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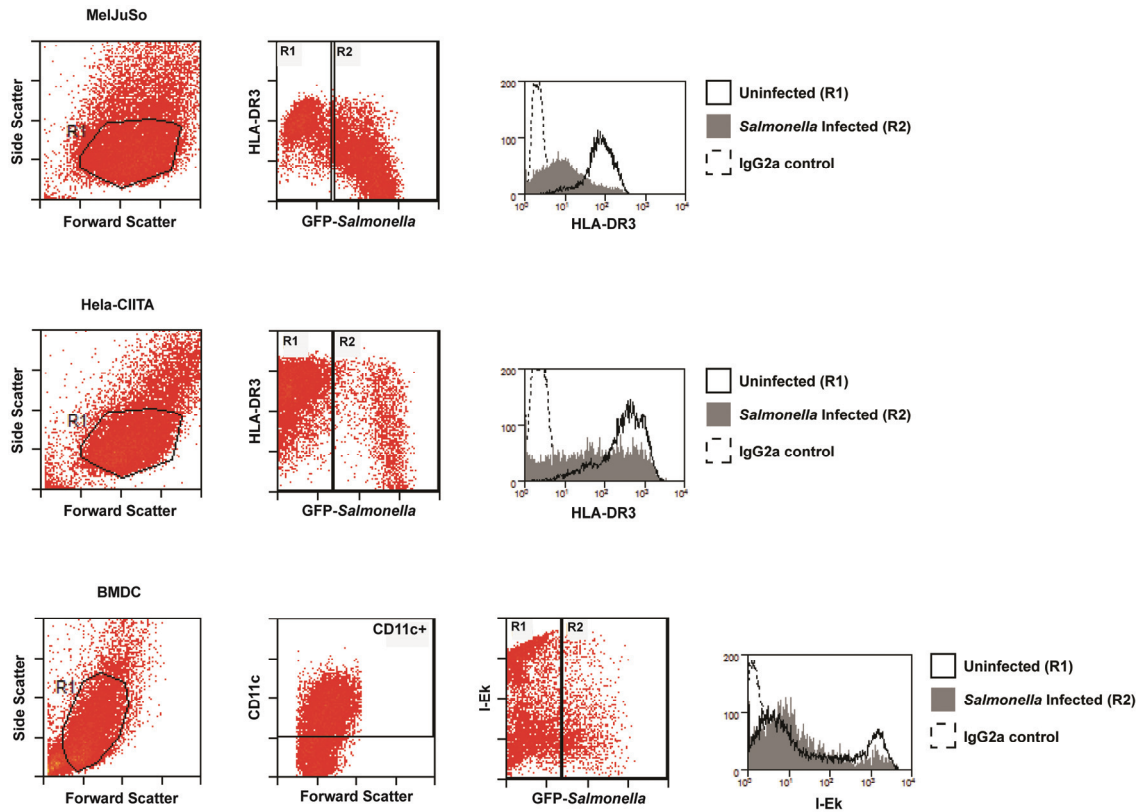
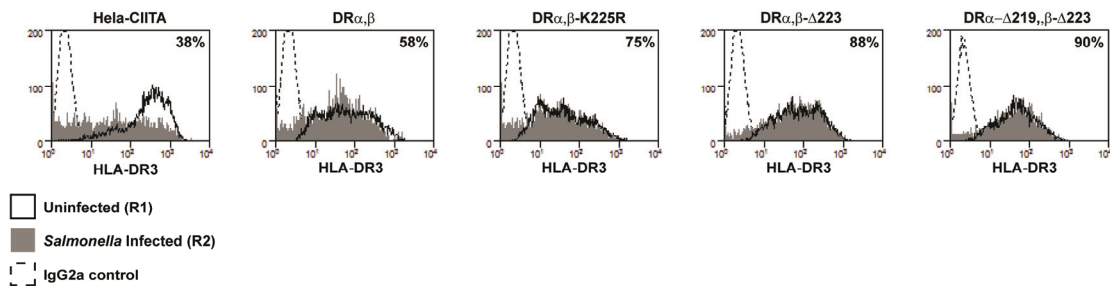
Supporting Information

