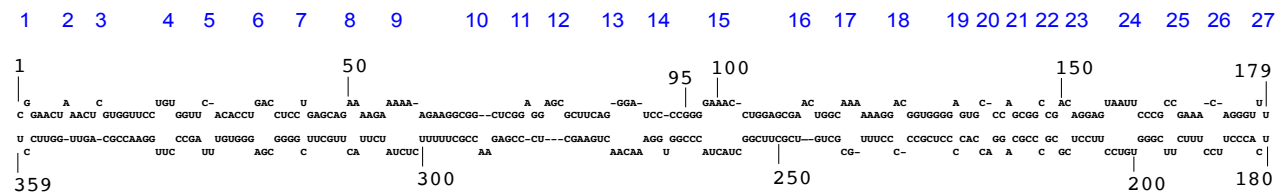
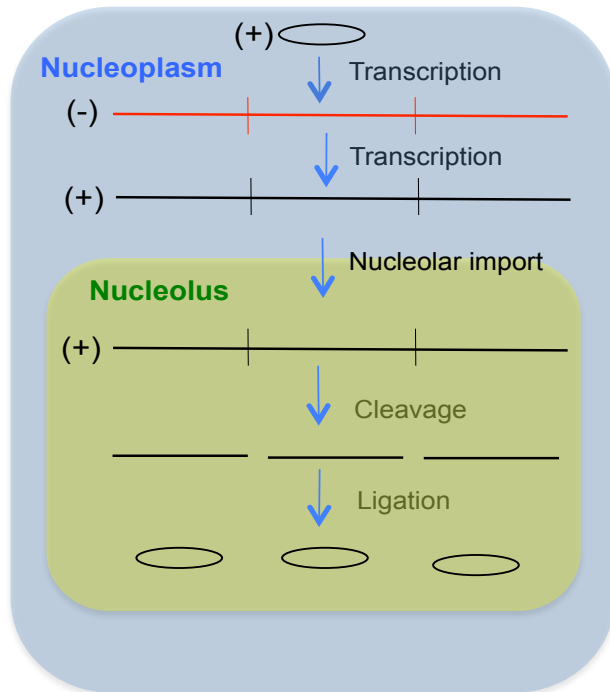


Supplemental Figure 1. Schematic representation of PSTVd secondary structure (A) and asymmetric rolling circle replication in the nucleus (B). In (A), the loops are numbered from left to right.

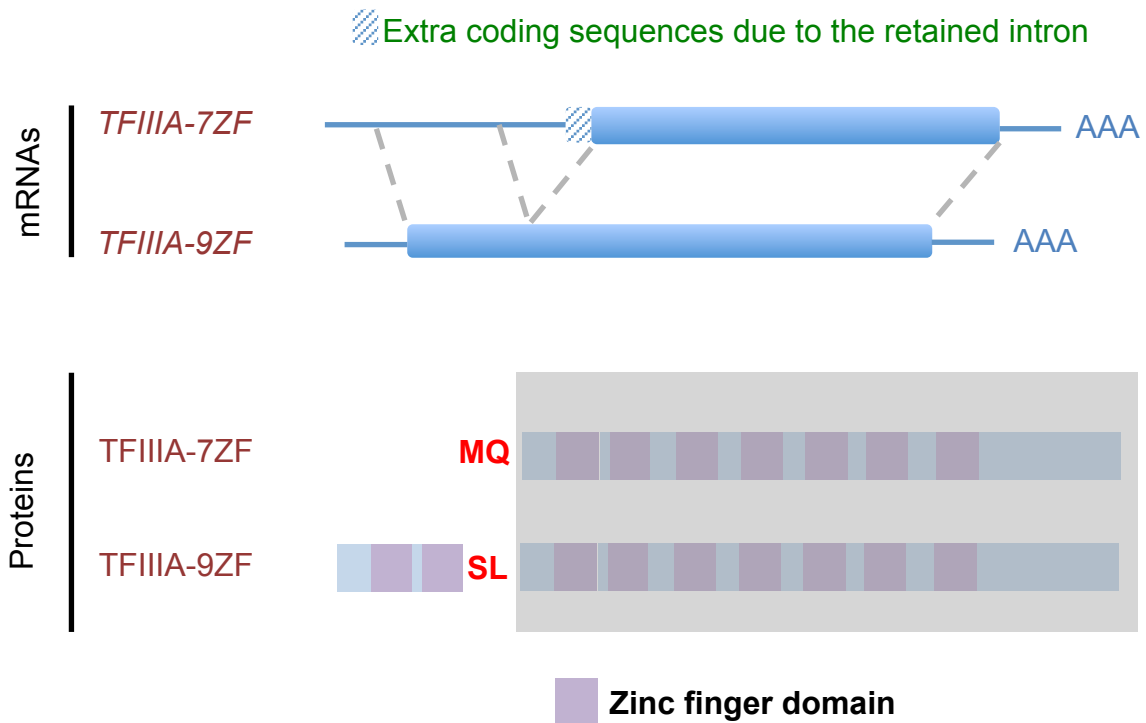
(A)



