

Validation data for HEK-Blue™ GM-CSF cells

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

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Version 21J04-NJ

HEK-Blue™ GM-CSF cells allow the detection of bioactive granulocyte-macrophage colony-stimulating factor (GM-CSF) by monitoring STAT5 activation. These human embryonic kidney 293 (HEK293)-derived cells express the human GMR α (CD116), GMR β (CD131), STAT5 and a STAT5-inducible secreted embryonic alkaline phosphatase (SEAP) reporter. Human GMR α expression by these cells has been verified by flow cytometry (**Figure 1**). HEK-Blue™ GM-CSF cells respond to human but not murine GM-CSF (**Figure 2**). Of note, HEK-Blue™ GM-CSF are knock-out for human IFNAR2, and are not responding to human type I IFN (**Figure 3**).

Validation of GMR α expression

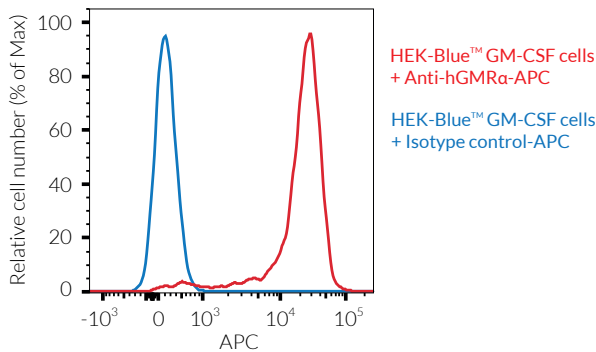


Figure 1. Validation of the expression of human GMR α by HEK-Blue™ GM-CSF cells. 5×10^5 cells were incubated with either a APC-conjugated isotype control (blue) or a APC-conjugated Anti-GMR α mAb (red) for 1 hour. The binding affinity was then measured using flow cytometry.

Cellular response to GM-CSF

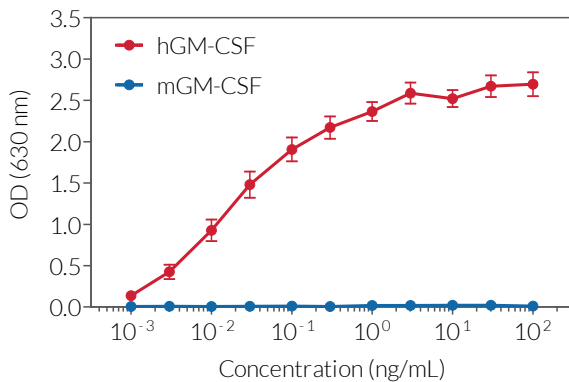


Figure 2. Dose-response of HEK-Blue™ GM-CSF cells to recombinant GM-CSF. Cells were stimulated with increasing concentrations of recombinant human GM-CSF (hGM-CSF) and murine GM-CSF (mGM-CSF). After overnight incubation, the STAT5 response was determined using QUANTI-Blue™ Solution, a SEAP detection reagent. The optical density (OD) at 630 nm is shown as mean \pm SEM.

Cell line specificity

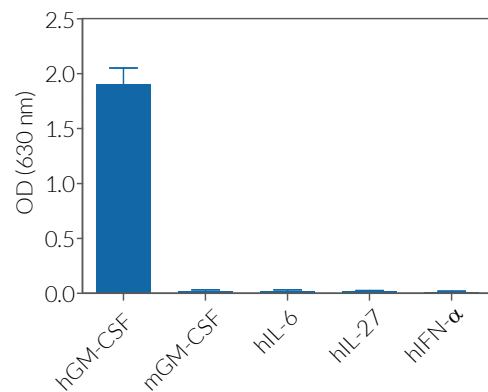


Figure 3. Response of HEK-Blue™ GM-CSF cells to a panel of cytokines. Cells were stimulated with various human and murine recombinant cytokines: 0.1 ng/ml of hGM-CSF, 10 ng/ml mGM-CSF, hIL-6, hIL-27, or 1000 U/ml hIFN- α . After overnight incubation, SEAP activity was assessed using QUANTI-Blue™ Solution. The optical density (OD) at 630 nm is shown as mean \pm SEM.

TECHNICAL SUPPORT

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