



Supplemental Figure 4. Transient expression of DUSP11 rescues BLV 5p miRNA accumulation and RISC activity in DUSP11-KO cells.

(A) Western blot analysis of cells transfected with pcDNA3.1-EV, the DUSP11 expression vector (pDUSP11), or the DUSP11-C152S catalytic mutant expression vector (pDUSP11-C152S). (B) Northern blot analysis of BLV-B2 and BLV-B5 miRNAs in parental (WT) and DUSP11-KO HEK293T cells. Cells were first transfected with pcDNA3.1-EV, pDUSP11, or pDUSP11-C152S. Cells were then co-transfected with pHSUR4 and either pBLV-B2 or pBLV-B5. The membrane was first blotted with probes specific for the 5p miRNAs, stripped and re-probed for the 3p miRNAs, and then stripped and re-probed for HSUR4 RNA. Note, the blot for the B2 and B5 pre-miRNAs was probed with the respective 5p probes. (C) Luciferase assay to measure the RISC activity of BLV-B5 miRNAs in parental (WT) cells co-transfected with pcDNA3.1-EV and in DUSP11-Ko HEK293T cells co-transfected with pcDNA3.1-EV, pDUSP11, or pDUSP11-C152S. Bars represent the mean ratio (Renilla/Firefly) +/- SEM (n=5) from five experiments performed in triplicate.