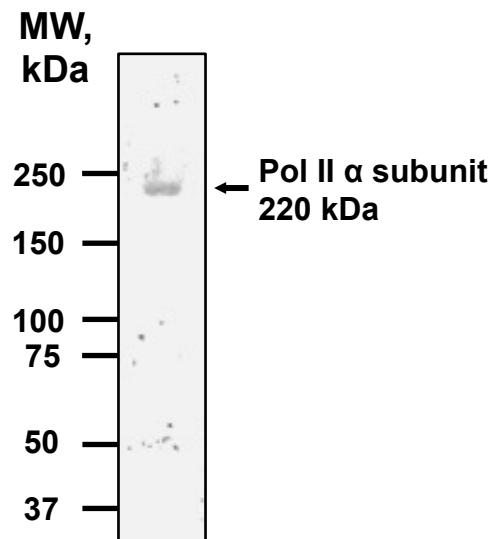
(B)



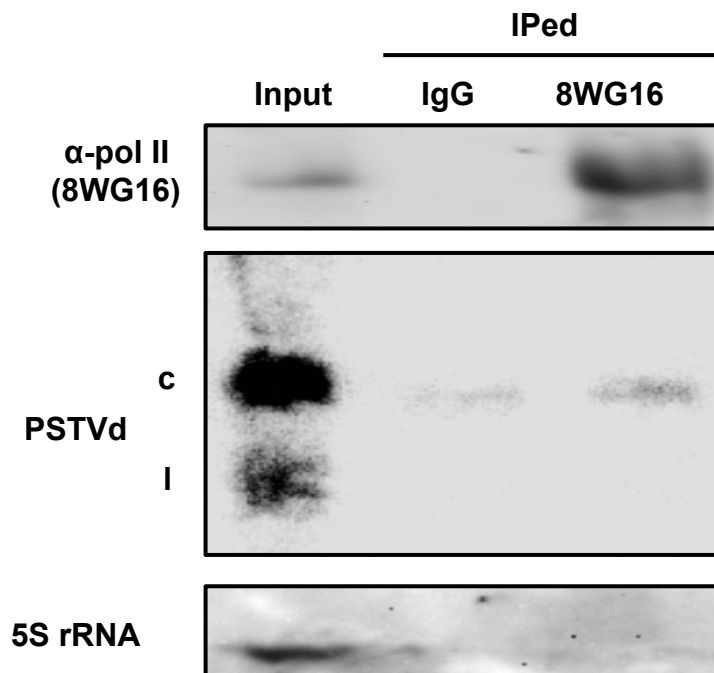
Supplemental Figure 2. Schematic representation of TFIIIA mRNA splicing variants and their translational products.



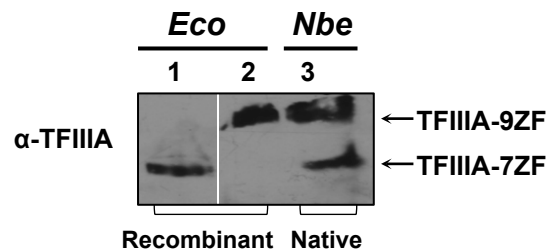
Supplemental Figure 3. Demonstration of the specificity of Pol II monoclonal antibody (8WG16) against the largest subunit of Pol II in *N. benthamiana*.



