Supplementary Figure 1. YY1 relocalizes to the decondensing DNA in telophase prior to nuclear envelope reconstitution. Indirect immunofluorescence of HeLa cells during early, mid, and late telophase was performed. Endogenous YY1 (Red) was immunostained with anti-YY1 antibody (H-414). The nuclear envelope was visualized by immunostaining of Lamin A/C (Green), followed by DAPI staining of DNA (Blue). Bar, 5μm.

Supplementary Figure 2. YY1 distributes to the cytoplasm away from condensed DNA in nocodazole arrested cells. HeLa cells were grown on coverslips, blocked at pro-metaphase by nocodazole for 18 hours, fixed and stained. Endogenous YY1 (Red) was visualized by immunostaining with anti-YY1 antibody (H-414), followed by DAPI staining of DNA (Blue). Bar, 5μm.

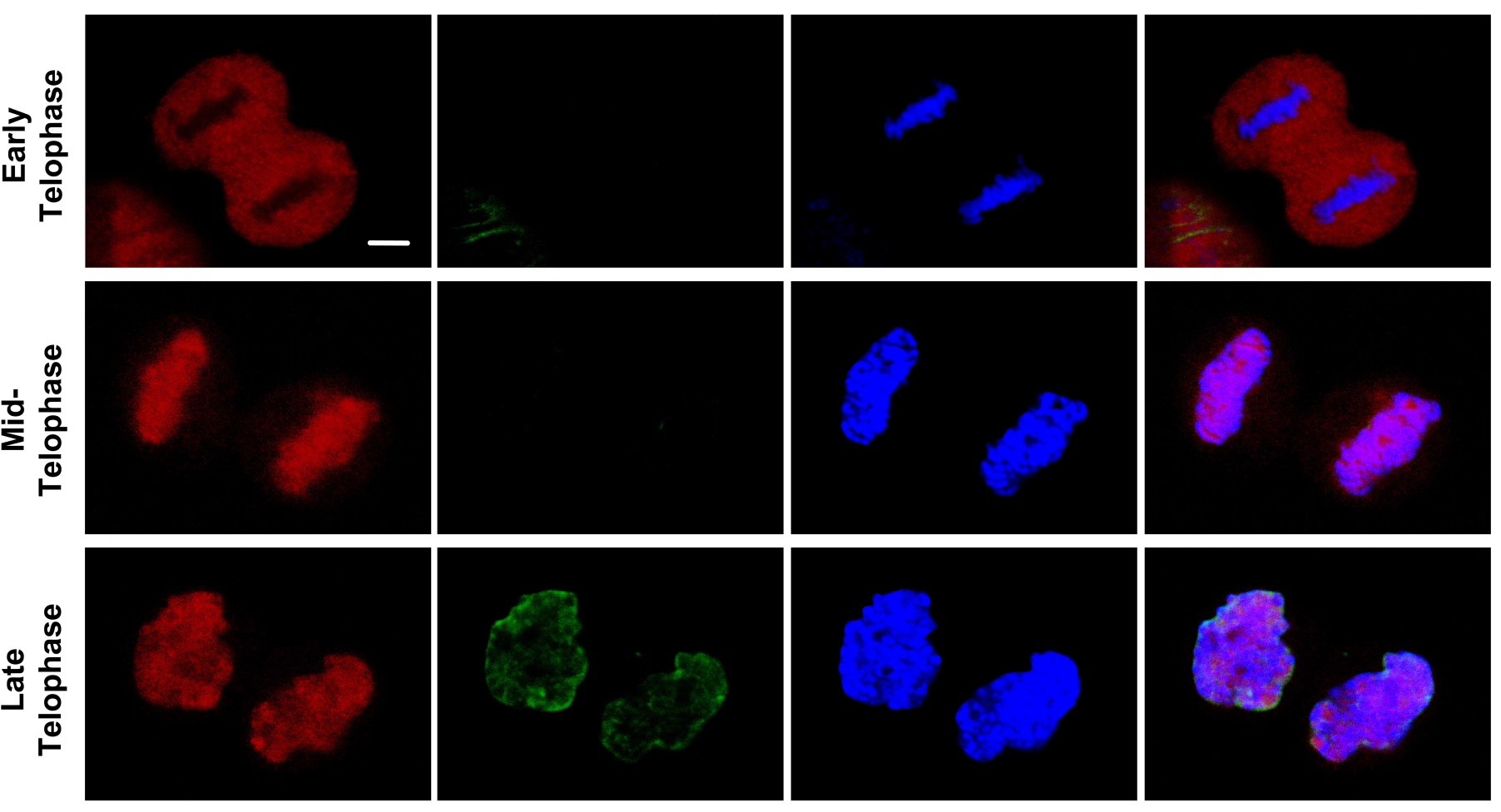
Supplementary Figure 3. Dephosphorylation of Flag-YY1 from nocodazole-arrested HeLa cells increases its DNA binding activity. Flag-YY1, purified from nocodazole-arrested HeLa cells, stably transfected with pCS2(+)-Flag-YY1, was incubated at 30ºC, with or without lambda phosphatase for 30 minutes prior to use in the electrophoretic mobility shift assay (EMSA) with p5-60 or Cdc6 probes, as described in the Methods section. The