

ADDITIONAL FILE 1

RNA virus interference via CRISPR/Cas13a system in plants

Rashid Aman^{1,3}, Zahir Ali^{1,3}, Haroon Butt^{1,3}, Ahmed Mahas¹, Fatimah Aljedaani¹, Muhammad Zuhaib Khan¹, Shouwei Ding², and Magdy Mahfouz^{1,*}

¹Laboratory for Genome Engineering, Division of Biological Sciences, 4700 King Abdullah University of Science and Technology, Thuwal 23955-6900, Saudi Arabia, and ²Center for Plant Cell Biology, Department of Microbiology and Plant Pathology, University of California, Riverside, CA 92521

*Corresponding author: Magdy M. Mahfouz (magdy.mahfouz@kaust.edu.sa)

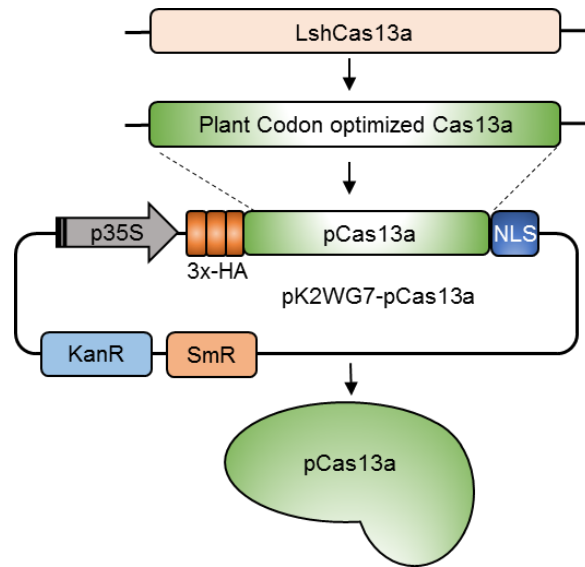
Additional file 1

Figure S1: Confirmation of *pCas13a* expression in *planta*.

Figure S2: pCas13a-mediated interference with TuMV-GFP in *planta*.

Figure S3: GFP quantification of TuMV-GFP interference in transgenic pCas13a-OE plants.

A



B

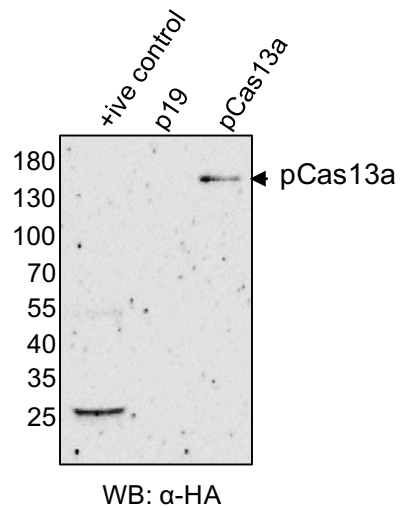
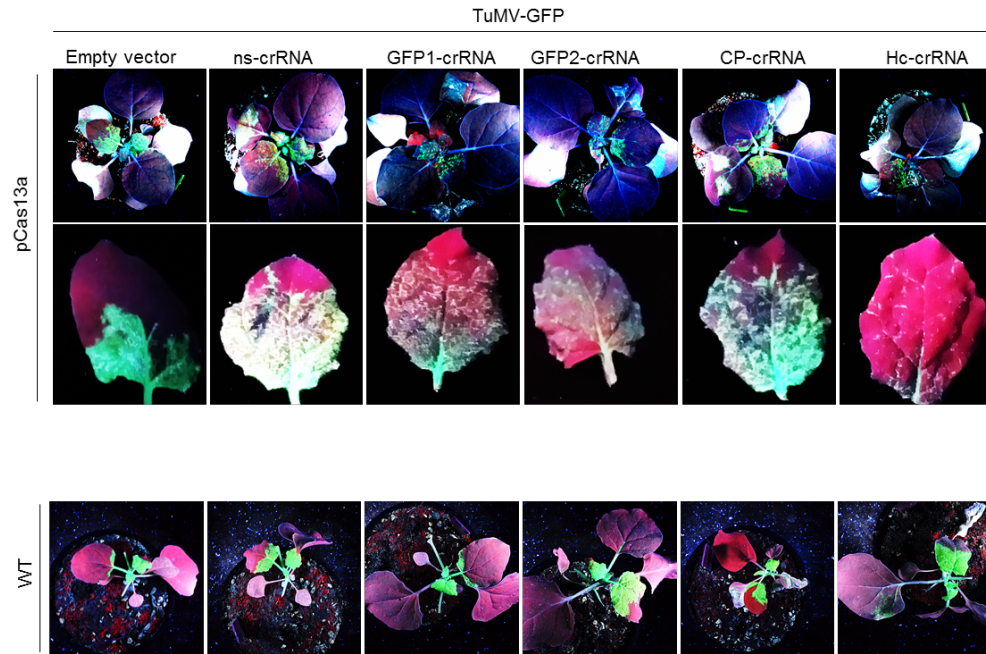


Figure S1. Confirmation of *pCas13a* expression in *planta*.