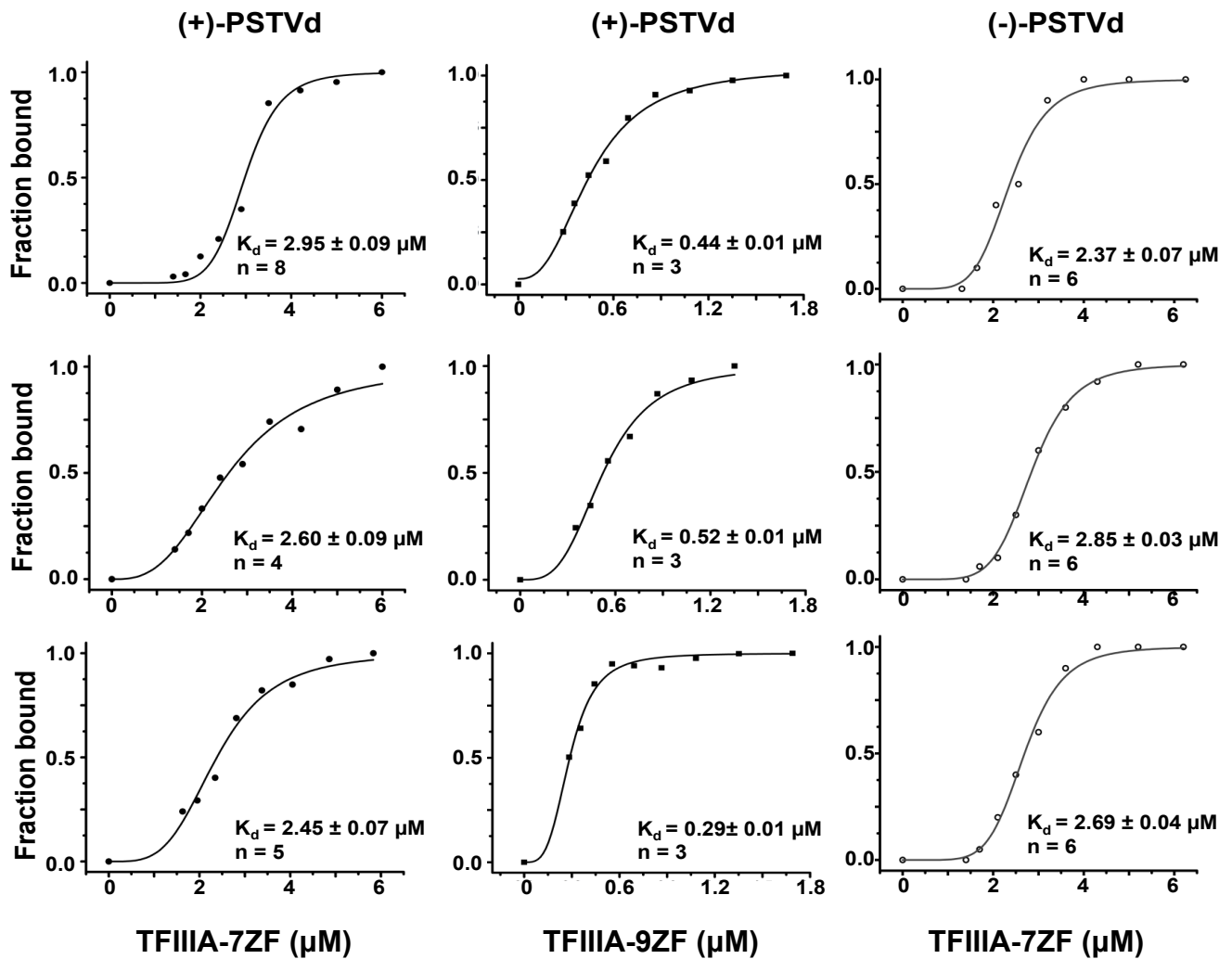
Supplemental Figure 4. *In vivo* interactions between Pol II and circular (+)-PSTVd revealed by RNA immunoprecipitation assays. The cell lysates were incubated with 8WG16 antibodies (α -Pol II) to pull down Pol II. Mouse IgG was used as a negative control. The immunoprecipitated fractions were subject to PSTVd detection by RNA gel blotting analysis. 5S rRNA was used as a negative control. The largest subunit and circular (+)-PSTVd RNAs were detected in the α -Pol II precipitated fraction, but not in IgG precipitated fraction. 5S rRNA was not detected in either fraction.



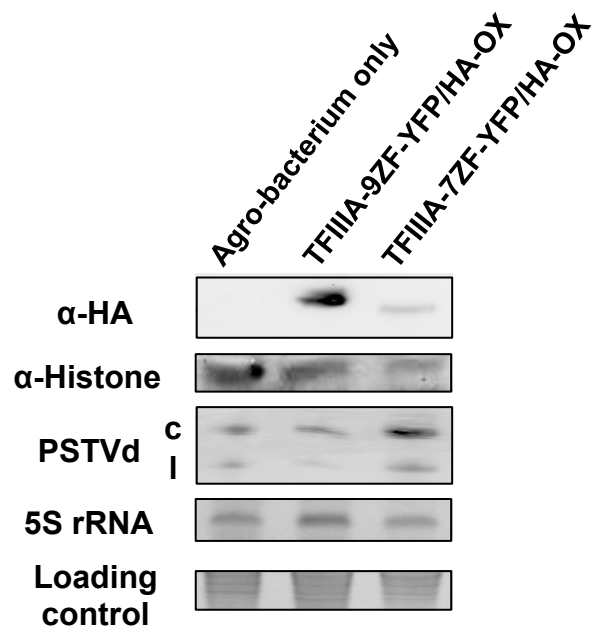
Supplemental Figure 5. Expression of TFIIIA-9ZF and TFIIIA-7ZF in *N. benthamiana*. Total lysates from *N. benthamiana* seedlings were used for detecting the two forms of native TFIIIA using α -TFIIIA polyclonal serum. Recombinant TFIIIA-9ZF and TFIIIA-7ZF proteins produced in *E. coli* serve as size markers (lanes 1 and 2).



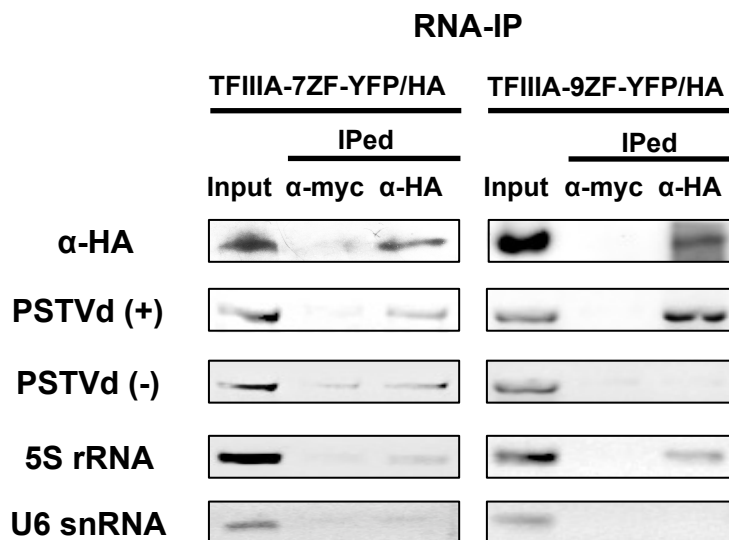
Supplemental Figure 6. Binding curves for the *in vitro* interaction between TFIIIA proteins and PSTVd, as determined from EMSA results.



