



Attenuation of Zika Virus by Passage in Human HeLa Cells

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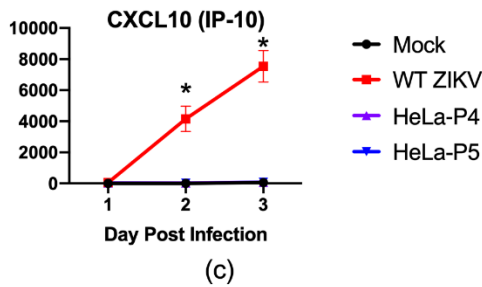
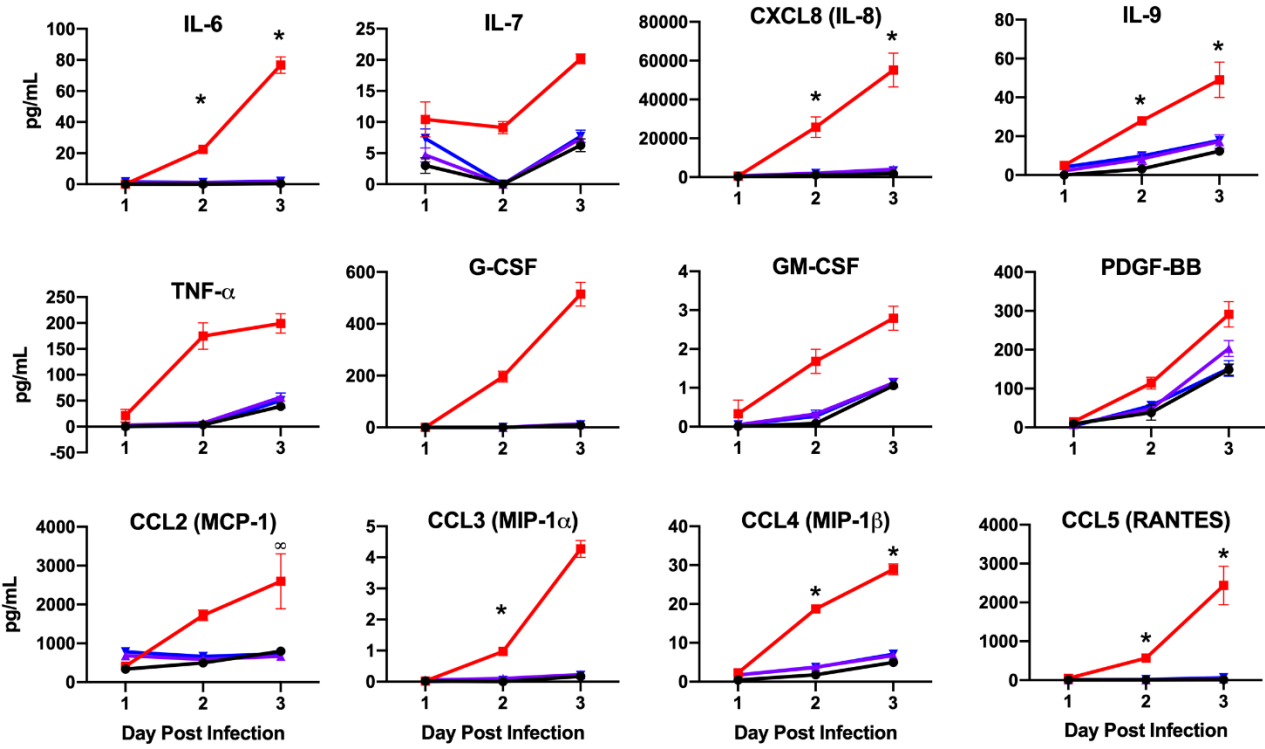
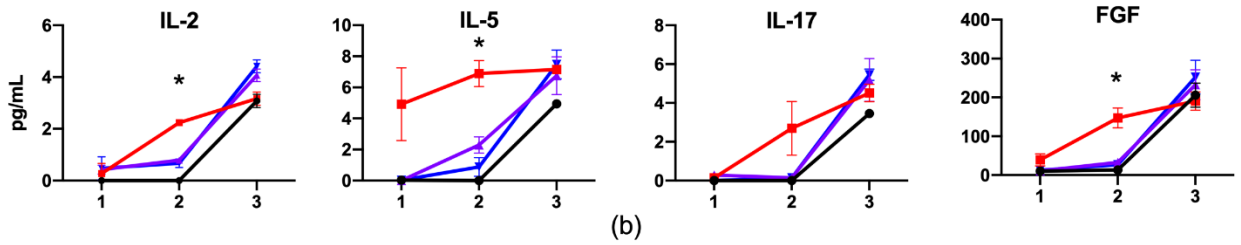
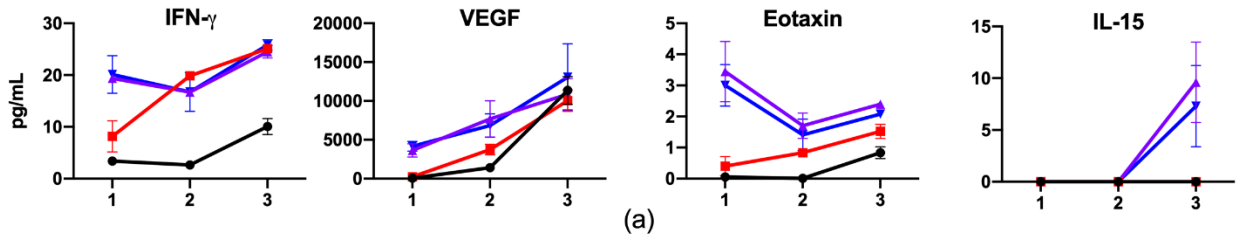


Figure S1. Passaging of ZIKV in HeLa cells attenuates cytokine production in A549 epithelial cells. Secreted cytokine levels from A549 cells infected with WT ZIKV, HeLa-P4, HeLa-P5, or mock at 1, 2, or 3 days p.i. were quantified from 50 microliter samples ($n = 2$). (a) Cytokine and chemokine levels increased on day 1, 2, or 3 following infection with HeLa-P4 and HeLa-P5 but not with WT ZIKV; (b) Cytokines and chemokines increased by infection with WT ZIKV on day 2 p.i. or (c) on days 2 and 3 p.i. Not shown: IL-1 β , IL-4, IL-12p70, and IL-13 were detected at very low levels (< 2 pg/mL); IL-1RA and IL-10 were not detected in any samples. Kruskal-Wallis tests with Dunn's multiple comparisons were used to compare WT ZIKV, HeLa-P4, and HeLa-P5 to mock and to compare mock, HeLa-P4, and HeLa-P5 to WT ZIKV. Error bars represent standard deviation. $*p < 0.05$ WT ZIKV compared to mock; $\infty p < 0.05$ WT ZIKV compared to HeLa-P4 or HeLa-P5. No differences were detected between HeLa-P4 and HeLa-P5 induction of cytokines.