for

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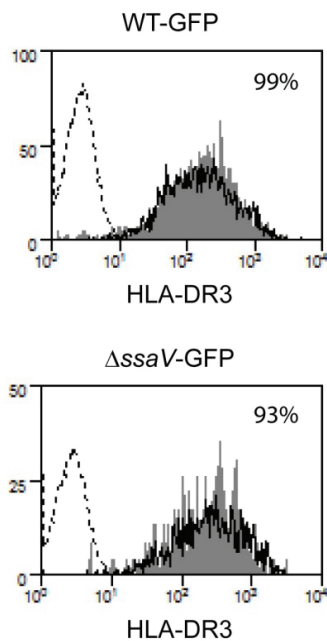
***Salmonella* polarises peptide-MHC-II presentation towards an unconventional
Type B CD4⁺ T-cell response**

A**B**

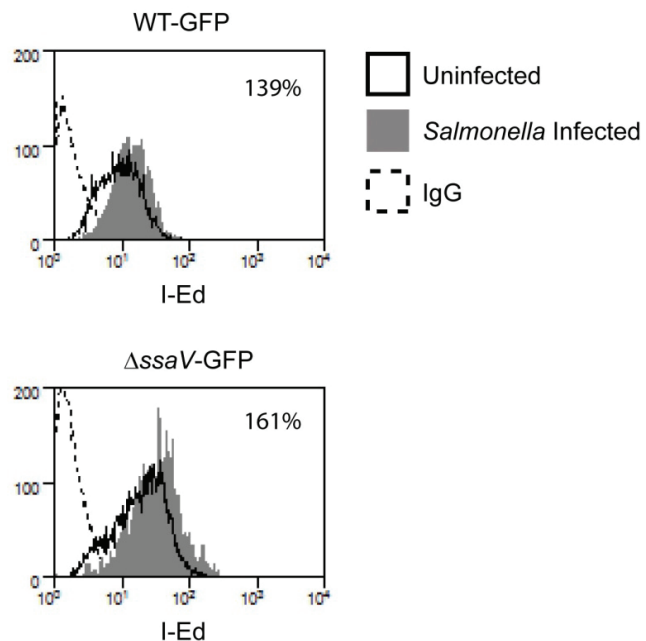
Supporting Information Figure 1: Representative gating strategies for MelJuSo, HeLa-CIITA and BMDCs (A) and flow cytometry plots showing representative MHC-II down-regulation by *Salmonella* for each HeLa-HLA-DR3 transfectant used in Figure 2 (B)

