Supporting Information

Yu et al. 10.1073/pnas.1310779111

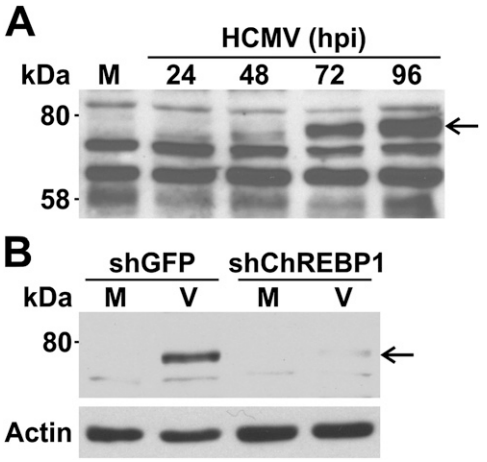

Fig. S1. A potential ChREBP-β protein is detected in human cytomegalovirus (HCMV)-infected cells. (*A*) Whole-cell extracts from an HCMV infection time course, as described in Fig. 1*B*, were analyzed by Western blot analysis using anti-ChREBP antibody. The arrow indicates an induced band that migrates at the predicted molecular weight of ChREBP-β. (*B*) Whole-cell extracts from mock- and HCMV-infected cells treated with shGFP and shChREBP1 were evaluated by Western blot analysis using anti-ChREBP and anti-actin antibodies. The band migrating at the predicted molecular weight of ChREBP-β is depleted by shChREBP1.

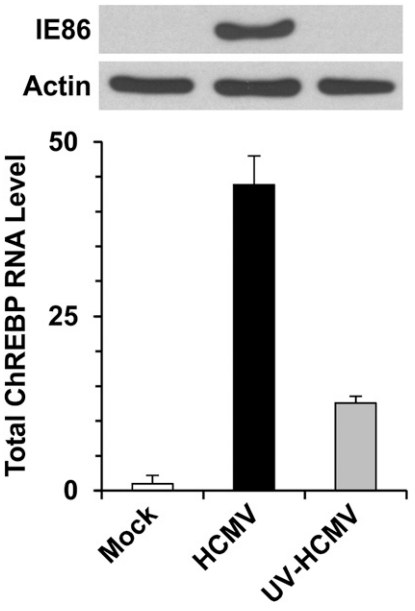

Fig. S2. Total ChREBP RNA levels in cells infected with UV-irradiated HCMV at 48 hpi. Before infection, the virus was exposed to 254-nm UV light at 480 mJ/cm^2 in a Stratalinker 1800 UV crosslinker (Stratagene). HFs were infected with either HCMV or UV-irradiated HCMV (UV-HCMV) for 48 h, after which total ChREBP RNA levels were measured by quantitative PCR and normalized to actin RNA levels. IE86 and actin protein levels in whole-cell extracts were measured by Western blot analysis to ensure that the UV-irradiated viruses were incapable of gene expression.

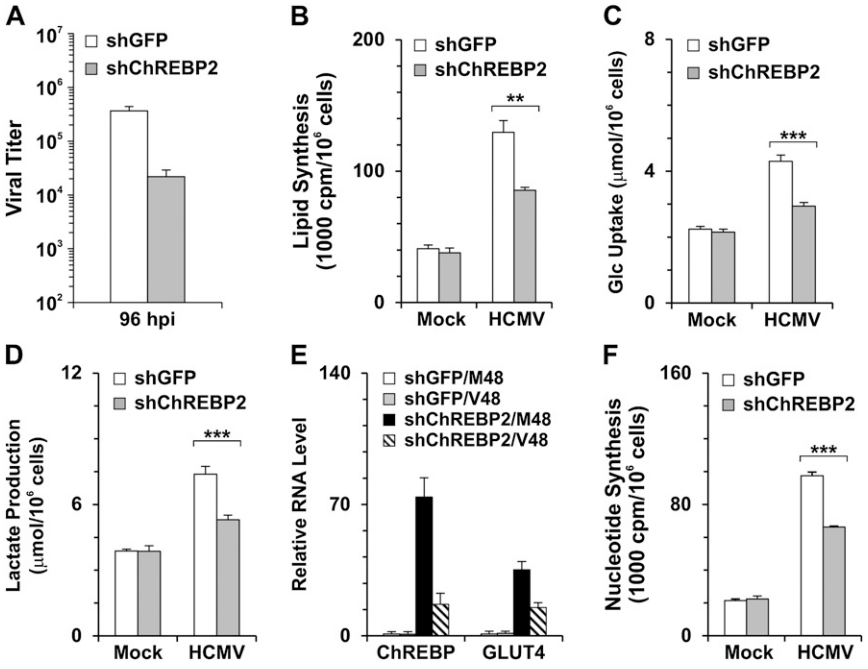

Fig. S3. Effect of ChREBP depletion by shChREBP2 on metabolism and HCMV growth. HF cells were treated with shGFP or shChREBP2 before HCMV infection as described in Fig. 4. Metabolic assays were performed at 48 hpi; and viral titers were measured at 96 hpi. (*A*) HCMV viral titers in HF cells treated with shGFP or shChREBP2. (*B*) Total lipid synthesis in HF cells. Total lipid synthesis was assayed by measuring the incorporation of ^{14}C-acetate into lipids. (*C*) Glucose uptake. Glc, glucose. (*D*) Lactate production. Levels of glucose and lactate in the cultured medium were measured with the YSI 7100 Multiparameter Bioanalytical System. (*E*) Depletion of ChREBP by shChREBP2 reduces glucose transporter (GLUT) 4 RNA levels in HCMV-infected cells. GLUT4 RNA levels were measured at 48 hpi as described in Fig. 4*B*. (*F*) Reduced nucleotide biosynthesis in infected cells from depletion of ChREBP by shChREBP2. Total nucleotide synthesis was assayed by measuring the incorporation of D-[U-^{14}C]-glucose into RNA at 48 hpi. M48, mock infection at 48 hpi; V48, HCMV infection at 48 hpi; $^{**}P < 0.005$; $^{***}P < 0.002$.

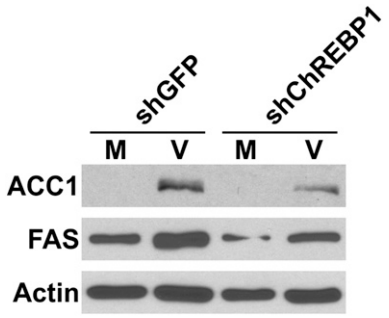

Fig. S4. Increases in levels of of lipogenic enzymes acetyl-CoA carboxylase 1 (ACC1) and fatty acid synthetase (FAS) by HCMV infection are diminished by ChREBP depletion. HF cells treated with shGFP or shChREBP1 were mock- or HCMV-infected for 48 h. Whole-cell extracts were evaluated by Western blot analysis using anti-ACC1, anti-FAS, and anti-actin antibodies.

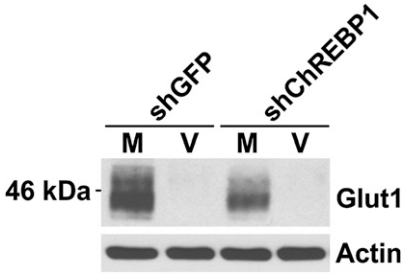

Fig. S5. Depletion of ChREBPs in normal HF cells decreases GLUT1 protein levels. HFs were treated with shGFP or shChREBP1 for 5 d. Whole-cell lysates were analyzed by Western blot analysis using anti-GLUT1 (NB300-666; Novus Biologicals) and anti-actin antibodies.