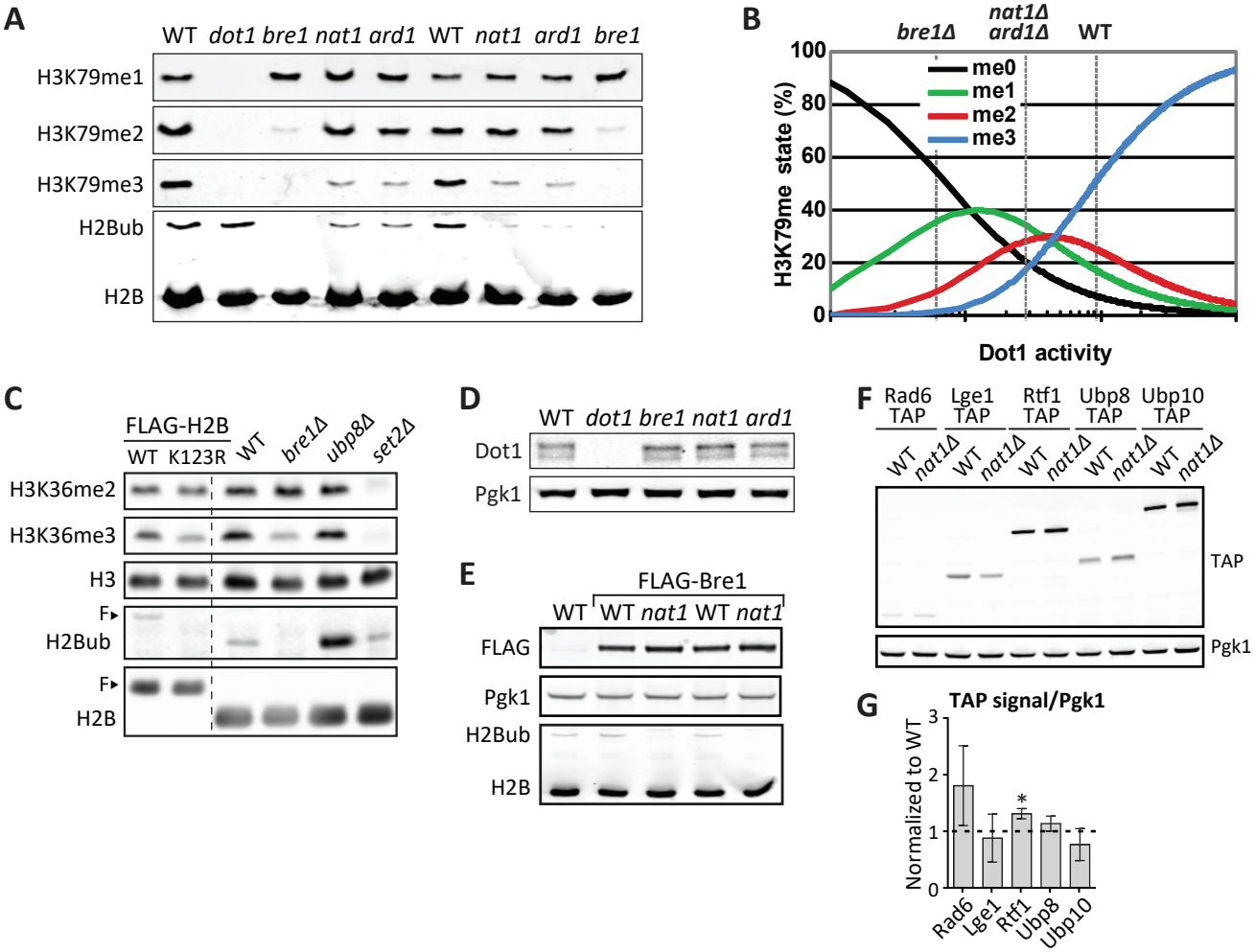

Figure 3 -figure supplement 1. NatA immunoblots.

A) Immunoblots of H3 methylation and H2B in the indicated deletion strains. Biological replicates of Figure 3A. B) Model of the dependence of H3K79 methylation on Dot1 activity (modified from De Vos et al. (2011)), with dashed lines indicating the estimated Dot1 activities in the indicated strains. The decrease in H3K79me3 and increase in K79me1 fit with lower overall Dot1 activity, but the effect of deletion of the NatA complex is not as strong as BRE1 deletion. C) Immunoblots showing the effect of an H2B-K123R mutation and *BRE1* deletion on H3K36me2 (no effect) and H3K36me3 (decrease). Bands shifted by the extra weight of the FLAG tagged have been marked with an F. D) Dot1 expression levels are unaltered in *nat1Δ* and *ard1Δ* strains. E) Because a C-terminal tag on Bre1 disrupts its function (Wood et al., 2013) and an N-terminal tag interferes with potential N acetylation of the native N terminus of Bre1, we performed an epistasis experiment to address whether the role of NatA in H2B ubiquitination was mediated by N-acetylation of Bre1. FLAG-Bre1 has a normal activity that is still NatA dependent, which demonstrates that the NatA complex does not act though N-acetylation of Bre1. F) TAP blots were used to measure the expression levels of C-terminally TAP-tagged versions of the indicated proteins. Representative blot. G) Quantification of the blot shown in panel F, as well as blots from two independent experiments. Only Rtf1 expression is significantly altered, but an increase in Rtf1 is not consistent with a decrease in H2Bub.