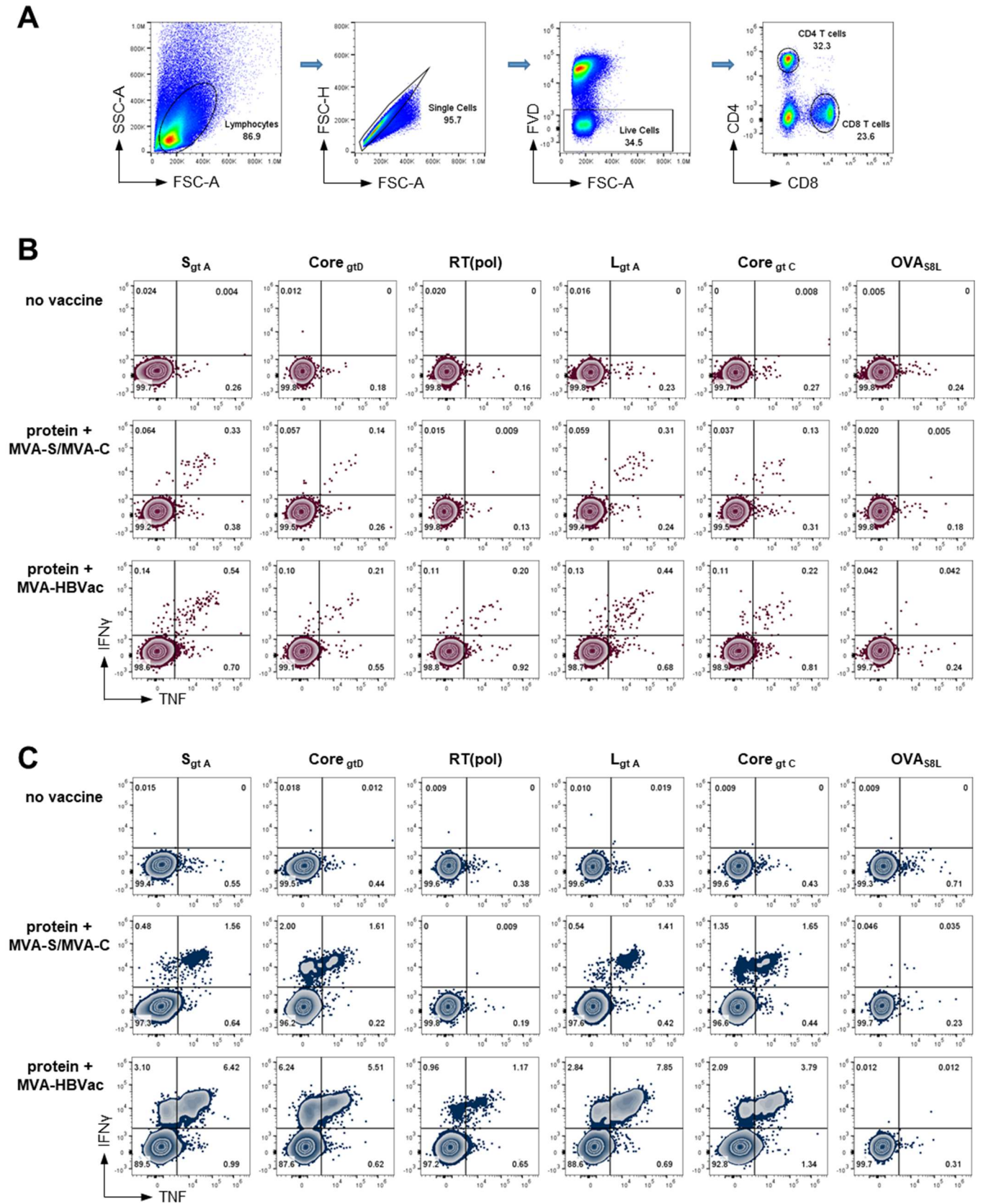


## **Supplemental information**

### **MVA-HBVac—A novel vaccine vector that allows pan-genotypic targeting of hepatitis B virus by therapeutic vaccination**

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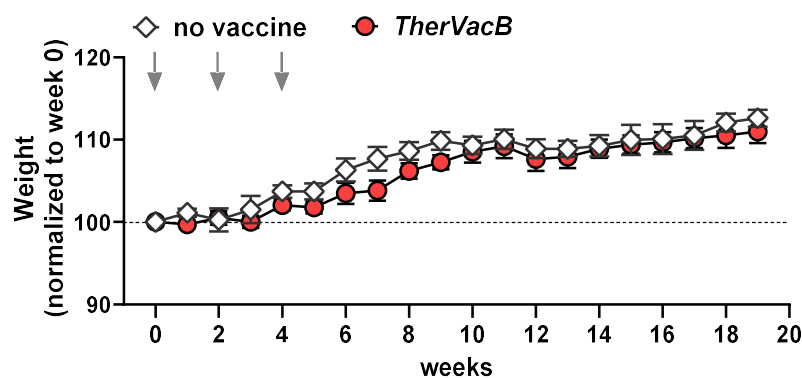
**Figure S1**



**Figure S1. Gating strategy and representative results of flow cytometric analysis.** (A) The lymphocyte population was defined using forward-scattered-light-area (FSC-A) and side-scattered-light-area (SSC-A) parameters. Doublets were excluded from the lymphocyte population by plotting forward-scattered-light height (FSC-H) against area (FSC-A) and gating on the diagonal

population. Dead cells were excluded from analysis by gating on fixable viability dye (FVD-eFluor780-A)-negative cells. CD4 or CD8 T cells were gated within the viable lymphocyte population using CD4 APC-A and CD8 PB450-A. (B-C) Representative dot-plots of HBV-specific IFN $\gamma$ <sup>+</sup> TNF<sup>+</sup> CD4 (B) and IFN $\gamma$ <sup>+</sup> TNF<sup>+</sup> CD8 T cells (C) detected by intracellular cytokine staining (ICS) of isolated splenocytes after 16h *ex vivo* stimulation with 5 overlapping peptide pools covering the proteins expressed by MVA-HBVac. Positioning of gates was determined using controls stimulated with ovalbumin (OVA)-derived peptide S8L<sub>257-264</sub> (OVA<sub>S8L</sub>; SIINFELK). A–area, H – height.

**Figure S2**



**Figure S2. Monitoring of mouse weight.** C57BL/6 mice (n=5) were intravenously injected with 4x10<sup>9</sup> GE of an AAV vector carrying a 1.2-fold overlength HBV genome of gtD. *TherVacB* vaccination was initiated 6 weeks after AAV-HBV infection (week 0), and mice were sacrificed 15 weeks after the MVA-HBVac boost (week 19). The body weight of the mice was monitored weekly. The presented values from the beginning of vaccination (week 0) until the end-analysis (week 19) were normalized according to the baseline values at week 0. Arrows indicate the time points of vaccination.