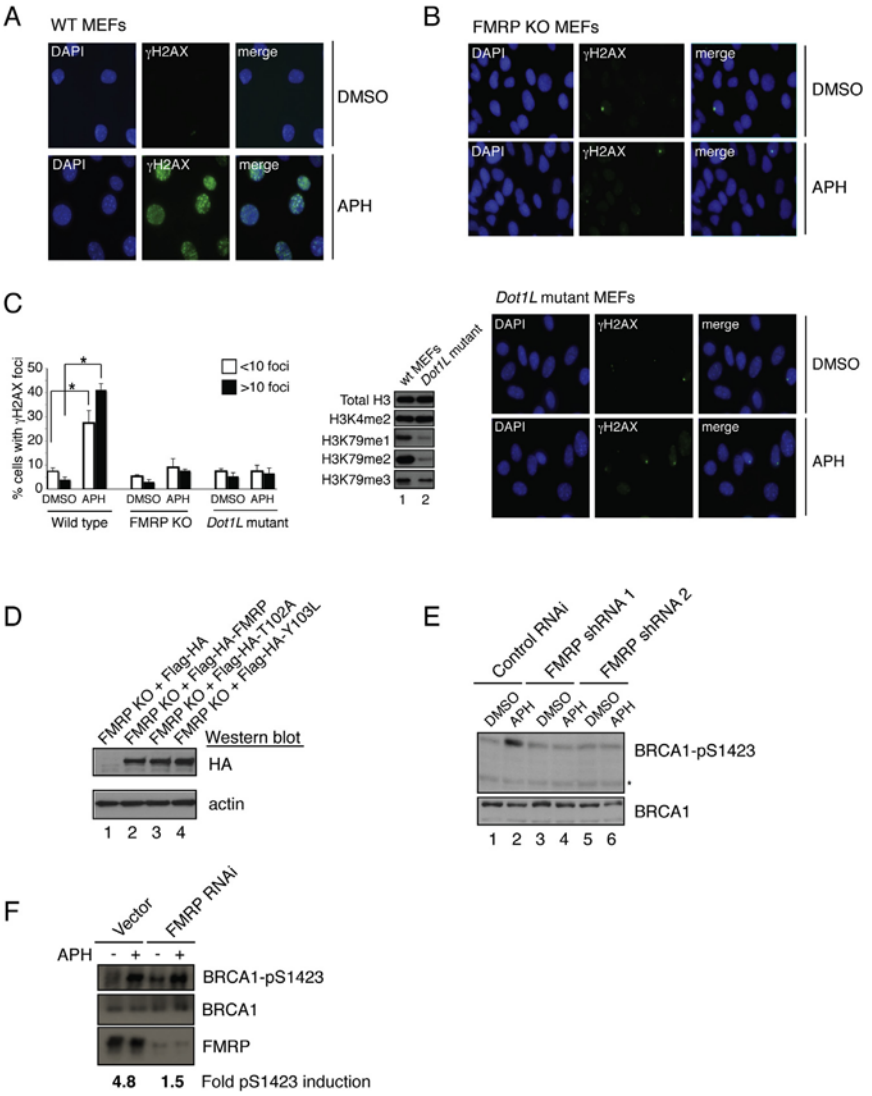

Figure S1. FMRP-Mediated γH2A.X Induction and BRCA1 Phosphorylation, Related to Figures 1, 3, and 7

(A and B) Induction of γH2A.X foci formation is impaired in FMRP KO (B) as compared to wild-type MEFs (A) in response to aphidicolin (APH) treatment.

(C) Left graph, wild-type but not FMRP KO and *Dot1L* mutant MEFs exhibited a 3-fold (cells with < 10 foci) and 4-fold (cells with > 10 foci) increase in γH2A.X foci in response to APH. Asterisks, $p < 0.05$, Student t test. Data are represented as average of three independent experiments with SD. Western blot shows H3K79 methylation levels in WT and *Dot1L* mutant MEFs. Right: induction of γH2A.X foci formation in response to APH is impaired in *Dot1L* mutant MEFs as compared to wild-type MEFs (A).

(D) Western blot showing the levels of expression of exogenous Flag-HA-FMRP (lane 2) and FMRP Agenet domain mutants (lanes 3 and 4) in FMRP KO MEFs.

(E) HeLa cells were subjected to scrambled or two independent FMRP shRNAs (FMRP shRNA1 and FMRP shRNA2). Cells were then treated with DMSO as a control or APH for 24 hr, followed by western blotting for BRCA1 phospho-Ser1423. In contrast to control RNAi cells (3.5-fold induction in Ser1423 phosphorylation), FMRP RNAi cells showed no phosphorylation of BRCA1 at Ser1423 in response to APH treatment (compare lane 2 to 4 and 6). Asterisk indicates a nonspecific band.

(F) Unlike in HeLa cells, in 293 cells FMRP RNAi-mediated dampening of phospho-Ser1423 could be partially attributed to the increased levels of phospho-Ser1423 even in the absence of APH treatment, suggesting that the FMRP effect on BRCA1 phosphorylation might be context dependent. Blots are representative of three experiments.