Figure S1. Excluding an influence of the FLAG tag on PRC2-RNA interactions.

(A) Western blot of Flag-PRC2 complexes treated (+) or mock treated (-) with rEnterokinase to remove the FLAG tag. Top: Anti-FLAG. Bottom: Anti-EZH2. (B) EMSA shows similar pattern of RNA recognition by PRC2 containing the FLAG-tag or after FLAG-tag removal, suggesting no influence of the FLAG-tag on PRC2-RNA binding. FLAG-GFP [200nM] was used as control. (Left) RepA I-IV. (Right) MBP 1-300. Figure S1 relates to main Figure 1.



Figure S2. RNA-binding potential of PRC2 subunits and subcomplexes to truncated RepA and HOTAIR RNAs.

(A-D) RNA EMSA to test binding of single PRC2 subunits and PRC2 subcomplexes to truncated RNAs.

(A) EZH2. (B) SUZ12. (C) EED. (D) Subcomplexes as indicated. EZ, EZH2. EE, EED. S, SUZ12.

Probes are used at 2 nM throughout. Figure S2 relates to main Figures 1 and 3.

Α EZH2 [nM] EZH2 [nM] EZH2 [nM] EZH2 [nM] 1000 1000 GST 40 200 1000 GST 40 200 GST 1000 GST 40 200 200 9 RepA I-II mHotair 1-150 P4-P6 hHOTAIR 1-200 В SUZ12 [nM] SUZ12 [nM] SUZ12 [nM] SUZ12 [nM] GST 40 200 1000 1000 40 200 1000 GST 40 200 1000 GST 40 200 GST hHOTAIR 1-200 RepA I-II mHotair 1-150 P4-P6 С EED [nM] EED [nM] EED [nM] GST 40 200 1000 1000 40 200 1000 GST 40 200 GST RepA I-II mHotair 1-150 P4-P6 D EZ+EE [nM] EZ+EE [nM] S+EE [nM] S+EE [nM] EZ+EE [nM] S+EE [nM] 40 200 1000 1000 1000 1000 GST 1000 GST 1000 200 GST 40 200 GST 40 200 GST 40 200 GST 200 4 RepA I-II mHotair 1-150 P4-P6 RepA I-II mHotair 1-150 P4-P6 EZ+S+EE [nM] EZ+S+EE [nM] EZ+S+EE [nM] EZ+S [nM] EZ+S [nM] EZ+S [nM] 1000 1000 1000 40 200 1000 GST 40 200 GST 200 1000 GST GST 40 200 001 200 GST 40 200 GST 6 6 RepA I-II mHotair 1-150 P4-P6 RepA I-II mHotair 1-150 P4-P6

Figure S3. Effect of JARID2 on the binding affinity of PRC2 for RNA.

Binding isotherms of co-purified PRC2-JARID2 complexes to 2 nM of indicated RNA species. Equilibrium dissociation constants (K_d) and R² values shown in the table. N/A, not applicable because curves were too flat. ">1000 nM" denotes a K_d in excess of that which could be measured. Values for PRC2 + RNA and JARID2 + RNA are shown in Figures 2B and 5B. This Figure relates to main Figure 5.



→ PRC2:JARID2 + RepA
→ PRC2 + RepA
→ JARID2 + RepA
→ PRC2:JARID2 + MBP
→ PRC2 + MBP
→ JARID2 + MBP

EMSA with co-purified PRC2:JARID2

RNA	Kd [nM]	R ²
RepA I-IV	344.3±71.6	0.982
MBP	>1000	N/A