

# Validation data for Nano-SiO<sub>2</sub>

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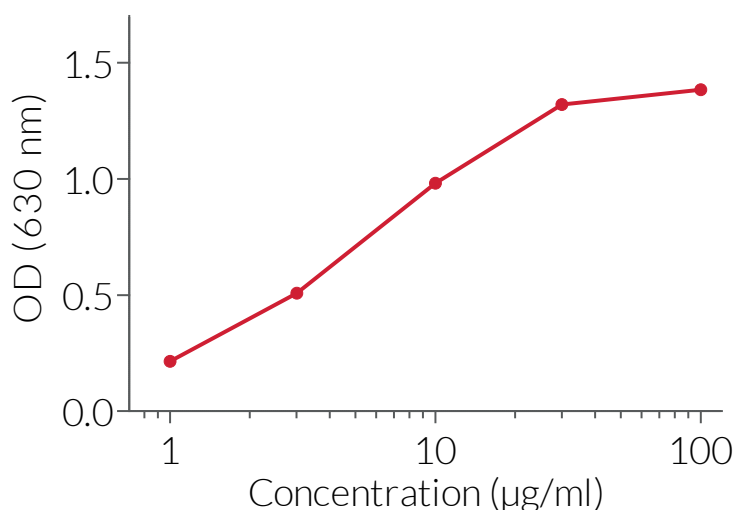
Version 22H22-AK

Silica dioxide nanoparticles (Nano-SiO<sub>2</sub>) are inducers 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 bacterial toxins, endogenous molecules, crystals or nanoparticles such as Nano-SiO<sub>2</sub>. 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 Nano-SiO<sub>2</sub> 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 Nano-SiO<sub>2</sub> 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



**IL-1 $\beta$  production in THP1-Null cells.** THP1-Null cells, primed with LPS (1 µg/ml for 3h), were stimulated with increasing concentrations of Nano-SiO<sub>2</sub>. After overnight incubation, IL-1 $\beta$  secretion was analyzed by adding 50 µl of supernatant from treated THP1-Null cells to HEK-Blue™ IL-1 $\beta$  cells. IL-1 $\beta$ -induced activation of NF- $\kappa$ B was assessed by measuring the levels of SEAP in the supernatant of HEK-Blue™ IL-1 $\beta$  cells using QUANTI-Blue™ Solution, a SEAP detection reagent, and by reading the optical density (OD) at 630 nm.

### TECHNICAL SUPPORT

InvivoGen USA (Toll-Free): 888-457-5873

InvivoGen USA (International): +1 (858) 457-5873

InvivoGen Europe: +33 (0) 5-62-71-69-39

InvivoGen Hong Kong: +852 3622-3480

E-mail: [info@invivogen.com](mailto:info@invivogen.com)