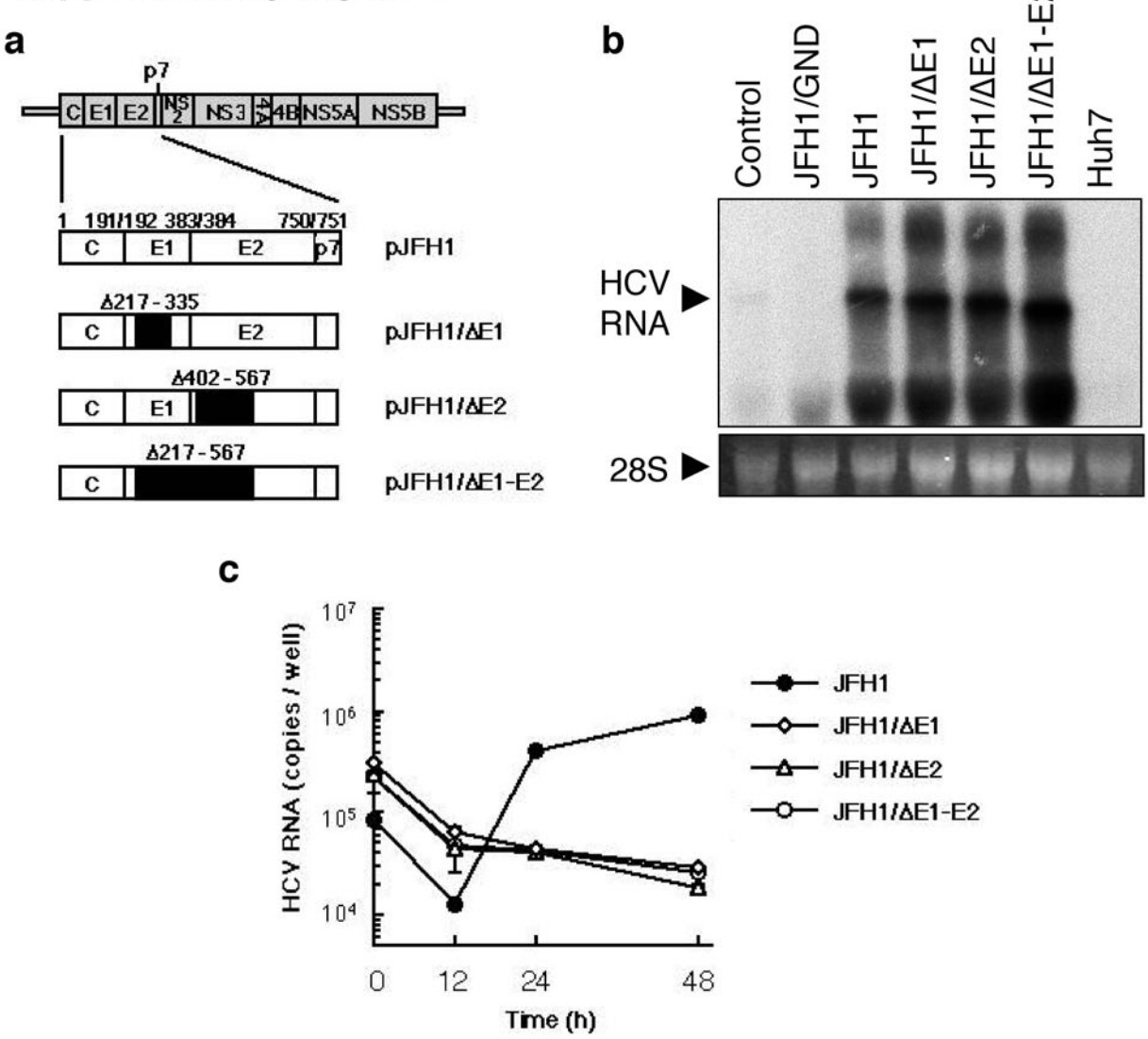
Supplementary Figure 4



Supplementary Fig. 4: Virus particle release depends on envelope glycoproteins. (a) Maps of JFH1 HCV constructs with deleted regions indicated by closed boxes and amino acid positions specified above. pJFH1/ΔE1, pJFH1/ΔE2 and pJFH1/ΔE1-E2 contain a 119 amino acid deletion (aa 217-335), a 166 amino acid deletion (aa 402-567) or a 351 amino acid deletion (aa 217-567), respectively. (b) Northern blot analysis of HCV RNA replication after RNA transfection. Transfected Huh7 cells were harvested 48 h after transfection. Control, positive controls comprised 10^7 copies of synthetic JFH-1 RNA; Huh7, cellular RNA isolated from naive Huh7 cells. Arrowheads indicate full-length HCV RNA (HCV RNA) and 28S ribosomal RNA (28S). Similar levels of RNA replication of JFH1/ΔE1, ΔE2 and ΔE1-E2 RNA were observed compared to JFH-1 RNA. (c) HCV RNA replication in infected Huh7 cells. Cells were inoculated with concentrated supernatant of full-length JFH-1 RNA, JFH1/ΔE1, JFH1/ΔE2, or JFH1/ΔE1-E2 RNA-transfected cells. In all cases, HCV RNA titers of concentrated culture supernatants used for inoculation were adjusted to identical HCV RNA copy numbers (2×10^7 copies/ml). Cells were harvested at 0, 12, 24 and 48 h after infection and HCV RNA was determined by RTD-PCR. Experiments were performed in triplicate, and mean titers are shown with SD (bars). Inspite of comparable RNA replication, efficient core protein release occurred only with JFH1 RNA-transfected cells.