

Supplementary figures

Figure S1. CEFs infected with EGFP⁺MDV contain cells of the macrophage lineage which are infected with MDV.

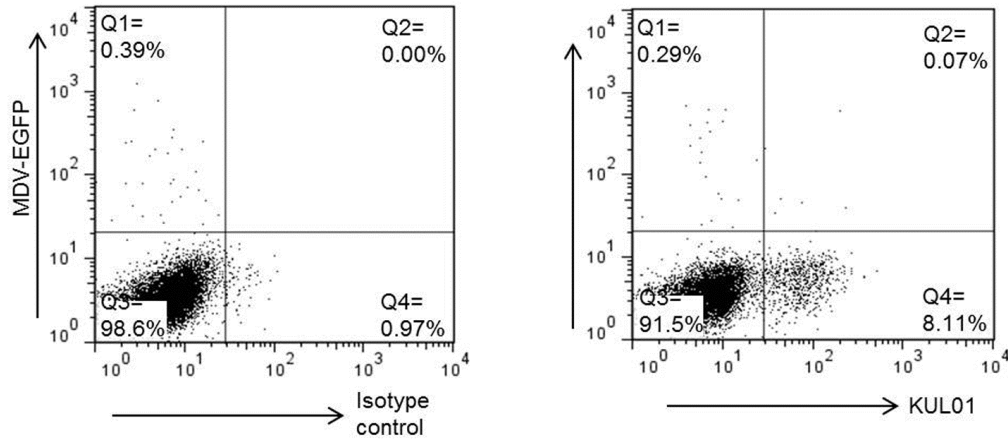
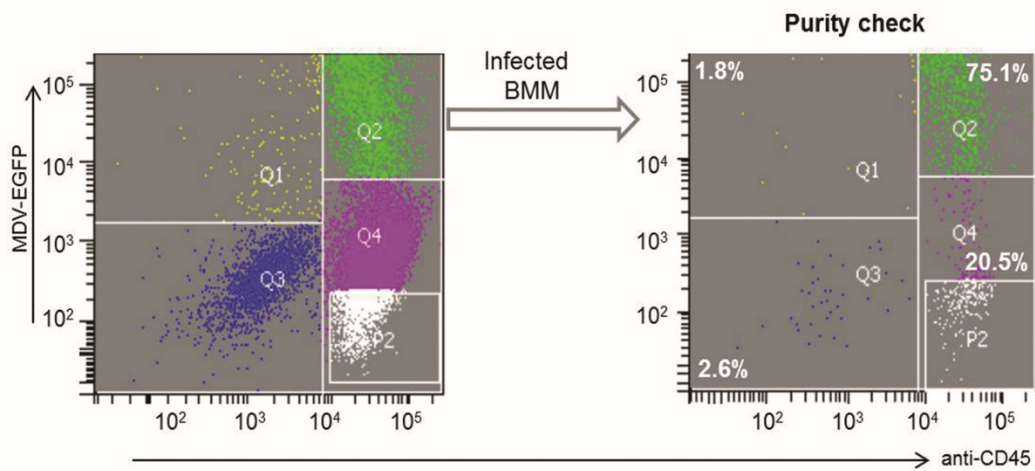


Figure S2. Analysis of MDV-infected BMM (EGFP⁺CD45⁺) reveals that a single round of sorting does not remove all EGFP⁺CD45⁻ CEFs present in the inoculum.



Supplementary figure legends

Fig. S1. Presence of macrophages in CEF cultures and staining pattern of macrophages. (a) CEF cultures naturally contain macrophage-lineage cells. In order to detect macrophage-lineage cells in CEF cultures, previously frozen EGFP expressing MDV-infected CEF were thawed and stained with a mouse monoclonal antibody to chicken macrophages, KUL01 followed by secondary antibody conjugated with AF647. Flow cytometric analysis shows that the CEF culture contains 8% macrophages including a small percentage (0.07%) of EGFP⁺MDV-infected macrophages.

Fig. S2. Purity of infected BMM. *In vitro* derived BMM were infected for 3 days with MDV infected CEF expressing EGFP. Virus positive BMM were selected by cell sorting for CD45⁺EGFP⁺ cells (left). Sorted cells were reanalysed for the presence of contaminant cells by placing 1000 cells in the same sorting plot (right). Whilst the majority of cells were CD45⁺EGFP⁺ (Q2, the sorted population consistently contained around 1-2% infected CEF (Q1) as well as EGFP⁻CEF (Q3) and uninfected BMM (Q4). P2 indicates the sorting zone for uninfected macrophage.