Validation data for HEK-Blue[™]IL-7 Cells

https://www.invivogen.com/hek-blue-il7

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Version 22D19-MM

HEK-Blue^M IL-7 cells allow the detection of bioactive interleukin-7 (IL-7) by monitoring STAT5 activation. These human embryonic kidney 293 (HEK293)-derived cells express the human IL-7 heterodimeric receptor (IL-7R α and IL-2R γ). They also express human JAK3, human STAT5b, and a STAT5-inducible secreted embryonic alkaline phosphatase (SEAP) reporter. Human IL-7R α expression by these cells has been verified by flow cytometry (**Figure 1**). HEK-Blue^M IL-7 cells respond to human and murine IL-7 (**Figure 2**). These cells can be used for screening of antibodies targeting the IL-7 pathway (**Figure 3**). Of note, HEK-Blue^M IL-7 cells are knock-out for human IFNAR2, and do not respond to human type I IFNs (**Figure 4**).

Validation of IL-7R α expression

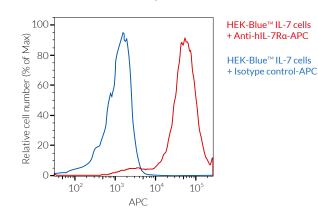


Figure 1. Validation of the expression of human IL-7R α by HEK-Blue[™] IL-7 cells. 5 x 10⁵ cells were incubated with either an APC-conjugated isotype control (blue) or an APC-conjugated Anti-IL-7R α mAb (red) for 30 minutes. The binding affinity was then measured using flow cytometry.

IL-7 signaling inhibition

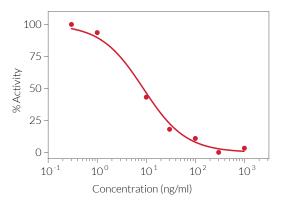


Figure 3. Dose-dependent inhibition of HEK-Blue[™] IL-7 cellular response using a neutralizing antibody against IL-2Rγ, a subunit of IL-7R. The antibody was incubated with HEK-Blue[™] IL-7 cells for 2 hours prior to the addition of hIL-7 (0.3 ng/ml). After overnight incubation, SEAP activity in the cell culture supernatant was assessed using QUANTI-Blue[™] Solution. Data are shown as a percentage (%) of activity.

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Cellular response to IL-7

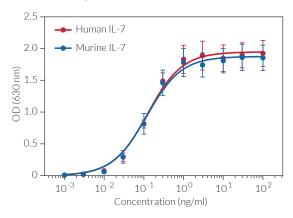


Figure 2. Dose-response of HEK-Blue[™] IL-7 cells to recombinant IL-7. Cells were stimulated with increasing concentrations of recombinant human IL-7 (hIL-7) and murine IL-7 (mIL-7). After overnight incubation, the STAT5 response was determined using QUANTI-Blue[™] Solution, a SEAP detection reagent. The optical density (OD) at 630 nm is shown as mean ± SEM.

Cell line specificity

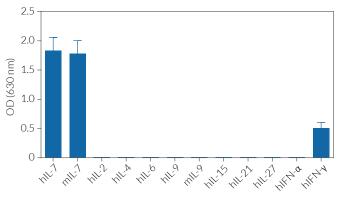


Figure 4. Response of HEK-Blue[™] IL-7 cells to a panel of cytokines. Cells were stimulated with various human and murine recombinant cytokines: 1 ng/ml of hIL-7 or mIL-7, and 100 ng/ml hIL-2, hIL-4, hIL-6, hIL-9, mIL-9, hIL-15, hIL-21, or hIL-27, and 1000 U/ml hIFN- α , and 10 ng/ml hIFN- γ . After overnight incubation, SEAP activity was assessed using QUANTI-Blue[™] Solution. The OD at 630 nm is shown as mean ± SEM.

