Appendix View

Structural basis of RNAPII transcription on the nucleosome containing histone variant H2A.B

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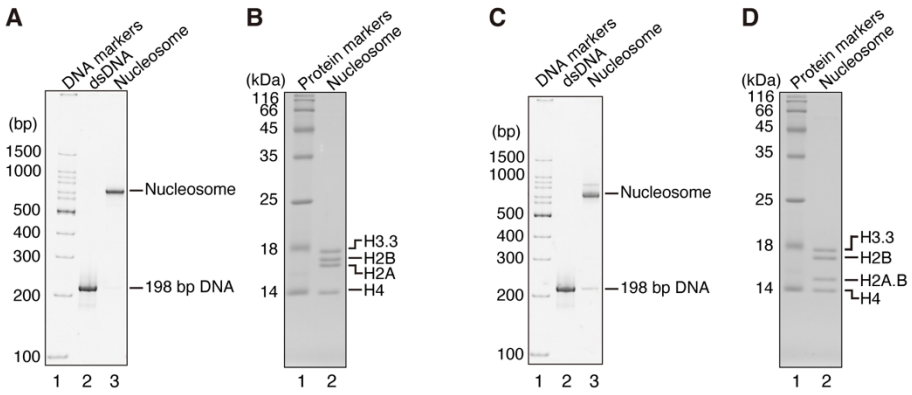
The Appendix View includes

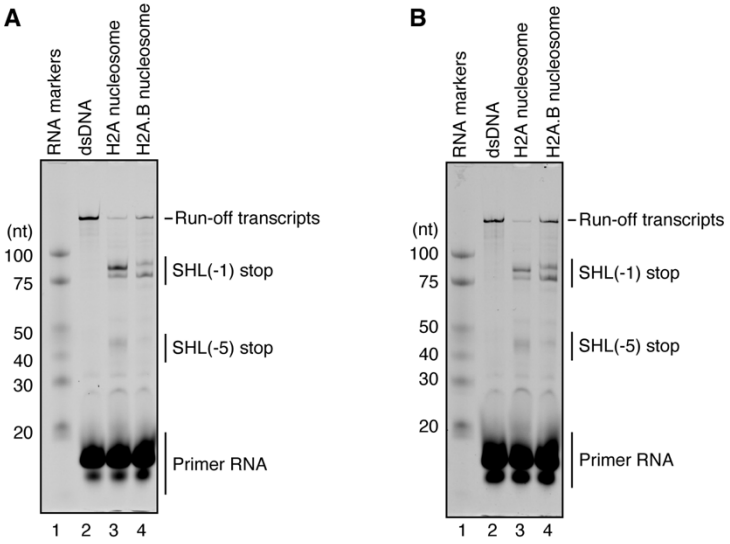
Appendix Fig. S1



Appendix Fig. S1- The H2A nucleosome and H2A.B nucleosome used in transcription assays and cryo-EM analyses.

(A, B) The H2A nucleosome was analyzed by non-denaturing 6% polyacrylamide gel electrophoresis with ethidium bromide staining (A) and SDS 20% polyacrylamide gel electrophoresis with CBB staining (B). (C, D) The H2A.B nucleosome was analyzed by non-denaturing 6% polyacrylamide gel electrophoresis with ethidium bromide staining (C) and SDS 20% polyacrylamide gel electrophoresis with CBB staining (D).

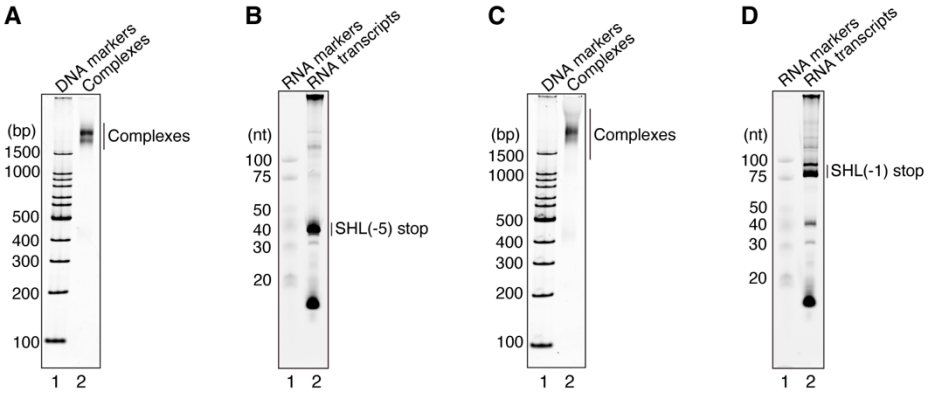
Appendix Fig. S2



Appendix Fig. S2- Repeated H2A nucleosome and H2A.B nucleosome transcription assays.

