

Validation data for HEK-Blue-Lucia™ hTLR9 (NF/IL8) cells (NF-κB-SEAP/KI-[IL-8]Lucia)

<https://www.invivogen.com/hek-blue-lucia-htlr9>

For research use only

Version 24B05-AK

HEK-Blue-Lucia™ hTLR9 (NF/IL8) cells are designed for studying the stimulation of human TLR9 (hTLR9) by monitoring the activation of the NF-κB/AP-1 and the IL-8 pathways. They stably express hTLR9 and two different reporter proteins: secreted embryonic alkaline phosphatase (SEAP) and secreted Lucia luciferase. In addition, these cells are triple knockout for TLR3, TLR5, and the TNF receptor, thus enabling the study of hTLR9 signaling without interference from other TLRs. They respond to very low concentrations of TLR9 agonists, such as oligonucleotides containing CpG motifs (CpG ODNs). They do not respond to other TLR agonists or to the cytokine TNF-α. The NF-κB response of these cells has been compared to that of HEK-Blue™ hTLR9 cells (SEAP reporter cells) using QUANTI-Blue™ Solution, a SEAP detection medium (Figure 1). Furthermore, HEK-Blue-Lucia™ hTLR9 (NF/IL8) cells also enable monitoring activation of the IL-8 pathway using QUANTI-Luc™ 4 Lucia/Gaussia, a Lucia and Gaussia luciferase detection reagent (Figure 2).

NF-κB (SEAP) response

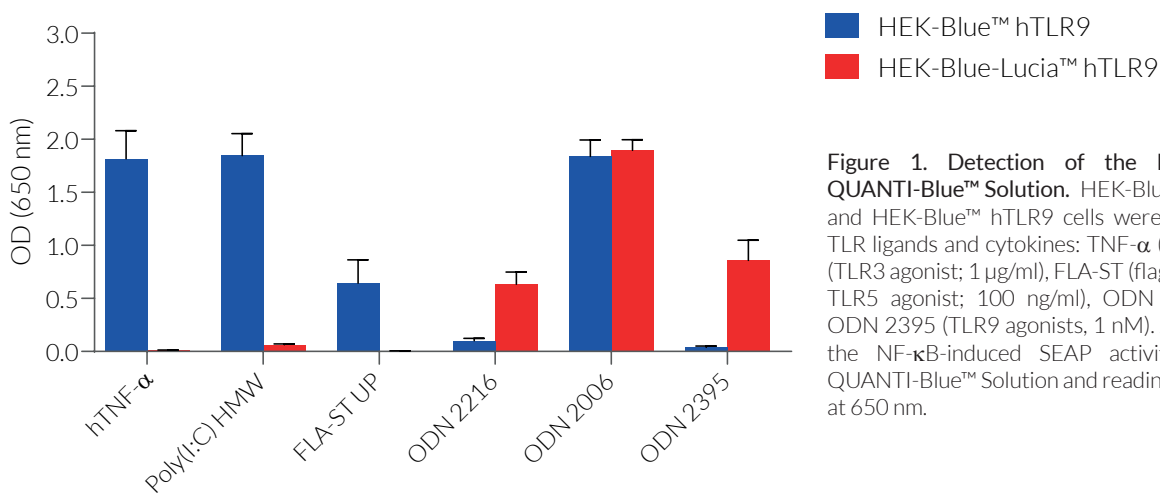


Figure 1. Detection of the NF-κB response using QUANTI-Blue™ Solution. HEK-Blue-Lucia™ hTLR9 (NF/IL8) and HEK-Blue™ hTLR9 cells were stimulated with various TLR ligands and cytokines: TNF-α (1 ng/ml), Poly(I:C) HMW (TLR3 agonist; 1 μg/ml), FLA-ST (flagellin from *S. typhimurium*, TLR5 agonist; 100 ng/ml), ODN 2216, ODN 2006, and ODN 2395 (TLR9 agonists, 1 nM). After 24 hour incubation, the NF-κB-induced SEAP activity was assessed using QUANTI-Blue™ Solution and reading the optical density (OD) at 650 nm.

KI-IL-8 (Lucia) response

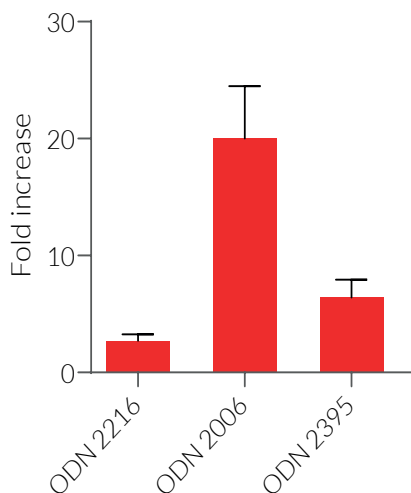


Figure 2. Detection of the IL-8 response using QUANTI-Luc™ 4 Lucia/Gaussia. HEK-Blue-Lucia™ hTLR9 (NF/IL8) cells were stimulated with 1 nM of the following TLR9 agonists: ODN 2216, ODN 2006, and ODN 2395. After 24 hour incubation, activation of the IL-8 promoter was determined by measuring the relative light units (RLUs) in a luminometer using QUANTI-Luc™ 4 Lucia/Gaussia, a Lucia and Gaussia luciferase detection reagent. Data are shown in fold response over non-induced cells (mean ± SEM).

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 Asia: +852 3-622-34-80
E-mail: info@invivogen.com