A

Monocyte-derived MΦ



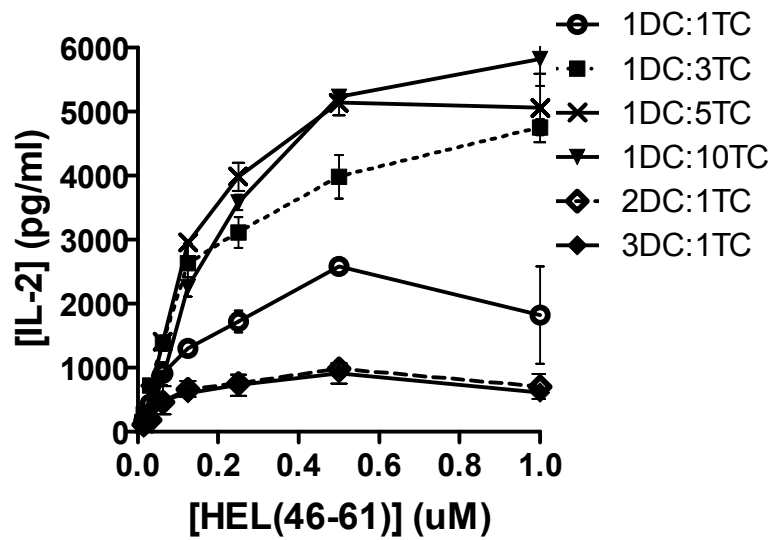
B

RAW264.7_CIITA + IFN- γ 

Supporting Information Figure 2: *Salmonella* does not down-regulate MHC-II in

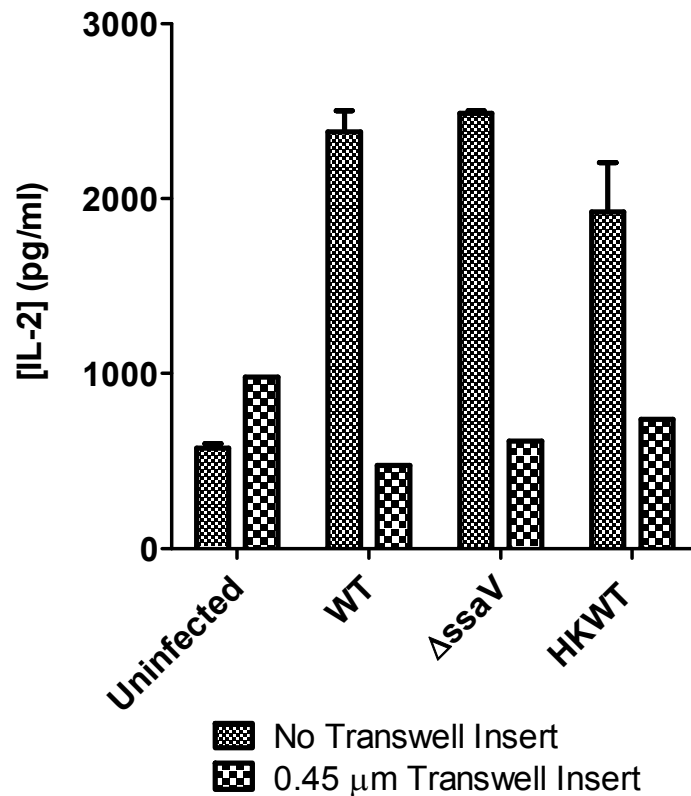
human monocyte-derived macrophage or a murine macrophage cell line (RAW264.7-CIITA)

Monocyte-derived macrophage (MΦ)(A) or IFN- γ -treated RAW264.7-CIITA cells (B) were infected with opsonised WT or SPI2-deficient (Δ ssaV) GFP-S. Typhimurium at an MOI of 50 and HLA-DR3 (L243 reactivity)(A) or I-E^d (14.4.4s reactivity)(B) surface expression was compared in infected (GFP positive) and uninfected (GFP negative) cells by flow cytometry. Histograms show HLA-DR3 (A) or I-E^d (B) surface expression in infected (shaded) or uninfected (clear) monocyte-derived MΦ (A) or RAW264.7-CIITA cells (B) from a representative of four independent experiments. Isotype controls are shown as a dashed line.



Supporting Information Figure 3: Relationship between BMDC: T hybridoma cell ratio and IL-2 response

BMDCs (in triplicate) were incubated with HEL₄₆₋₆₁ peptide and Type B CD4⁺ T hybridoma cells (11A10) at different BMDC: T cell ratios to determine the optimal ratio for infection studies. After 24 h, culture supernatants were harvested and T cell activation was quantified by IL-2 ELISA. Graph shows mean IL-2 concentration from a representative of three independent experiments. Error bars represent SD.



Supporting Information Figure 4: Transwell experiment showing exposure to *Salmonella* is sufficient to enhance presentation of exogenous peptide to Type B T cells

BMDCs (in triplicate) were infected with opsonised WT, SPI2-deficient (Δ ssaV) or HK WT GFP-S. Typhimurium (MOI 10). For antigen presentation, BMDCs were incubated with 0.5 μ M HEL₄₆₋₆₁ peptide and Type B CD4⁺ T hybridoma cells (11A10) at a ratio of 5 T cells: 1 BMDC. Where indicated, infected BMDCs were separated from fresh BMDCs and T hybridoma cells using a 0.45 μ m transwell insert (Millicell-HA, Millipore). After 24 h, culture supernatants were harvested and T cell activation was quantified by IL-2 ELISA. Graph shows mean IL-2 concentration from a representative of three independent experiments. Error bars represent SD.