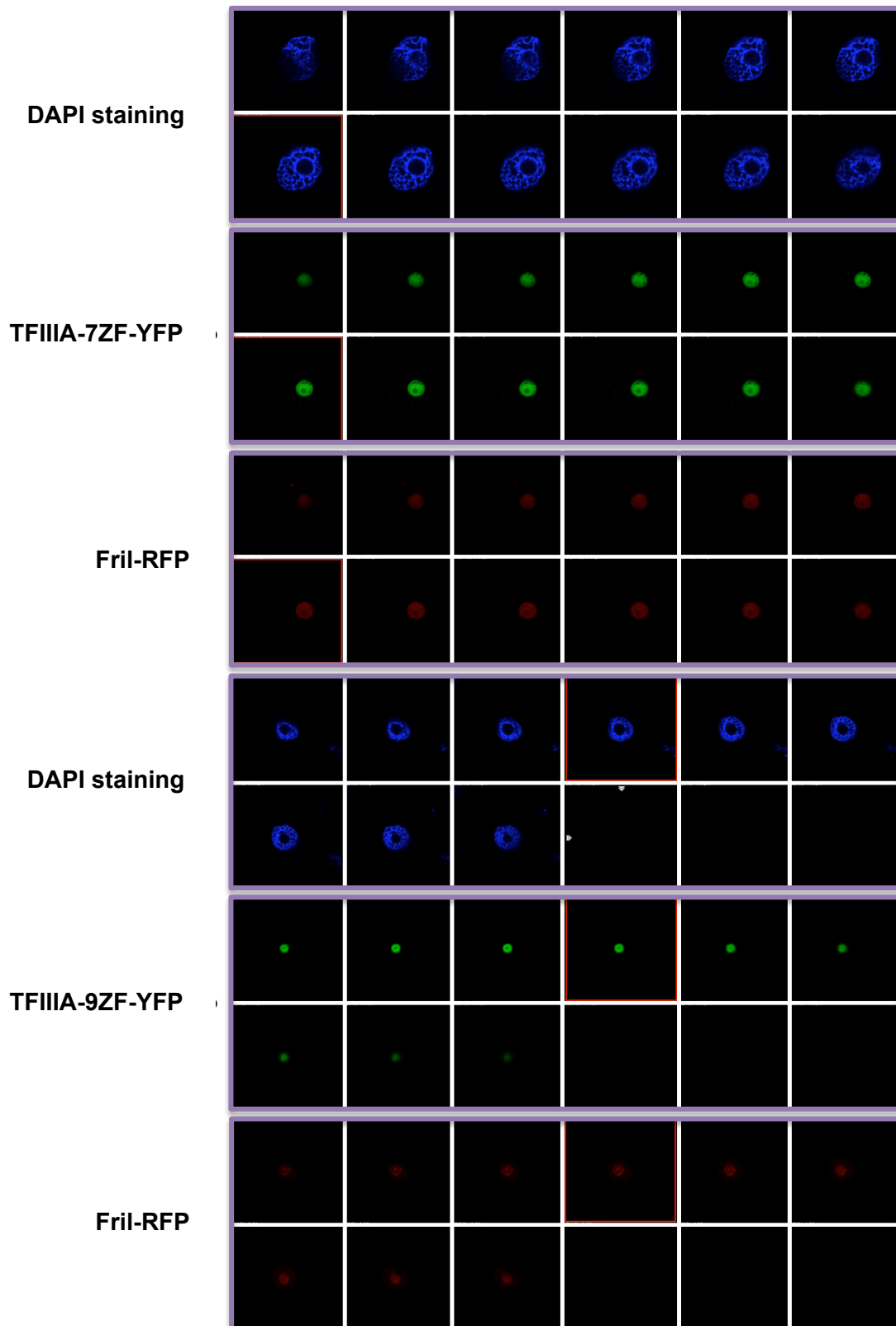
Supplemental Figure 7. Specific enhancement of PSTVd replication with ectopic expression of TFIIIA-7ZF. YFP/HA-tagged TFIIIA-9ZF and TFIIIA-7ZF were detected by α -HA, and endogenous histone H3 was detected by α -histone H3 in immunoblotting analyses. RNA gel blotting analyses were employed to detect PSTVd and 5S rRNA. c, circular PSTVd, l, linear PSTVd. Ethidium bromide staining of ribosomal RNAs served as the loading control.