(A, B) Replicated experiments of the transcription assay shown in Figure 1C.

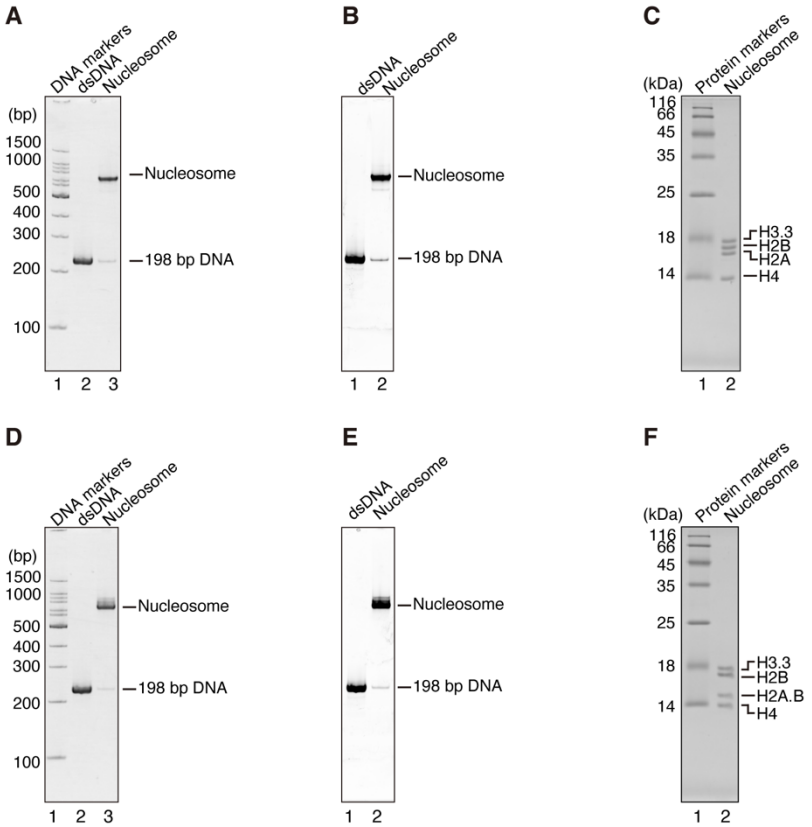
Appendix Fig. S3



Appendix Fig. S3- The EC-H2A.B nucleosome complexes used in cryo-EM analyses.

(A, B) The EC-H2A.B nucleosome complex paused at the SHL(-5) position for the cryo-EM analysis was analyzed by non-denaturing 4% polyacrylamide gel electrophoresis with SYBR Gold staining (A). RNA transcripts were separated by denaturing 10% polyacrylamide gel electrophoresis and detected with DY647 fluorescent dye (B). (C, D) The EC-H2A.B nucleosome complex paused at the SHL(-1) position for the cryo-EM analysis was analyzed by non-denaturing 4% polyacrylamide gel electrophoresis with SYBR Gold staining (C). RNA transcripts were separated by denaturing 10% polyacrylamide gel electrophoresis and detected with DY647 fluorescent dye (D).

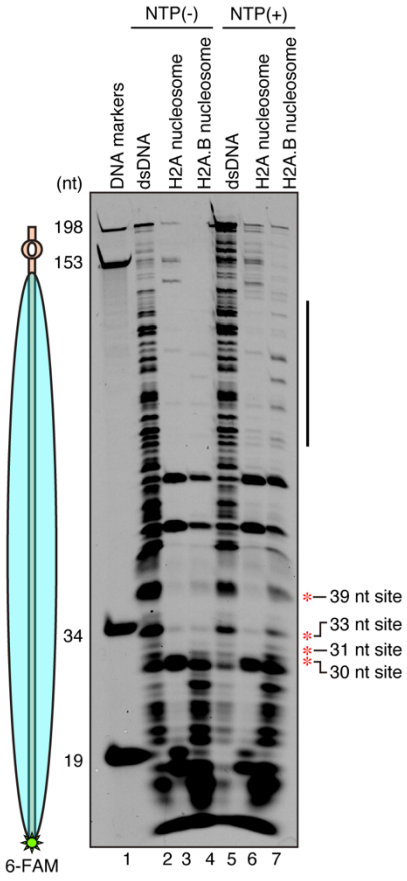
Appendix Fig. S4



Appendix Fig. S4- The canonical H2A and H2A.B nucleosomes used in DNase I footprinting analysis.

