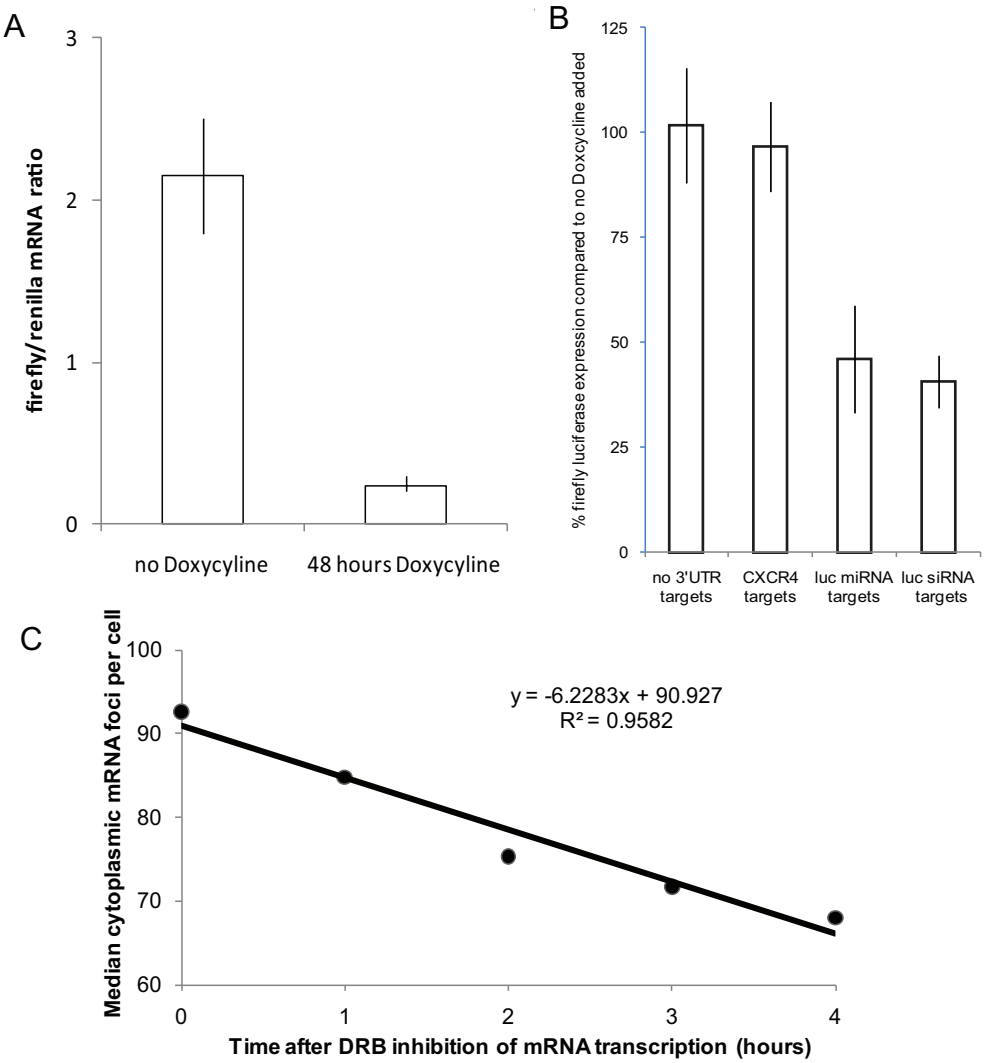
Supplemental Figure S1



Supplemental Figure S1. A) Firefly luciferase mRNA levels decrease 10-fold after doxycyline induction of miRNA. Quantitative RT-PCR of firefly luciferase mRNA levels normalized to a renilla luciferase mRNA control. N = 9 replicates per data point. B) Luc miRNA specifically targets and represses complementary target sequences. Plasmid expressing luc miRNA under doxycycline induction was transiently co-transfected with luciferase reporters containing no 3'UTR targets, mismatched CXCR4 targets, imperfectly complementary miRNA targets, or perfectly complementary siRNA targets. Doxycycline was added 24 hours post-transfection and luciferase levels were assayed 48 hours after doxycyline addtion. Each target under induction was compared to the same transfection without doxycyline addition grown for the same amount of time. N = 3 replicates per data point. C) mRNA lifetime calculation. Cells were fixed 0 - 4 hours after DRB inhibition of mRNA transcription and smFISH stained for luciferase mRNA. Median cytoplasmic mRNA foci for each hour (circles) was used to extrapolate a linear mRNA decay equation shown in the figure. N = 53 cells for 0 hours, 51 cells for 1 hour, 53 cells for 2 hours, 47 cells for 3 hours, and 47 cells for 4 hours.