specificity of the YY1 shift was assessed by addition of anti-YY1 (C-20), anti-GFP, or anti-Sp1 to the reaction of Flag-YY1 with lambda phosphatase; as indicated.

Supplementary Figure 4. Mutation of S247 has no effect on YY1 nuclear localization. HeLa cells were grown on coverslips were transfected with pCS2(+)-Flag-YY1 WT, S247D, or S247A. 18 hours post-transfection, cells were fixed and immunostained with

anti-Flag antibody (Red) to visualize the overexpressed proteins and DAPI to visualize the DNA/nucleus (Blue). Bar, 10μm.

Supplementary Figure 5. Mutation of threonine 348 and 378 residues to alanine abolishes the DNA binding activity of YY1. Whole cell extracts prepared from HeLa cells transfected with pCS2(+)-Flag-YY1 (WT) or (T348,378A) mutant were tested for equal expression of the exogenous proteins on a Western blot with anti-YY1 (H-10) (A). The same extracts were incubated with radioactively labeled H3.2α as probe in an EMSA reaction (B). Endogenous and Flag-YY1 shifts are as indicated.

Supplementary Figure 6. Mutations of T348 and T378 have no effect on YY1 nuclear localization. HeLa cells grown on coverslips were transfected with pCS2(+)-Flag-YY1 wild type (WT), (T348,378D), or (T348,378A) mutants . 