Validation data for TL7-887 VacciGrade™

https://www.invivogen.com/tlr7-conjugatable-ligands

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Version 22D21-NJ

TL7-887 is a ready-to-use "pre-linked" conjugatable TLR7 ligand, synthesized from the base molecule CL307, a well-known TLR7 agonist. TL7-887 efficiently triggers a cellular response upon recognition by either human or murine TLR7 **(Figure 1)**. TL7-887 can be used to generate immune-stimulating antibody conjugates (ISACs) as conjugation to a Anti-HER2-hIgG1 and subsequent activation of TLR7 has been validated using cellular assays. The Anti-HER2/TL7-887 ISAC is more potent at inducing a TLR7-mediated response than TL7-887 only in cells expressing HER2 **(Figure 2A)**. Of note, at high concentrations, TL7-887, Anti-HER2/TL7-887 ISAC and Anti-βGal/TL7-887 ISAC, induce a TLR7-mediated cellular response, independently of HER2 expression **(Figure 2A and B)**. This observation could be explained by cellular uptake through endocytosis/pinocytosis.

Biological activity of TL7-887

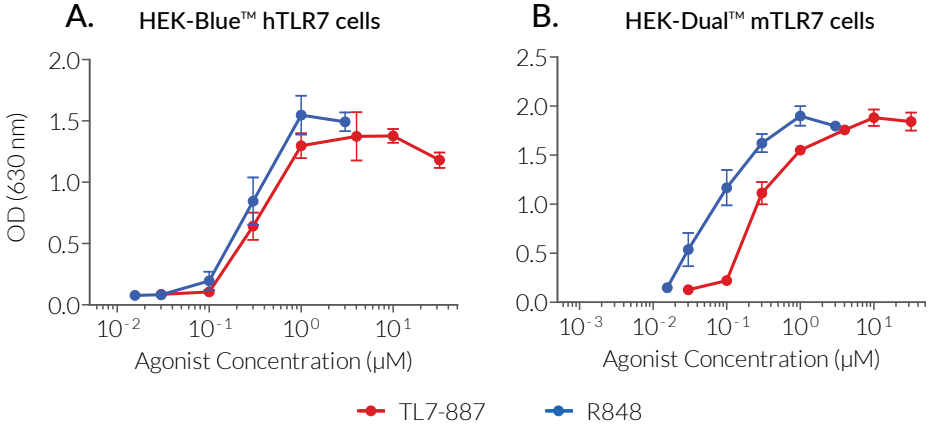

Figure 1: Dose-response of human and murine TLR7 reporter cells to TLR7 conjugatable ligand TL7-887.
~ $4x10^5$ HEK-Blue™ hTLR7 **(A)** or HEK-Dual™ mTLR7 **(B)** cells were stimulated with increasing concentrations of TL7-887 (TLR7 agonist), or R848 (TLR7/8 agonist) as a control. Cells were incubated overnight in HEK-Blue™ Detection, a cell culture medium that allows real-time detection of SEAP activity in the supernatant. The optical density (OD) at 630 nm is shown as mean ± SEM.

Biological activity of TL7-887 conjugated to Anti-HER2-hIgG1

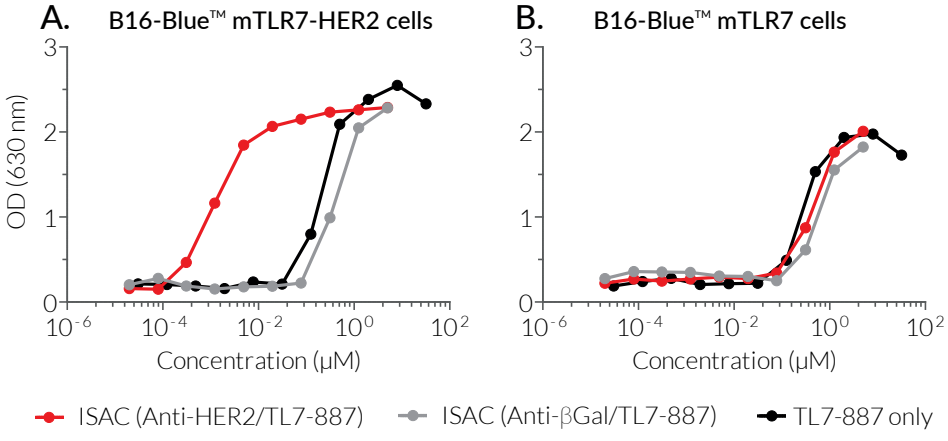

Figure 2: Dose-response of HER2-expressing and murine TLR7 reporter cells to Anti-HER2/TL7-887 ISAC.
~ $5x10^5$ B16-Blue™ mTLR7-HER2 cells **(A)** or B16-Blue™ mTLR7 control cells **(B)** were stimulated with increasing concentrations of Anti-HER2/TL7-887 ISAC (DAR ~8), Anti-βGal/TL7-887 ISAC (DAR ~8), or TL7-887 only. After overnight incubation, the TLR7 response was determined using QUANTI-Blue™ Solution, a SEAP detection reagent. The optical density (OD) at 630 nm is shown.

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