

Validation data for HEK-Blue™ hTLR2 cell collection

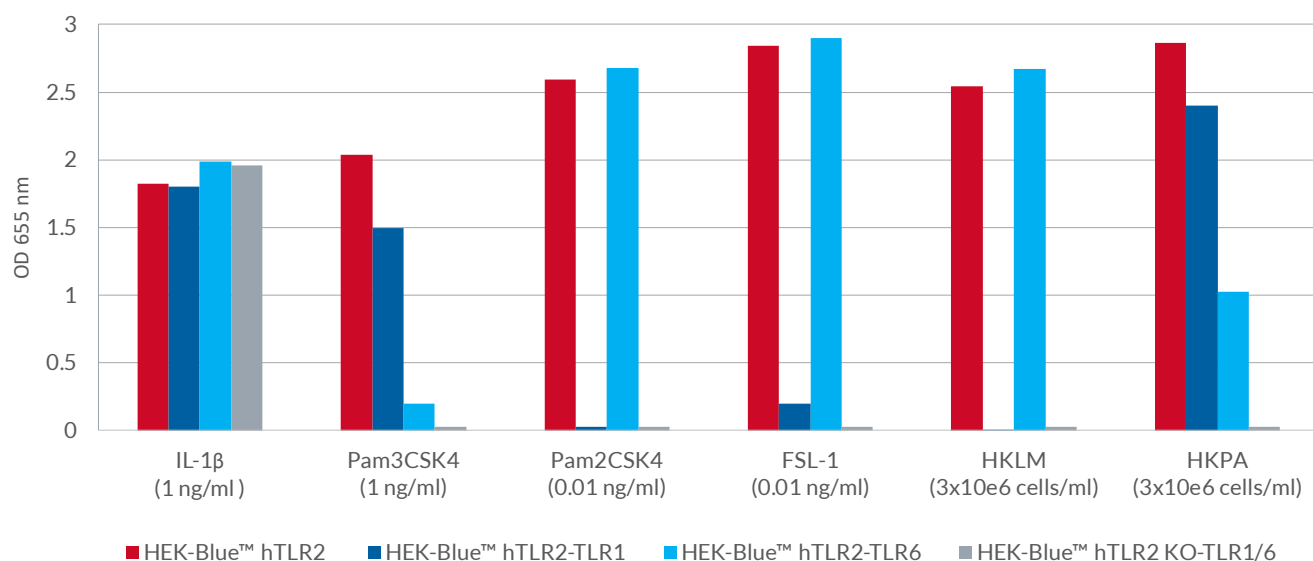
<https://www.invivogen.com/hek-blue-htlr2>

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InvivoGen provides a collection of engineered HEK293-derived cell lines designed for investigating the TLR1-TLR2 and/or TLR2-TLR6 signaling pathways. This collection was generated using HEK-Blue™ hTLR2 cells, which endogenously express TLR1 and TLR6 and have been stably transfected with human TLR2 and CD14 genes, together with an NF-κB/AP-1-inducible secreted embryonic alkaline phosphatase (SEAP) reporter gene. Cells in the collection include HEK-Blue™ hTLR2 KO-TLR1/6 which harbors a double knockout of TLR1 and TLR6, as well as HEK-Blue™ hTLR2-TLR1 and HEK-Blue™ hTLR2-TLR6 derived from the double knockout which stably express exogenous human TLR1 and TLR6, respectively. Biological activity has been assessed by measuring the levels of NF-κB-induced SEAP reporter activity. HEK-Blue™ hTLR2-TLR1 cells respond to triacylated lipoproteins (Pam₃CSK₄) whereas HEK-Blue™ hTLR2-TLR6 respond to diacylated lipoproteins (Pam₂CSK₄ and FSL-1; see figure below). Gram⁻ bacteria such as *Pseudomonas aeruginosa* are predominantly recognized by HEK-Blue™ hTLR2-TLR1 cells whereas Gram⁺ bacteria such as *Listeria monocytogenes* are predominantly recognized by HEK-Blue™ hTLR2-TLR6 cells.

Discrimination of TLR1-TLR2 and TLR2-TLR6 activation with synthetic ligands and heat-killed bacteria



HEK-Blue™ hTLR2, HEK-Blue™ hTLR2-TLR1, HEK-Blue™ hTLR2-TLR6 and HEK-Blue™ hTLR2 KO-TLR1/6 cells were incubated in HEK-Blue™ Detection medium and stimulated with 10 ng/ml Pam₃CSK₄, 0.01 ng/ml Pam₂CSK₄ and FSL-1, 3 x 10⁶ cells/ml HKLM (heat killed *L. monocytogenes*; Gram⁺ bacteria) and HKPA (heat killed *P. aeruginosa*; Gram⁻ bacteria), and 1 ng/ml of recombinant human IL-1β. After 24h the level of NF-κB-induced SEAP was determined by reading the optical density (OD) at 655 nm.

TECHNICAL SUPPORT

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