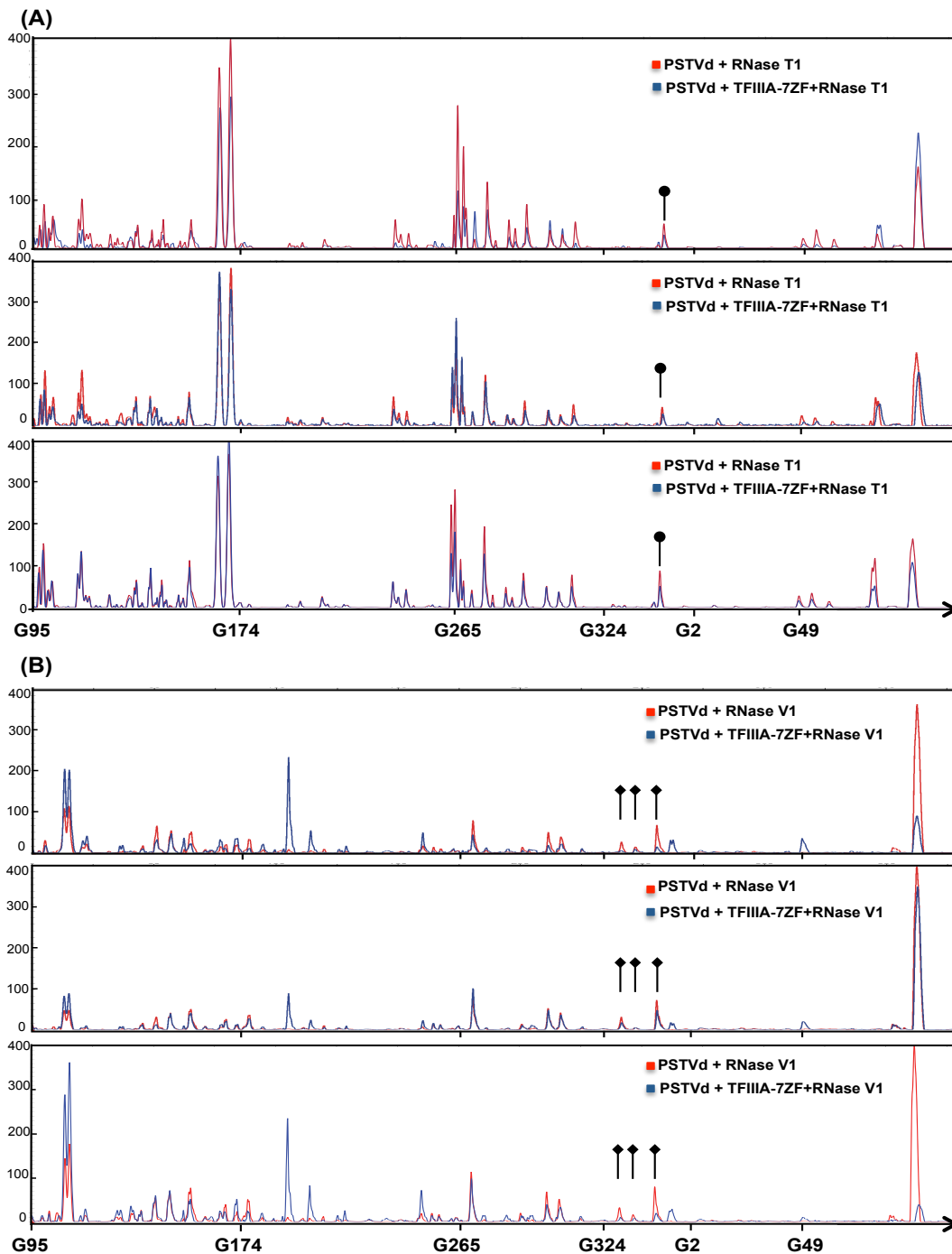
Supplemental Figure 8. *In vivo* interactions between TFIIIA and PSTVd revealed by RNA immunoprecipitation assays. The cell lysates were incubated with α -HA antibodies to detect TFIIIA-7ZF-YFP/HA and -9ZF-YFP/HA proteins or with α -myc antibodies, with the latter serving as a negative control. The immunoprecipitated fractions were subject to PSTVd detection by RT-PCR. 5S rRNA was used as a positive control. Both the (+)- and (-)-PSTVd RNAs were detected in the precipitated TFIIIA-7ZF fraction, but only the (+)-PSTVd was detected in the precipitated TFIIIA-9ZF fraction. U6 snRNA, a negative control, was not detected in any fraction.



