Validation data for CL075

https://www.invivogen.com/cl075

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Version 23L18-AK

CL075 is a thiazoloquinolone derivative and a potent agonist of the Toll-like receptors 7 and 8 (TLR7 and TLR8). The ability of CL075 to activate TLR7 and TLR8 signaling was validated using a panel of InvivoGen's reporter cell lines. CL075 efficiently activates human (h)TLR7, murine (m)TLR7 and hTLR8, but not mTLR8, as assessed by the expression of an NF- κ B-inducible secreted embryonic alkaline phosphatase (SEAP) reporter in HEK-BlueTM -derived cell lines (Figure 1). The induction of the NF- κ B and IRF pathways by CL075 has been tested using InvivoGen's HEK-DualTM cells featuring two reporter genes, the NF- κ B-inducible SEAP and IRF-inducible Lucia luciferase, as well as the overexpression of TLR7 or TLR8 (Figure 2).

Dose-dependent NF-κB response in HEK-Blue™-derived cells

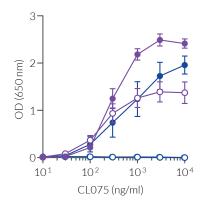




Figure 1. NF-κB response of HEK-Blue[™]-derived cells to CL075. HEK-Blue[™] cells expressing hTLR7, mTLR7, hTLR8, or mTLR8 were cultured in HEK-Blue[™] Detection reagent and stimulated with increasing concentrations of CL075. After 24h incubation, the NF-κB-induced SEAP activity was assessed by measuring the SEAP level in the supernatant. Data are shown as optical density (OD) at 650 nm (mean ± SEM). Of note, HEK-Blue[™] Null* comprises data from the parental cell lines HEK-Blue Null1, HEK-Blue Null1-v, and HEK-Blue Null2-k.

Dose-dependent NF-κB and IRF responses in HEK-Dual[™]-derived cells

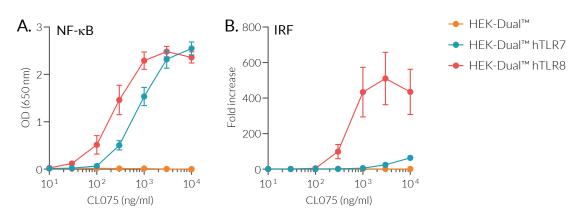


Figure 2. NF-κB and IRF responses of THP1-DualTM-derived cells to CL075. HEK-DualTM, HEK-DualTM hTLR7, and HEK-DualTM hTLR8 cells were incubated for 24 hours with increasing concentrations of CL075. After 24h incubation, the (A) NF-κB-induced SEAP activity was assessed using QUANTI-BlueTM. Data are shown as optical density (OD) at 650 nm (mean \pm SEM). (B) The IRF response was assessed by measuring the activity of Lucia luciferase in the supernatant using QUANTI-LucTM. Data are shown in fold response over non-induced cells (mean \pm SEM).



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