- A) Schematic for the expression of Cas13a in *planta*. The *LshCas13a* was plant codon optimized (*pCas13a*) and cloned into the binary vector *pK2GW7*.
- B) Proteins were extracted from *N. benthamiana* leaves infiltrated with *Agrobacterium* having the binary construct *pK2GW7-pCas13a* and p19. The protein blot was developed for the detection of HA-pCas13a with anti-HA antibody. The *N. benthamiana* line expressing an unrelated 27-kDa HA-

tagged protein was used as positive control. P19 alone was used as the negative control. The arrow indicates the detection of the expected size 171 kDa pCas13a.

A



B

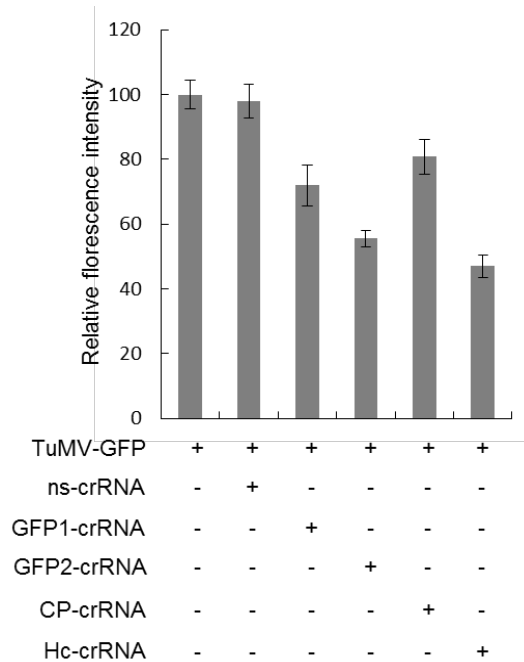


Figure S2. pCas13a-mediated interference with TuMV-GFP in *planta*.

A) GFP imaging of the viral interference in plants transiently expressing pCas13a and crRNA. *Agrobacterium* containing the *pK2GW7-pCas13a*, TuMV-GFP and TRV expressing crRNA (against GFP, HC-pro and CP of TuMV-GFP) were infiltrated into wild-type *N. benthamiana*.

Plants were imaged under UV-light for the systemic spread of TuMV-GFP at 7 dai. The representative third leaf of individual plants are shown in the middle panel. The lower panel represents the WT *N. benthamiana* infiltrated with TuMV-GFP and TRV expressing the respective crRNAs without pCas13a.

- B) Relative interference of TuMV-GFP. The GFP intensity was used as relative reference to measure the pCas13a-mediated interference of TuMV-GFP. GFP signal was quantified for the reduction of the TuMV-GFP in plants ($n=5$).

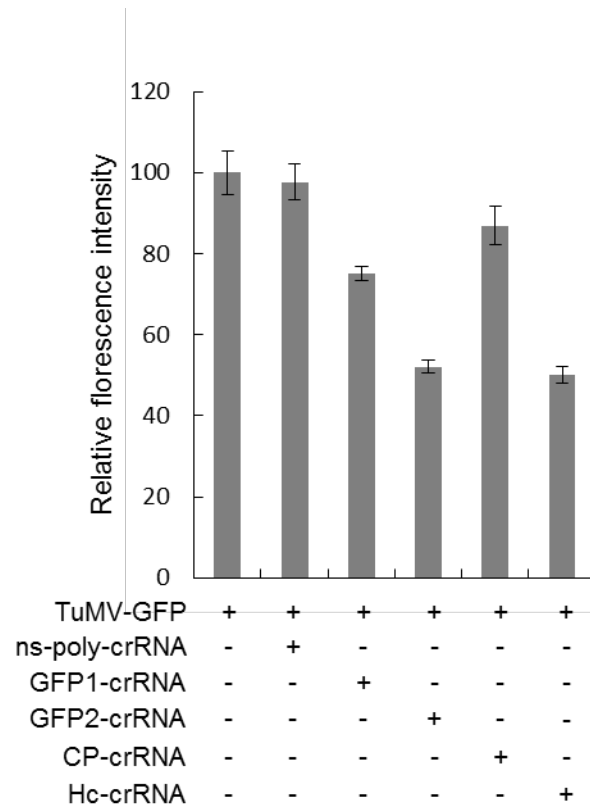


Figure S3. GFP quantification of TuMV-GFP interference in transgenic pCas13a-OE plants.

The GFP signal (plants $n=5$) was used for the generation of graph representing the comparative reduction of the GFP signal upon targeting.