Fig. S1. Production of infectious virus by HHV-6 infected LECs. LECs were infected with cell-free HHV-6A or HHV-6B, as described in Materials and Methods, and 5 days p.i. were cocultured with target lymphoid cells (JJhan for HHV-6A and SupT1 for HHV-6B) or culture supernatant was used to infect target lymphoid cells. Lymphoid cells were analyzed for signs of HHV-6 infection. The images show JJhan cells, 10 days p.i. Similar results were obtained in SupT1 cells. The development of a typical cytopathic effect was observed both in JJhan cells infected with culture supernatant from infected LECs (A, Left) and by coculture with infected LECs (A, Right). IFA staining for gp116 virus antigen showed bright positivity both in JJhan cells infected with LECs culture supernatant (B) and in cells infected by coculture with infected LECs (C). All images were taken at 10 d.p.i. with a Nikon Eclipse TE2000-S microscope equipped for phase contrast and fluorescence observation (magnification, ×100). The same field is shown: bright field observation (Left), DAPI nuclear stain (Middle), and HHV-6 specific IFA for gp116 (Right).
Fig. S2. Effects of HHV-6B infection on organization of LECs on BME. HHV-6B (strain CV) infection inhibited LECs organization into capillary-like networks. The inhibitory effect was partial 3 days post infection (d.p.i.) (B) and total 7 d.p.i. (D), whereas mock-infected LECs spontaneously formed tubes both at day 3 (A) and day 7 (C). Pictures were taken after 6 h incubation at 37 °C (original magnification, ×10). Data are representative of three independent experiments with similar results.
Fig. S3. Expression of HHV-6 U94/rep deleted mutants pSR2pH-ΔC and pSR2pH-ΔN does not inhibit network formation on BME. HUVECs were transfected with pSR2pH-ΔC or pSR2pH-ΔN plasmids and transferred on BME-coated plates 48 h after nucleofection. No differences in the capability to form the capillary-like network were observed when cells expressing the C-deleted (pSR2pH-ΔC) and the N-deleted (pSR2pH-ΔN) forms of U94/rep were compared both with not transfected (N.T.) and pSR2-transfected (pSR2) HUVECs. Images were taken 6 h after plating (original magnification, ×10). Data are representative of three independent experiments with similar results.