

# Validation data for ADP-Heptose

<https://www.invivogen.com/adp-heptose>

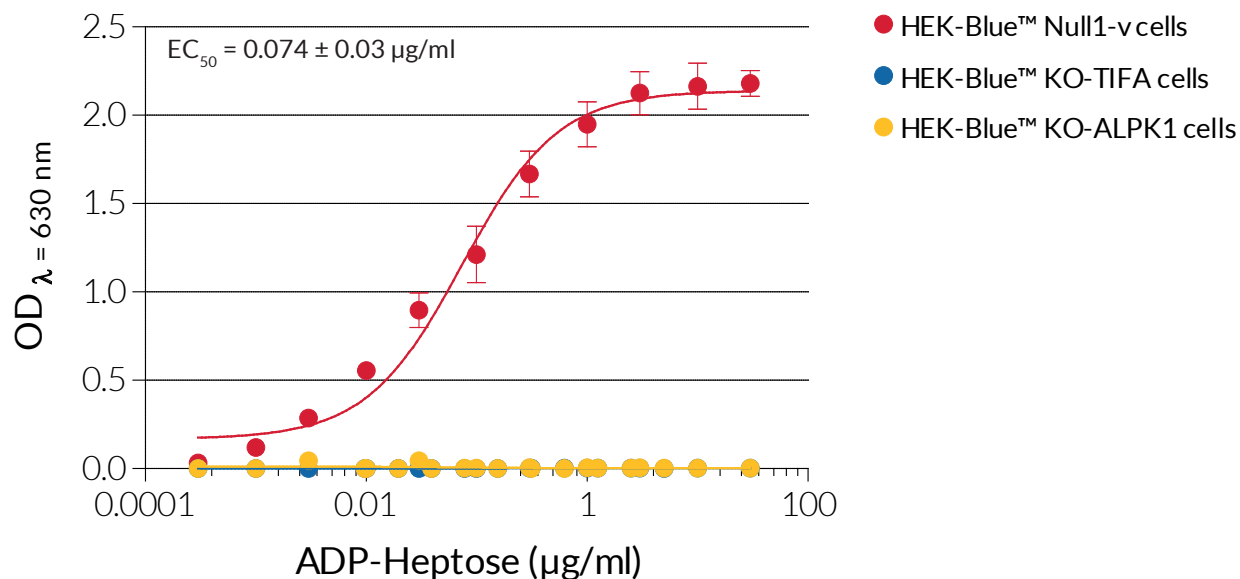
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Version 21F08-ED

InvivoGen has synthesized and purified ADP-D-*glycero*- $\beta$ -D-*manno*-heptose (ADP-Heptose; D isomer), an intermediary sugar in the biosynthesis of lipopolysaccharide (LPS), an essential component of the outer membrane of Gram-negative bacteria. ADP-Heptose has been identified as a potent pathogen associated molecular pattern (PAMP) that binds to the cytosolic pattern recognition receptor (PRR) ALPK1 and activates the ALPK1-TIFA signaling pathway. Ultimately, ADP-Heptose induces a pro-inflammatory NF- $\kappa$ B-dependent response in host cells (**Figure 1**).

## Functional validation of ADP-Heptose

Stimulation of InvivoGen's HEK-Blue™ Null1-v cells (red), which express a NF- $\kappa$ B-inducible secreted embryonic alkaline phosphatase (SEAP), with ADP-Heptose results in a clear dose-dependent response. Furthermore, this activation is dependent upon ALPK1 and TIFA, with no response observed in the derived knockout (KO) cell lines, HEK-Blue™ KO ALPK1 (yellow) and HEK-Blue™ KO-TIFA (blue) cells.



**Figure 1: ADP-Heptose induced NF- $\kappa$ B response.** HEK-Blue™ Null1-v, HEK-Blue™ KO-ALPK1, and HEK-Blue™ KO-TIFA cells were incubated with increasing concentrations of ADP-Heptose (0 - 30  $\mu\text{g/ml}$ ). After overnight incubation, the NF- $\kappa$ B response was assessed by measuring the activity of SEAP in the supernatant using QUANTI-Blue™ Solution, a SEAP detection reagent. Data are presented as optical density (OD) at 630 nm (mean  $\pm$  SEM).  $\text{EC}_{50}$  value is indicated ( $\pm$  std error).

### TECHNICAL SUPPORT

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