(A-C) The H2A nucleosome was analyzed using non-denaturing 6% polyacrylamide gel electrophoresis with ethidium bromide staining (A) and 6-FAM fluorescent dye detection (B). The histone composition was confirmed by SDS 20% polyacrylamide gel electrophoresis with CBB staining (C). (D-F) The H2A.B nucleosome was analyzed using non-denaturing 6% polyacrylamide gel electrophoresis with ethidium bromide staining (D) and 6-FAM fluorescent dye detection (E). The histone composition was confirmed by SDS 20% polyacrylamide gel electrophoresis with CBB staining (F).

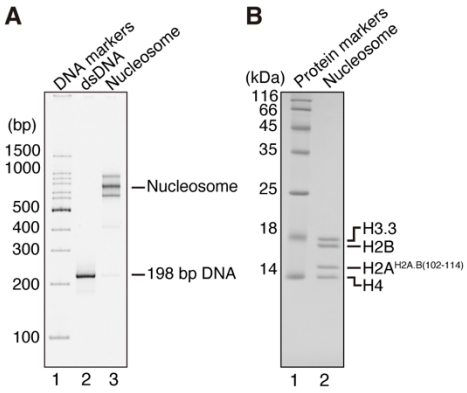
Appendix Fig. S5



Appendix Fig. S5- Repeated DNase I footprinting analysis.

A replicated experiment of the DNase I footprinting shown in Figure 3B.

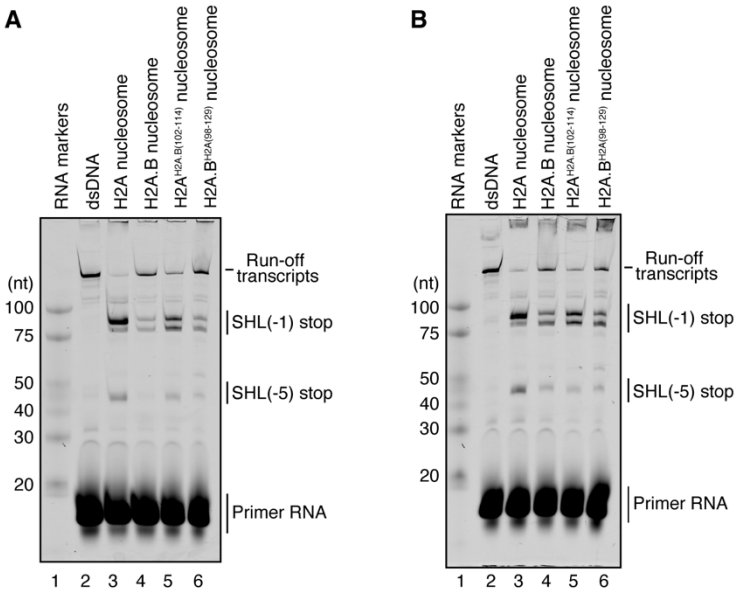
Appendix Fig. S6



Appendix Fig. S6- The H2A$^{H2A.B(102-114)}$ nucleosome used in transcription assays.

(A, B) The H2A$^{H2A.B(102-114)}$ nucleosome was analyzed by non-denaturing 6% polyacrylamide gel electrophoresis with ethidium bromide staining (A) and SDS 20% polyacrylamide gel electrophoresis with CBB staining (B).

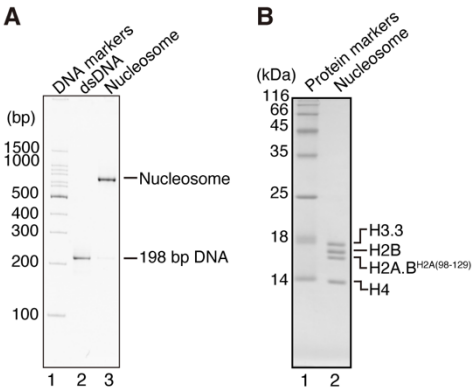
Appendix Fig. S7



Appendix Fig. S7- Repeated $H2A^{H2A.B(102-114)}$ nucleosome and $H2A.B^{H2A(98-129)}$ nucleosome transcription assays.

(A, B) Replicated experiments of the transcription assay shown in Figure 4C.

Appendix Fig. S8



Appendix Fig. S8- The H2A.B$^{H2A.B(98-129)}$ nucleosome used in transcription assays.

(G, H) The H2A.B$^{H2A(98-129)}$ nucleosome was analyzed by non-denaturing 6% polyacrylamide gel electrophoresis with ethidium bromide staining (G) and SDS 20% polyacrylamide gel electrophoresis with CBB staining (H).