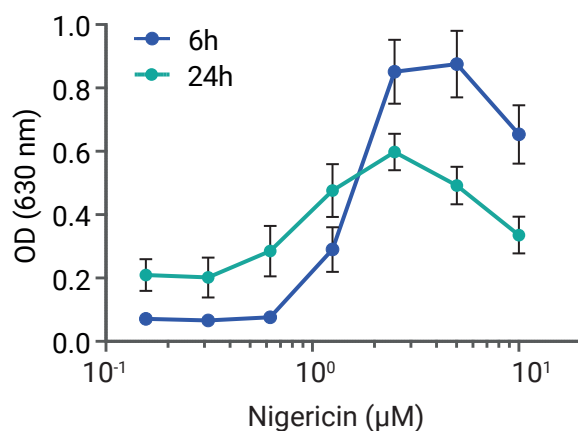
Version 21K03-MM

Nigericin, a bacterial toxin, is an inducer of the NLRP3 inflammasome, a large intracellular multiprotein complex that plays a central role in innate immunity<sup>1,2</sup>. NLRP3 inflammasome activation requires an initial signal ('priming'), provided by microbial molecules, such as lipopolysaccharide (LPS), and a secondary signal, provided by a wide array of stimuli including endogenous molecules, crystals or bacterial toxins, such as Nigericin. This triggers the multimerization of the NLRP3 inflammasome and caspase-1 activation with the subsequent maturation and secretion of IL-1 $\beta$  and IL-18.

The ability of Nigericin to induce the NLRP3 inflammasome has been validated using THP1-Null cells. The production of IL-1 $\beta$  by THP1-Null cells was measured using HEK-Blue™ IL-1 $\beta$  cells. Treatment with Nigericin induced IL-1 $\beta$  secretion, an indicator of NLRP3 inflammasome activation, in a dose-dependent manner.

1. Schroder K. & Tschopp J., 2010. The inflammasomes. Cell 140(6):821-32. 2. Franchi L. *et al.*, 2012. Sensing and reacting to microbes through the inflammasomes. Nat Immunol 13(4):325-32.

## Evaluation of NLRP3 inflammasome activation



**Figure 1. IL-1 $\beta$  production in THP1-Null cells.** Human THP-1 monocytes were primed with LPS-EK (1  $\mu$ g/ml) prior to incubation with increasing concentrations of Nigericin. After 6h and 24h activation, IL-1 $\beta$  secretion was assessed in the culture supernatant using HEK-Blue™ IL-1 $\beta$  sensor cells and the SEAP detection reagent QUANTI-Blue™ Solution. The optical density (OD) was read at 630 nm.

### TECHNICAL SUPPORT

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