MCC950 (also known as CP-456773) is a potent and specific NLRP3 inhibitor that prevents NLRP3 (NOD-like receptor (NLR) pyrin domain-containing protein 3) inflammasome assembly in a reversible manner. The NLRP3 inflammasome is an innate immune sensor that is activated by a two-step process; a first signal ('priming') is provided by microbial molecules such as lipopolysaccharide (LPS), while the second signal is provided by a wide array of stimuli including endogenous molecules or crystalline substances such as monosodium urate (MSU) crystals. The ability of MCC950 to inhibit the NLRP3 inflammasome was validated using InvivoGen’s THP-1/HEK-Blue™ IL-1β assay. This assay uses the secretion of IL-1β by THP1-Null2 cells as an indicator of NLRP3 inflammasome induction. The IL-1β production by these cells is measured using HEK-Blue™ IL-1β cells. Treatment with MCC950 inhibited IL-1β secretion in a dose-dependent manner (Figure 1).

![Dose-dependent inhibition of NLRP3 activity](image)

**Figure 1:** MCC950 inhibits the NLRP3 inflammasome response in a dose-dependent manner.

THP1-Null2 cells, primed with LPS-EK (1 µg/ml for 3 h), were stimulated with MSU (150 µg/ml) and increasing concentrations of MCC950. After overnight incubation, IL-1β secretion was analyzed by adding 50 µl of supernatant from treated THP1-Null2 cells to HEK-Blue™ IL-1β cells. IL-1β-induced activation of NF-κB was assessed by measuring the levels of SEAP in the supernatant of HEK-Blue™ IL-1β cells using QUANTI-Blue™ Solution, a SEAP detection reagent, and by reading the optical density (OD) at 655 nm. Data are shown as a percentage (%) inhibition of the maximal response for the ligand with no inhibitor.