

# Validation data for THP1-Dual™ hTLR7 cells

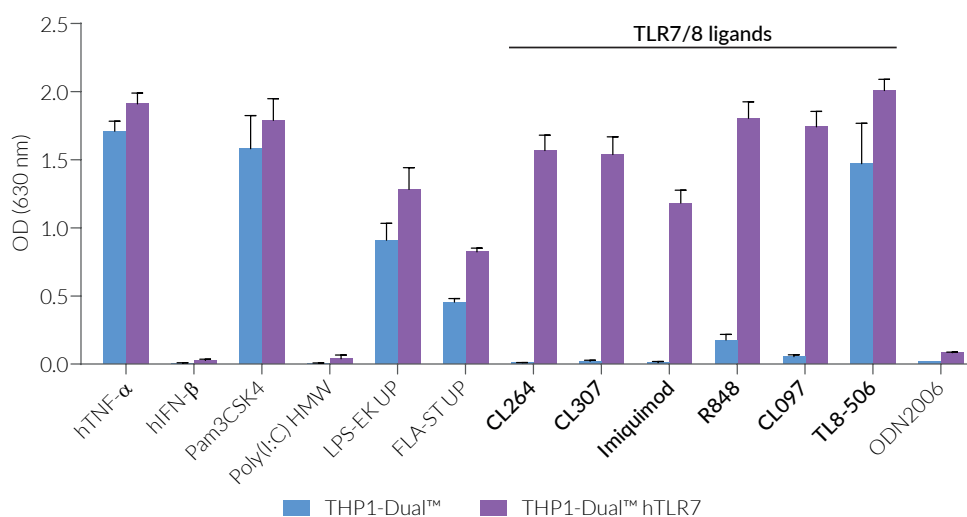
<https://www.invivogen.com/thp1-dual-htlr7-htlr8>

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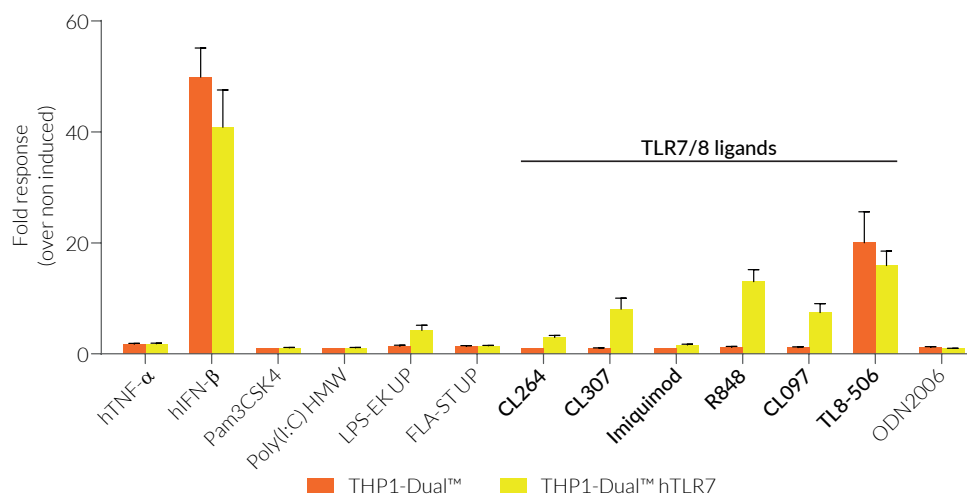
THP1-Dual™ hTLR7 cells were generated from the THP1-Dual™ cell line through the stable expression of the human Toll-like receptor 7 (hTLR7) and UNC93B1<sup>mut</sup>. These cells also feature two reporter genes allowing the simultaneous study of the NF-κB pathway by monitoring the activity of an inducible SEAP (secreted embryonic alkaline phosphatase), and the IRF pathway by monitoring the activity of an inducible secreted Lucia luciferase. Human TLR7 overexpression together with UNC93B1<sup>mut</sup> in THP1-Dual™ cells allows potent NF-κB and IRF responses upon incubation with TLR7-specific ligands, when compared to their parental cells (Figures 1 & 2). Of note, as THP-1 cells express endogenous levels of various TLRs, THP1-Dual™-derived cells respond to the cognate ligands including Pam3CSK4, LPS, or flagellin.

## Functional validation of THP1-Dual™-derived cells (NF-κB responses)



**Figure 1. NF-κB responses in THP1-Dual™-derived cells.** THP1-Dual™ and THP1-Dual™ hTLR7 cells were incubated for 24 hours with various TLR agonists: Pam3CSK4 (TLR2 ligand, 10 ng/ml), Poly(I:C) HMW (TLR3 ligand, 10 µg/ml), LPS-EK Ultrapure (UP) (TLR4 ligand, 1 ng/ml), FLA-ST UP (TLR5 ligand, 1 µg/ml), **CL264** (TLR7 ligand, 1 µg/ml), **CL307** (TLR7 ligand, 1 µg/ml), **Imiquimod** (TLR7 ligand, 10 µg/ml), **R848** (TLR7/8 ligand, 1 µg/ml), **CL097** (TLR7/8 ligand, 1 µg/ml), **TL8-506** (TLR8 ligand, 1 µg/ml), and ODN 2006 (TLR9 ligand, 10 µg/ml). Human TNF-α (1 ng/ml) was used as an NF-κB-positive control. After 24h incubation, the NF-κB-induced SEAP activity was assessed using QUANTI-Blue™. Data are shown as optical density (OD) at 630 nm (mean ± SEM).

## Functional validation of THP1-Dual™-derived cells (IRF response)



**Figure 2. IRF responses in THP1-Dual™-derived cells.** THP1-Dual™ and THP1-Dual™ hTLR7 cells were treated as described above. Human IFN-β (1000 U/ml) was used as an IRF-positive control. After 24h incubation, the IRF response was assessed by measuring the activity of Lucia luciferase in the supernatant using QUANTI-Luc™. Data are shown in fold response over non-induced cells (mean ± SEM).

### TECHNICAL SUPPORT

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