

# Validation data for HEK-Blue™ IFN-γ cells

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

For research use only

Version 23113-AK

HEK-Blue™ IFN-γ cells were generated by stable transfection of HEK293 cells with the human *STAT1* gene to obtain a fully active *STAT1* pathway. These cells were further transfected with a SEAP (secreted embryonic alkaline phosphatase) reporter gene under the control of an IGS54 promoter fused to four interferon-gamma-activated sites (GAS). Stimulation of HEK-Blue™ IFN-γ cells with human IFN-γ triggers the activation of the GAS-inducible promoter and the production of SEAP. They do not respond to murine (m) IFN-γ (Figure 1). Of note, HEK-Blue™ IFN-γ cells do not respond to either type I IFNs (IFN-α/β) or type III IFN (IFN-λ) (Figure 2). These cells can also be used to screen for molecules that inhibit IFN-γ signaling, such as antibodies targeting IFN-γ (Figure 3).

Dose-response to human and murine IFN-γ

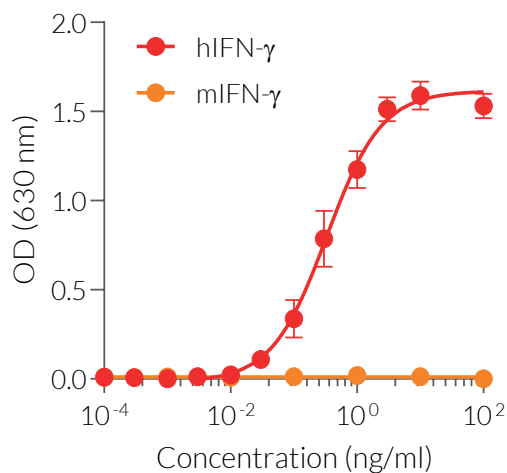


Figure 1. Dose-response of HEK-Blue™ IFN-γ cells to human and murine IFN-γ. Cells were stimulated with increasing concentrations of recombinant human (h) and murine (m)IFN-γ. After overnight incubation, the ISGF3 response was determined using QUANTI-Blue™ Solution, a SEAP detection reagent. The optical density (OD) at 630 nm is shown as mean ± SEM.

Response to human and murine IFN-γ

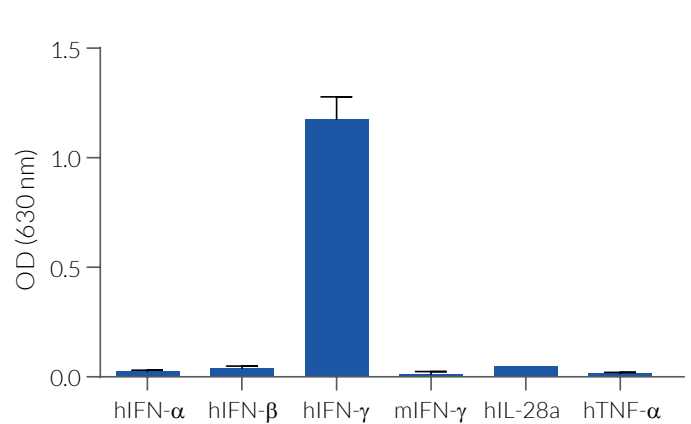


Figure 2. Response of HEK-Blue™ IFN-γ cells to a panel of cytokines. Cells were stimulated with various human recombinant cytokines: 100 U/ml hIFN-α2b or hIFN-β-1a, 1 ng/ml hIFN-γ, 10 ng/ml mIFN-γ, 100 ng/ml IL-28a, or 10 ng/ml hTNF-α. After overnight incubation, SEAP activity was assessed using QUANTI-Blue™ Solution. The OD at 630 nm is shown as mean ± SEM.

Neutralization of IFN-γ response using anti-hIFN-γ mAb

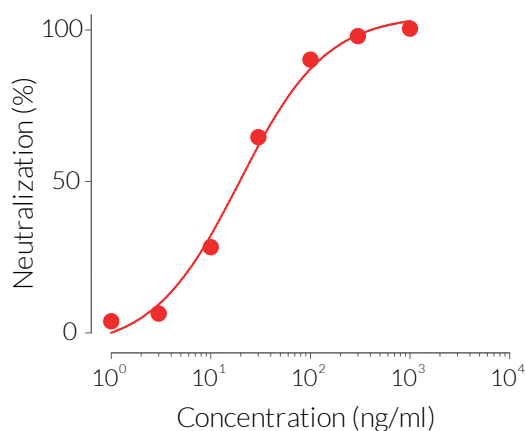


Figure 3. Dose-dependent inhibition of HEK-Blue™ IFN-γ cell response using Anti-IFN-γ-IgA. A serial dilution of Anti-IFN-γ-IgA monoclonal antibody (mAb) was incubated with 0.3 ng/ml of recombinant human IFN-γ for 30 minutes prior to the addition of the HEK-Blue™ IFN-γ cells. After overnight incubation, SEAP activity in the cell culture supernatant was determined using QUANTI-Blue™ Solution, a SEAP detection reagent. Data are presented as percentage of neutralization (mean).

## 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 3622-3480

E-mail: [info@invivogen.com](mailto:info@invivogen.com)