Validation data for RAW-Lucia™ ISG-KO-MAVS cells

For research use only

Version # 16F02-MM

RAW-Lucia™ ISG-KO-MAVS cells were generated from the RAW-Lucia™ ISG cell line through the stable knockout of the MAVS gene. These cells derive from the murine RAW 264.7 macrophage cell line. The knockout of the MAVS gene in RAW-Lucia™ ISG-KO-MAVS cells has been confirmed by PCR and sequencing. Biological activity has been assessed using the Lucia luciferase reporter assay to monitor IRF induction (see figures 1 & 2).

IRF INDUCTION (Lucia luciferase reporter)

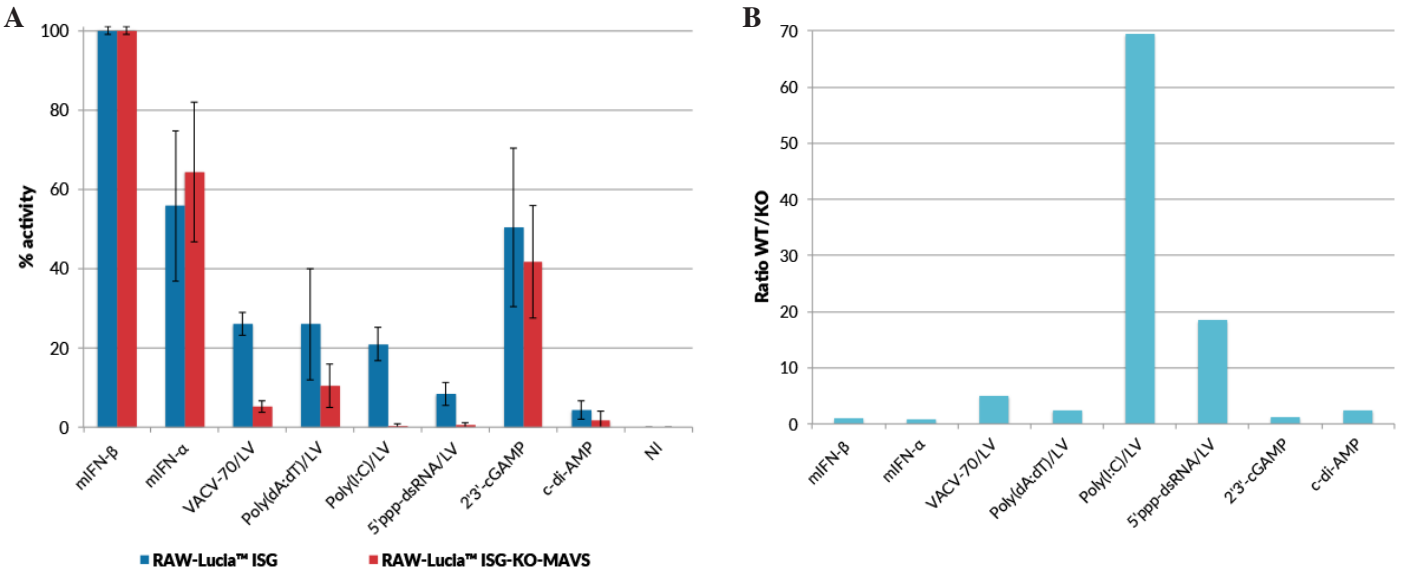

Figure 1: A) Stimulation of RAW-Lucia™ ISG-KO-MAVS and RAW-Lucia™ ISG cells (wild-type cell line) with VACV70/LyoVec™ (1 µg/ml), poly(dA:dT)/LyoVec™ (1 µg/ml), poly(I:C)/LyoVec™ (1 µg/ml), 5'ppp-dsRNA/LyoVec™ (1 µg/ml), 2'3'-cGAMP (3 µg/ml) and c-di-AMP (3 µg/ml). Mouse IFN-α ($1x10^4$ U/ml) and IFN-β ($1x10^4$ U/ml) serve as positive controls. Non-induced cells (NI) have been included as a negative control. After 24h incubation, IRF activation was determined by measuring the relative light units (RLUs) in a luminometer using QUANTI-Luc™, a Lucia luciferase detection reagent. The IRF induction of each ligand is expressed relative to that of mIFN-β at $1x10^4$ U/ml (taken as 100%). B) The difference in activity between the two cell lines is expressed as the ratio WT/KO, which was obtained by dividing each value in figure 1A for RAW-Lucia™ ISG (WT) cells by the corresponding value for RAW-Lucia™ ISG-KO-MAVS cells.

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