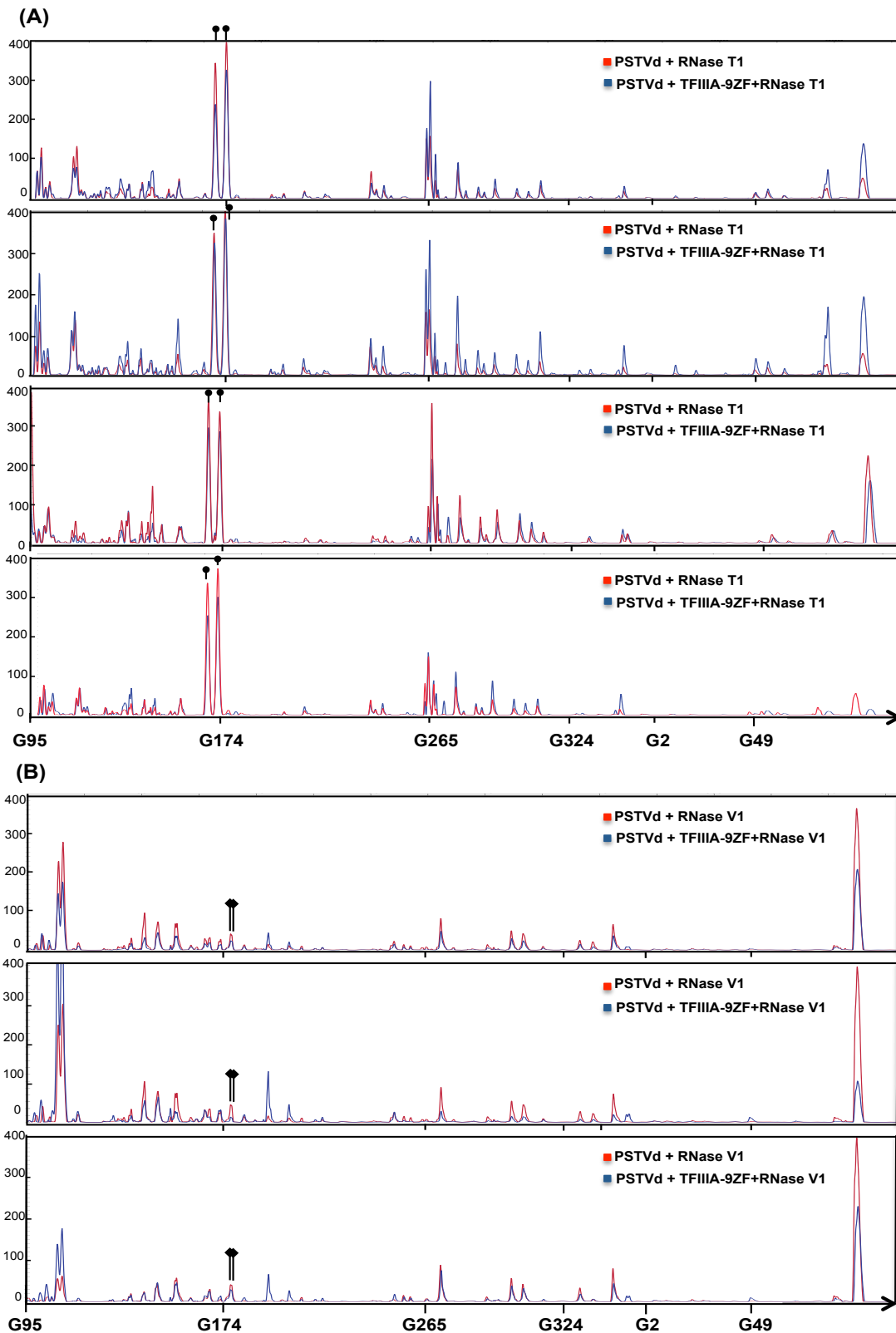
Supplemental Figure 9. Z-series confocal images from whole mounts of PSTVd-infected *N. benthamiana* leaves that ectopically express either TFIIIA-7ZF-YFP/HA or TFIIIA-9ZF-YFP/HA. Arabidopsis Fril-RFP was co-expressed to serve as nucleolar marker. DAPI was used to stain nuclei. Images were collected at 0.3 μ m intervals. The images in red-colored boxes are presented with contrast enhancement in Figure 5.



Supplemental Figure 10. Capillary electrophoretograms from three independent experiments for RNase T1 (A) and V1 (B) footprinting of the PSTVd-TFIIIA-7ZF complex. PSTVd fragments digested by RNase in the absence of TFIIIA-7ZF (red) or in the presence of TFIIIA-7ZF (blue) are presented by peaks in electrophoretogram. Y-axes indicate arbitrary intensities of peaks. Short black lines at the bottom of panels A and B indicate elution position of fluor-labeled PSTVd size markers whose compositions are identical to that in full-length PSTVd. Black arrows indicate the positions where protection by TFIIIA-7ZF is inferred from RNase T1 (round-end symbols) and V1 (square-end symbols) footprinting experiments.



Supplemental Figure 11. Capillary electrophoretograms from three independent experiments for RNase T1 (A) and V1 (B) footprinting of the PSTVd-TFIIIA-9ZF complex. The color schemes, symbols, and Y-axes are identical to those employed in Supplemental Figure 10.



Supplemental Table 1. Primers, vectors, and bacterial strains used for cloning.

Constructs for expression in plants

TFIIIA-antisense:

f: 5'-caccagcatctggccaaaga-3'

r: 5'-ggtgccaccagcctgtgtgtg-3'

Gateway Entry Vector: pCR8/GW/TOPO

(Life Technologies)

Gateway Destination Vector: pMDC32

(TAIR, #CD3-738)

Cloned from Total RNA by RT-PCR

HA-TFIIIA-7ZF:

f: 5'-ggtggtcatatgcaagagaggccatttgcattgc-3'

r: 5'-tcacagcttctcatctgattctg-3'

Gateway Entry Vector: pCR8/GW/TOPO

(Life Technologies)

Gateway Destination Vector: pEarleyGATE201

(TAIR, #CD3-687)

Cloned from Total RNA by RT-PCR

HA-TFIIIA-9ZF:

f: 5'-caccaaatgggagaagagatgagagaag-3'

r: 5'-tcacagcttctcatctgattctg-3'

Gateway Entry Vector: pCR8/GW/TOPO

(Life Technologies)

Gateway Destination Vector: pEarleyGATE201

(TAIR, #CD3-687)

Cloned from Total RNA by RT-PCR

TFIIIA-7ZF-YFP/HA:

f: 5'-ggtggtcatatgcaagagaggccatttgcattgc-3'

r: 5'-cagcttctcatctgattctg-3'