18 hours post-transfection, cells were fixed and immunostained with anti-Flag antibody (Red) to visualize the overexpressed proteins, and DAPI to visualize the DNA/nucleus (Blue). Bar, 10μm.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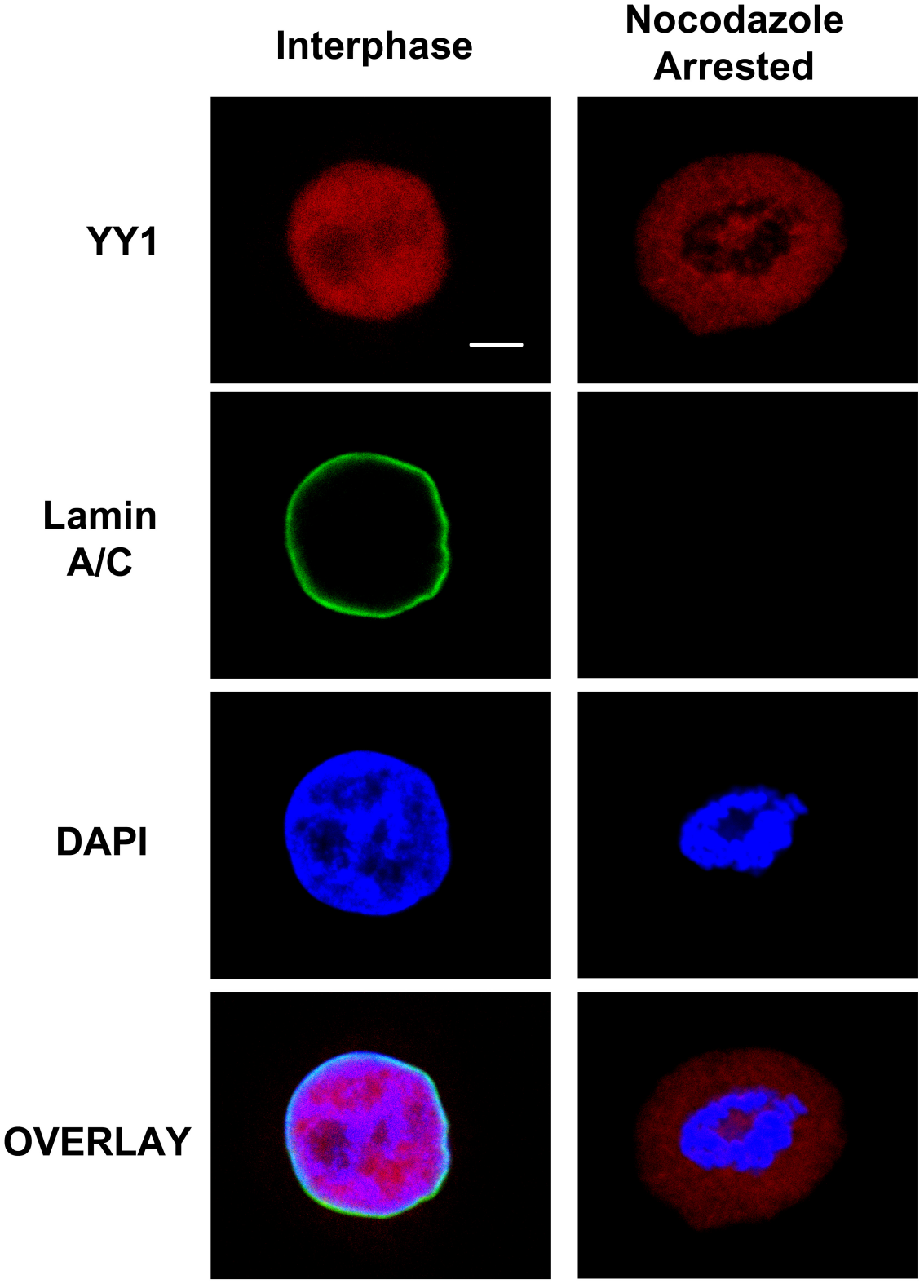
Mid-Telophase
Late Telophase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YY1
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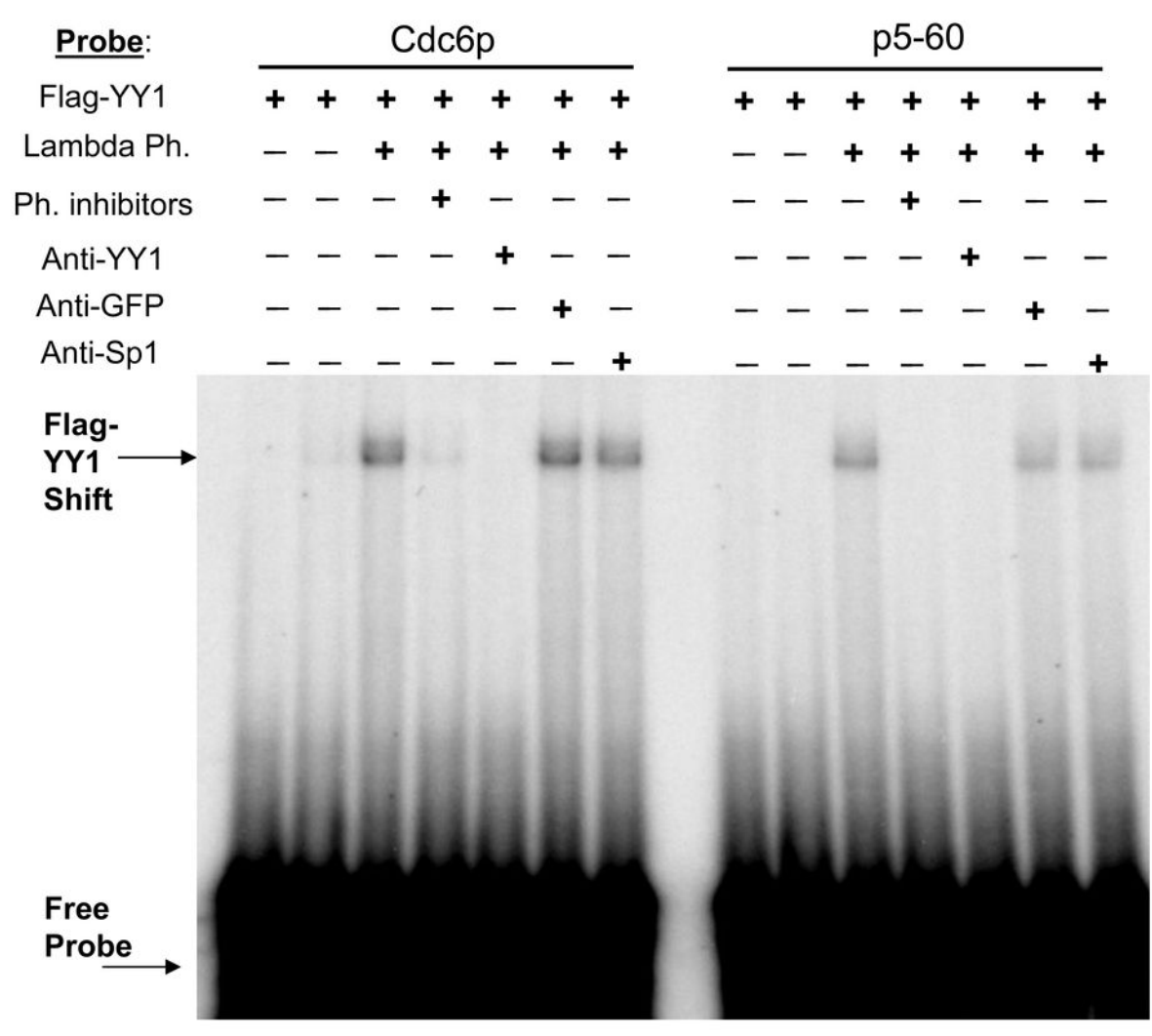
DAPI
OVERLAY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Lambda Ph.
Ph. inhibitors
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Flag-YY1 Shift
