We damaged cells treated with the XRCC4 shRNA and analyzed their rates of DSB joining. We found that a 50% reduction in XRCC4 protein levels did not generate a delay in DSB repair. Rather, we were surprised to find that the half-life of 53BP1 foci was shorter, indicating faster repair, in the knock down cells compared to normal controls (Figure 3.5E). At present we cannot explain why a reduction in XRCC4 protein levels increases the rates of DSB repair.

We analyzed how the reduced XRCC4 expression affects the dynamical properties of the p53 pulse in response to DNA DSBs. We found no difference in the amplitude of the p53 pulse between cells treated with the XRCC4 shRNA and normal controls (Figure 3.5F, G). However, the duration of the p53 pulse significantly increased in the knock down cells compared to normal controls (Figure 3.5F, H, p-value 0.007, t-test). The wider p53 pulses observed in the knock down cells are qualitatively similar to those observed in response to UV treatment, where p53 is primarily activated by the upstream kinase ATR (Ataxia telangiectasia and Rad3 related). We hypothesize that abatement of XRCC4 protein levels might prevent the effective ligation of a few DSBs in damaged cells. A failure to ligate DNA ends potentially promotes an increased resection of DNA at these break sites in an attempt to improve the annealing between the DNA strands and promote re-ligation. The resected ssDNA strands may then recruit and activate ATR, which subsequently modifies p53 and increases the duration of its pulse. Future work employing specific ATR inhibitors will aid the investigation of this hypothesis.