Gateway Entry Vector: pCR8/GW/TOPO

(Life Technologies)

Gateway Destination Vector: pEarleyGATE101

(TAIR, #CD3-683)

Cloned from Total RNA by RT-PCR

TFIIIA-9ZF-YFP/HA:

f: 5'-caccaaatgggagaagagatgagagaag-3'

r: 5'-cagcttctcatctgattctg-3'

Gateway Entry Vector: pCR8/GW/TOPO

(Life Technologies)

Gateway Destination Vector: pEarleyGATE101

(TAIR, #CD3-683)

Cloned from Total RNA by RT-PCR

AtFri I-RFP:

f: 5'-atgagacccccagttacaggag-3'

r: 5'-tgaggctggggtctttgttcttcg-3'

Gateway Entry Vector: pCR8/GW/TOPO

(Life Technologies)

Gateway Destination Vector: pK7WFR2.0

(Gift from Dr. Iris Meier, OSU)

Cloned from Total RNA by RT-PCR

HA-GFP:

f: 5'-atggtgagcaagggcgaggagc-3'

r: 5'-ttactgttacagctcgtccatgccg-3'

Gateway Entry Vector: pCR8/GW/TOPO

(Life Technologies)

Gateway Destination Vector: pEarleyGATE201

(TAIR, #CD3-687)

Cloned from pK7FWG2 template by PCR

Constructs for expression in *E. coli*

pTXB1-TFIIIA-7ZF:

f: 5'- ggtggcatatgcaagagaggccatttgcacgc-3'

r: 5'-aaagctactagtgcgtcccagtgatgcacagctcttcac-3'

Expression vector: pTXB1 (NEB)

E. coli BL21(DE3) Rosetta strain was used for expression

pTXB1-TFIIIA-9ZF:

f: 5'-ggtggcatatgatgggagaagagatgagagaag-3'

r: 5'-aaagctactagtgcgtcccagtgatgcacagctcttcac-3'

Expression vector: pTXB1 (NEB)

E. coli BL21(DE3) Rosetta strain was used for expression

His6-TFIIIA-7ZF:

f: 5'-ggtggcatatgcaagagaggccatttgcacgc-3'

r: 5'-tcacagctcttcacgattctg-3'

Expression vector: pDEST17 (Life Technologies)

E. coli BL21(DE3) Rosetta strain was used for expression

Constructs for in vitro transcripts/probes

5S rRNA probe:

f: 5'-ggatgcatcaccagc-3'

r: 5'-gagggatgcaacacgagg-3'

Vector: pGEM-T (Promega, Madison, WI)

pInt95-94+:

f: 5'-ggggacaagttgtacaaaaagcagaattaaccctcactaaaggggaaacctggagcga-3'

r: 5'-ggggaccactttgtacaagaaagctgggtcccggggatccctgaagcgctcc-3'

Vector: pDONR221 (Life Technologies)

pInt95-94-:

f: 5'-ggggacaagttgtacaaaaagcagaattaaccctcactaaaggggatccctgaagcg-3'

r: 5'-ggggaccactttgtacaagaaagctgggtcccgggaaacctggagcgaact-3'

Vector: pDONR221 (Life Technologies)

pInt95-174+:

f: 5'-ggggacaagttgtacaaaaagcagaattaaccctcactaaaggggaaacctggagcga-3'

r: 5'-ctgttcggcggaattactcc-3'

Vector: pCR8/GW/TOPO (Life Technologies)

pInt95-265+:

f: 5'-ggggacaagttgtacaaaaaagcagaattaaccctcactaaaggggaaacctggagcga-3'

r: 5'-cgggtagtagccgaagcgac-3'

Vector: pCR8/GW/TOPO (Life Technologies)

pInt95-324+:

f: 5'-ggggacaagttgtacaaaaaagcagaattaaccctcactaaaggggaaacctggagcga-3'

r: 5'-cgccccgaagcaagtaagatag-3'

Vector: pCR8/GW/TOPO (Life Technologies)

pInt95-2+:

f: 5'-ggggacaagttgtacaaaaaagcagaattaaccctcactaaaggggaaacctggagcga-3'

r: 5'-cgaggaaccaactgcggtcca-3'

Vector: pCR8/GW/TOPO (Life Technologies)

pInt95-49+:

f: 5'-ggggacaagttgtacaaaaaagcagaattaaccctcactaaaggggaaacctggagcga-3'

r: 5'-ctgctcaggaggtcaggtgtga-3'

Vector: pCR8/GW/TOPO (Life Technologies)

CaMV35S promoter construct:

35Sf: 5'-gcccaaccggtcaacatgtggag-3'

35Sr: 5'-acggtcctaaggtagcgcgatggc-3'

Gateway Entry Plasmid: pENTR-MIR4376 (Wang et al., 2011, Plant Cell; PMID: 21917547)

Gateway Destination Vector: CD3-1656 (TAIR)

(The strategy for this construct is to recombine pENTR-MIR4376 with CD3-1656, then use the above primer pairs to PCR clone the fragment for template)

Primers for RNA immunoprecipitation experiments

PSTVd:

f: 5'-ggggaaacctggagcgaactgg-3'

r: 5'-cccgggatccctgaagcgctcc-3'

5S rRNA:

f: 5'-ggatgcatcaccagc-3'

r: 5'-gagggatgcaacacgagg-3'

U6 snRNA:

f: 5'-gtcccttcggggacatccgata-3'

r: 5'-tttgaccatttctcgatttgctg-3'

NOTE:

- 1) Agrobacterium strain GV3101 was used for agroinfiltration in all instances
- 2) All the recombination reactions were carried out using LR clonase II (Life Technologies)

Supplemental Data 1. *N. benthamiana* TFIIIA-9ZF and TFIIIA-7ZF cDNA sequences.