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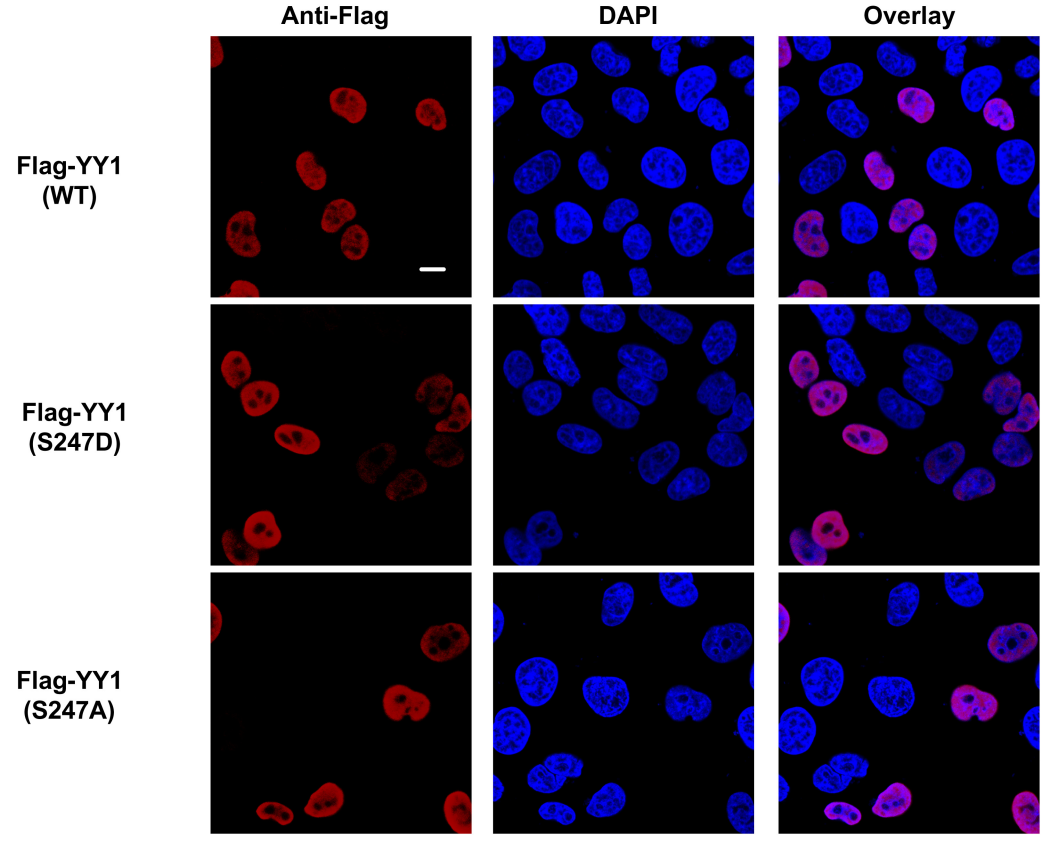
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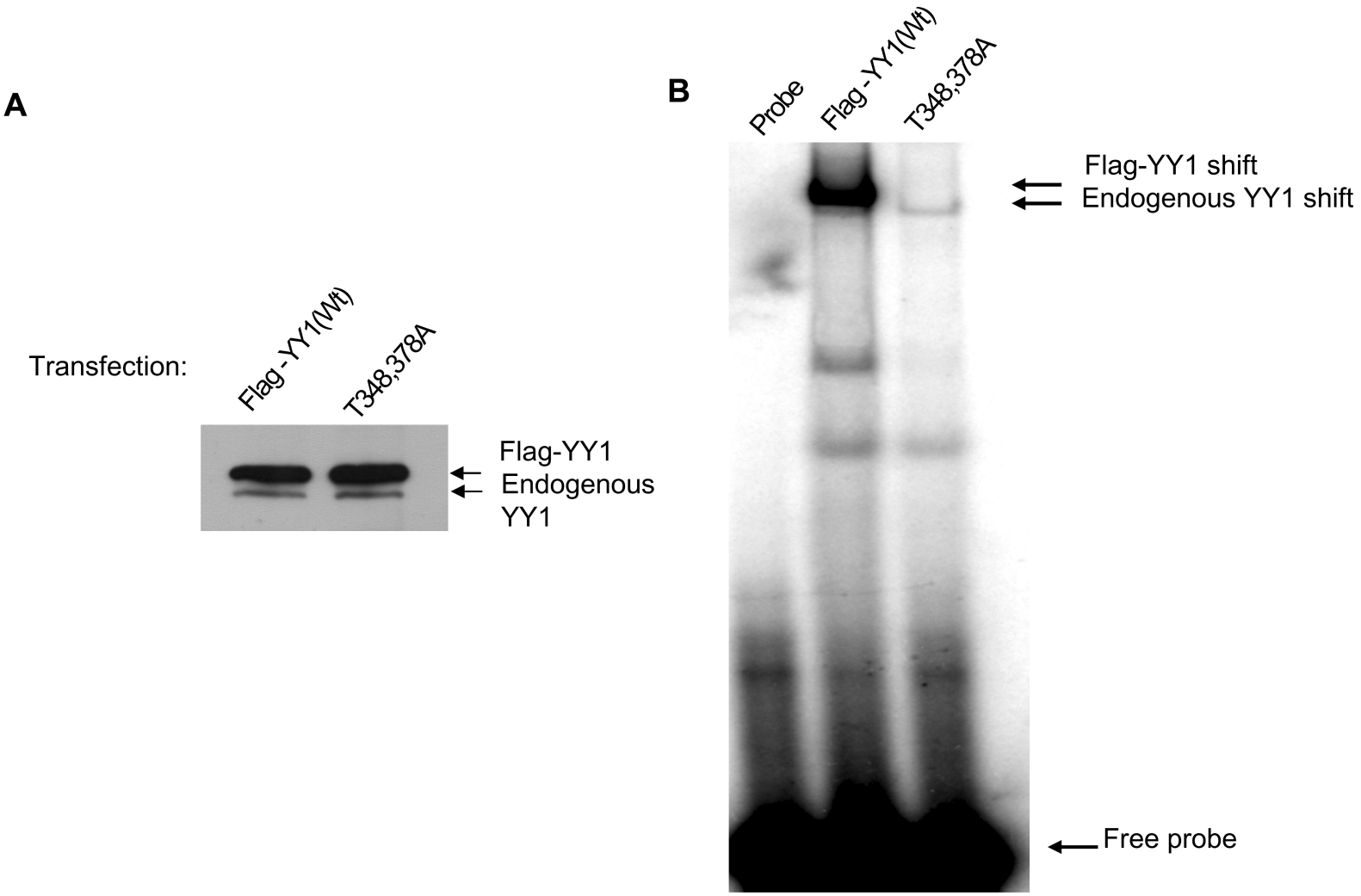
DAPI
Overlay
Flag-YY1
(WT)
Flag-YY1
(S247D)
Flag-YY1
(S247A)


A
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T348,378A
Flag-YY1
Endogenous
YY1
B
Probe
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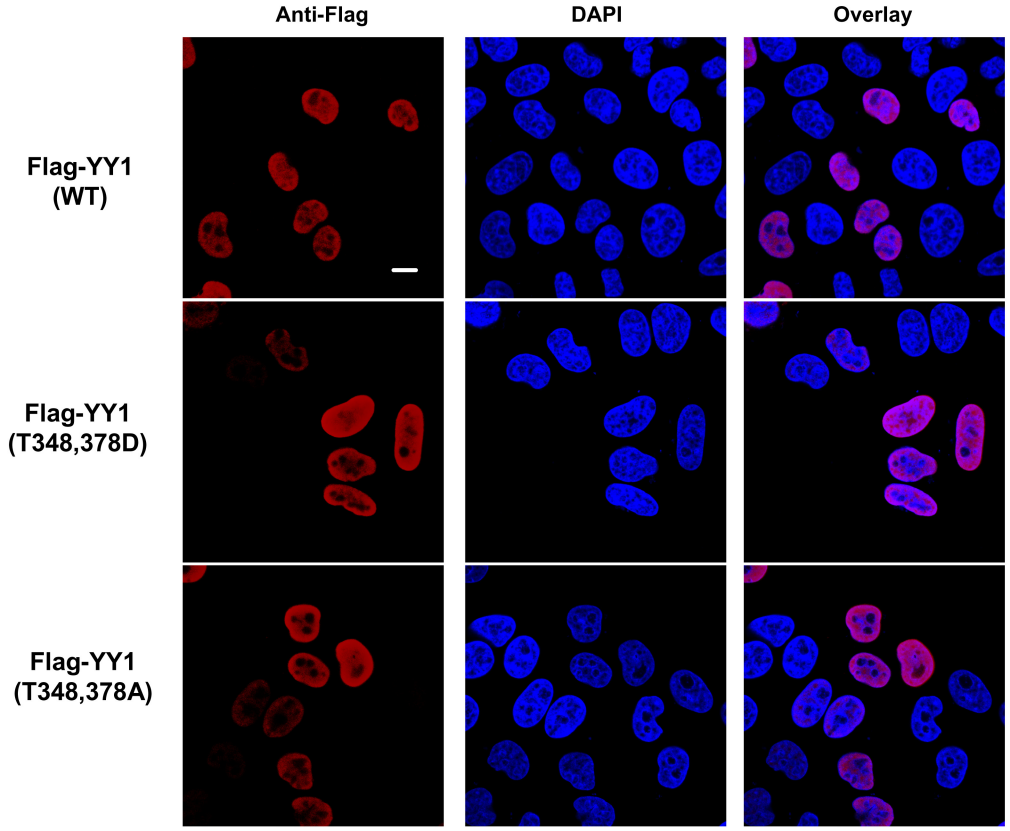
Endogenous YY1 shift
Free probe


Anti-Flag
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Flag-YY1
(WT)
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Flag-YY1
(T348,378A)