TFIIIA-9ZF

ATGGGAGAAGAGATGAGAGAAGTAATATTCAGAGACATAAGACGATATTACTG
TGAATTTTGTGGGATTTGCCGCTCCAAAAAGTCCCTTATCTCCTCTCATATCCT
CTCCTATCATCAAGATGAAATGGAGAAGCGGAAAGATGAGGCAAACGAAGATG
CAAAGATAAACGAAGGCCCAAACCTGAATATTTGTGAAGAATGTGGTGTGAGC
TTTCAGAAGCCTGCTCATTGAAGCAGCATATGCAGAGTCATTCACTCGAGAG
GCCATTTGCATGCCATATAGATGACTGCCAGTCCAGCTACCGTAGAAAGGATC
ACTTGACGAGACATCTCTTGCAGCACCAAGGGAAGTTGTTTGAATGTCCTTTTG
ATGGGTGCAAACGCGCATTAGTATCCAAGGCAACATGACTCGGCATGTCAAT
GAGATGCATGACCAGCATGCCTCCCCTGAGGCGAGTCTTCCAAAGCAGTATGT
CTGTGCAGAACCTGGTTGTGGGAAGGTGTTTAAATCTGCATCCAAATTAAGA
AACATGAGGATTCTCACGTTAAGTTGGAGACAATGGAAGCACTTTGTTTAGAG
CCGGGCTGTATGAAACATTTACAAATGAGAAATGCCTCAAGGAACACGTGGA
GAGTTGCCACCAGCCTGTTGTGTGTGAAATATGTGGAACCAAGCAATTGAAGA
AGAATATTAAGCGGCATCTCCGTACGCATGAAGGAGAGTCTACATTGGAGAGA
ATAAAATGTGAATTCAGGATTGTCAGCACACTTTCTCCACTAAATCAAATCTTA
TTCAGCACGTCAAAGCTGTGCACCTGGGGGATAAACCATTCTCATGTAGCGTT
GCTGTTTGTGGTATGAAATTTGCCTTCGAGCACGTGAGAGATAGACATGAAAA
GTCTGGCTGTCATGTTTATACTCCTGGTGATTTTCATAGAGGCTGACGAGCAATT
CCGCTCCAGACCAAGAGGTGGTAGGAAGAGAAAGCTTCCTGTTTTTCGAGGCA
ATTATGCGGAAAAAGATAACACCACCCAGTGGTACAGATCCTGTGTTCTATCAA
GGATCAGAATATCTCTCATGGTTACTCTCAGCAGAATCAGATGAAGAGCTGTG
AGTCTATCTCTTTGGCCAGATGCTGGGT

TFIIIA-7ZF

ATGCAAGAGAGGCCATTTGCATGCCATATAGATGACTGCCAGTCCAGCTACCG
TAGAAAGGATCACTTGACGAGACATCTCTTGCAGCACCAAGGGAAGTTGTTTG
AATGTCCTTTTATGATGGGTGCAAACGCGCATTAGTATCCAAGGCAACATGACT
CGGCATGTCAATGAGATGCATGACCAGCATGCCTCCCCTGAGGCGAGTCTTC
CAAAGCAGTATGTCTGTGCAGAACCTGGTTGTGGGAAGGTGTTTAAATCTGCA
TCCAAATTAAGAAACATGAGGATTCTCACGTTAAGTTGGAGACAATGGAAGCA
CTTTGTTTAGAGCCGGGCTGTATGAAACATTTACAAATGAGAAATGCCTCAAG
GAACACGTGGAGAGTTGCCACCAGCCTGTTGTGTGTGAAATATGTGGAACCAA
GCAATTGAAGAAGAATATTAAGCGGCATCTCCGTACGCATGAAGGAGAGTCTA
CATTGGAGAGAATAAAATGTGAATTCAGGATTGTCAGCACACTTTCTCCACTA
AATCAAATCTTATTCAGCACGTCAAAGCTGTGCACCTGGGGGATAAACCATTCT
CATGTAGCGTTGCTGTTTGTGGTATGAAATTTGCCTTCGAGCACGTGAGAGAT
AGACATGAAAAGTCTGGCTGTCATGTTTATACTCCTGGTGATTTTCATAGAGGCT
GACGAGCAATTCCGCTCCAGACCAAGAGGTGGTAGGAAGAGAAAGCTTCCTG
TTTTCGAGGCAATTATGCGGAAAAAGATAACACCACCCAGTGGTACAGATCCT
GTGTTCTATCAAGGATCAGAATATCTCTCATGGTTACTCTCAGCAGAATCAGAT
GAAGAGCTGTGA