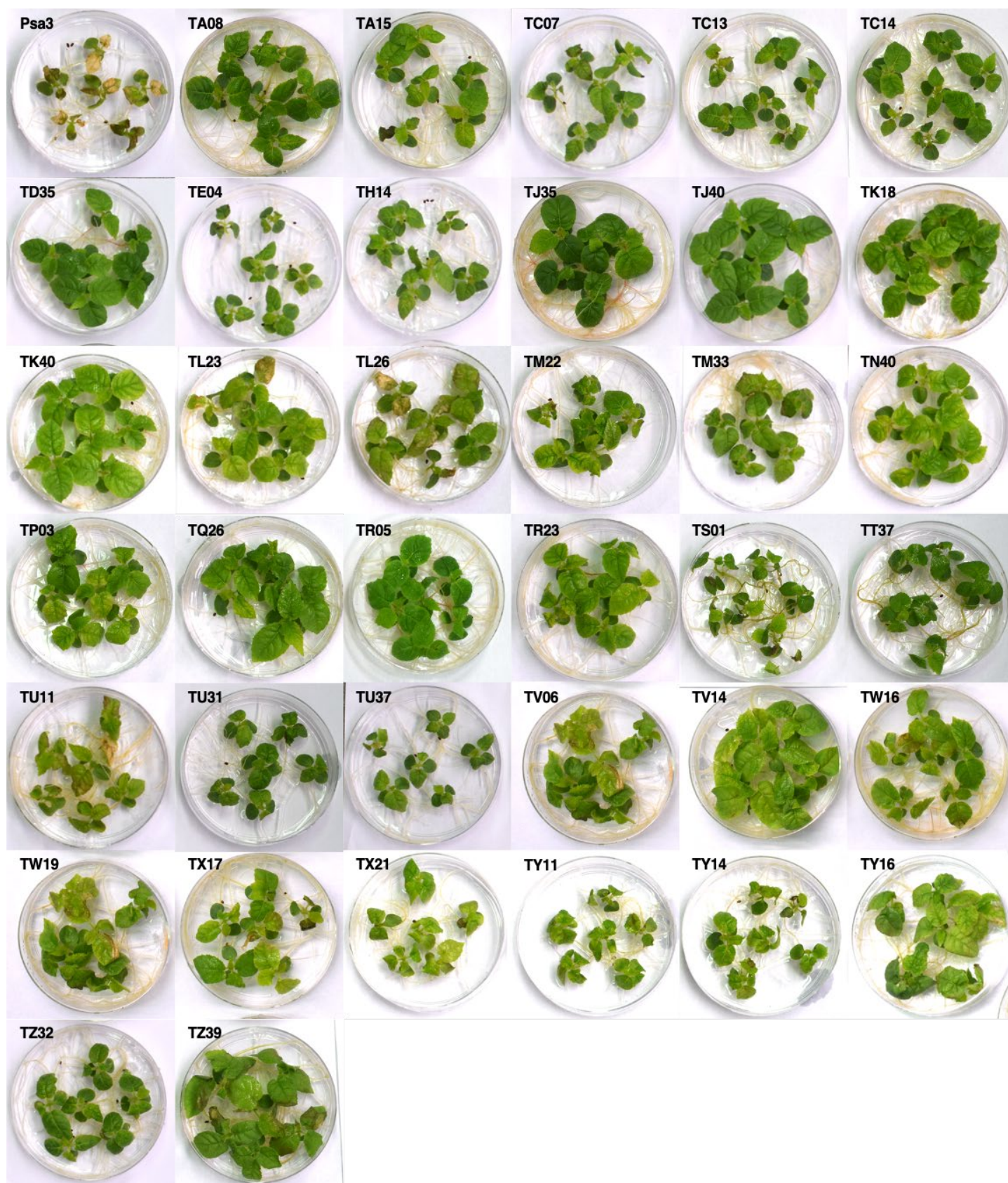


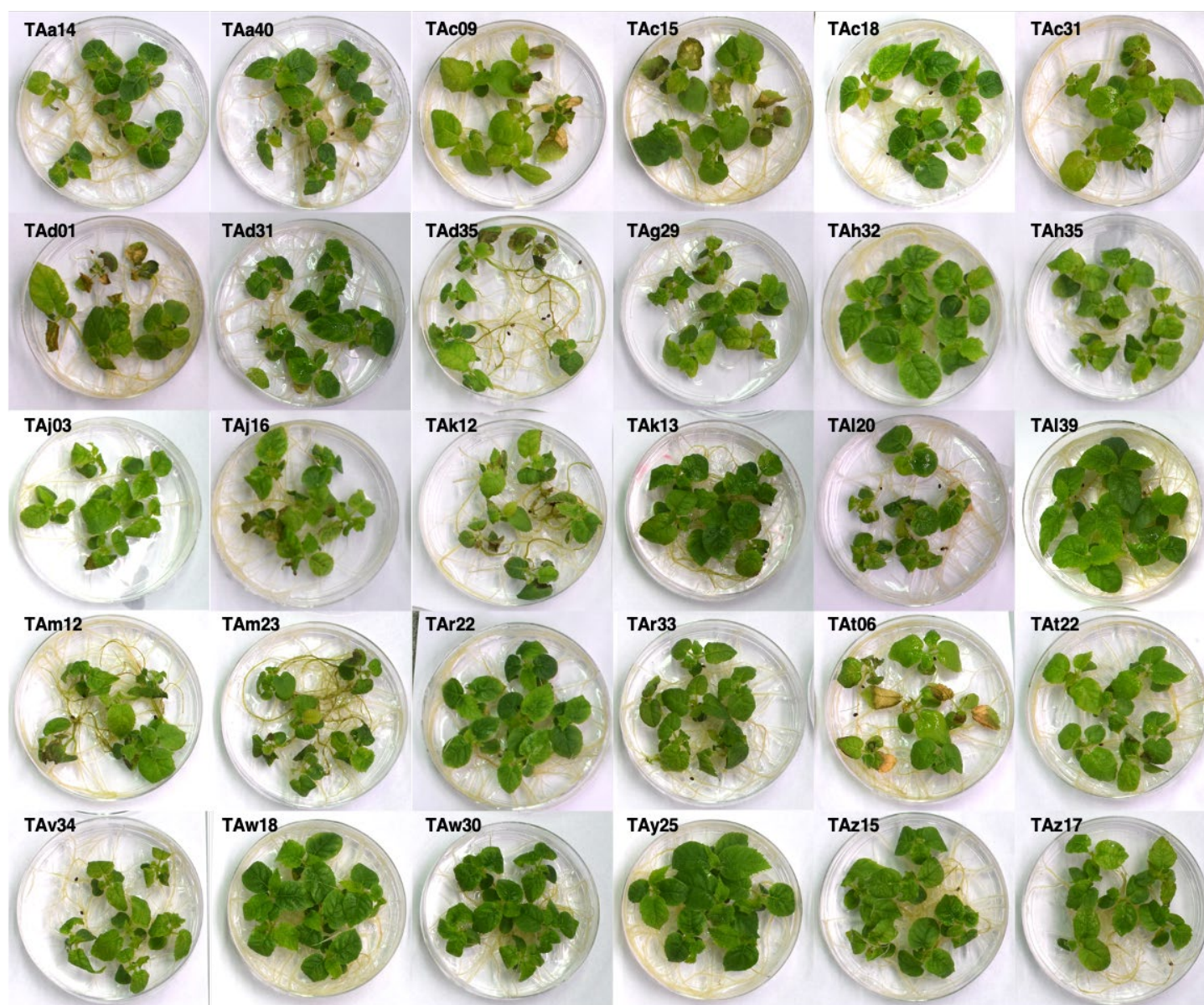
**Figure S1:** Large-scale Psa3 transposon mutagenesis screening using kiwifruit seedlings. (A) Planting the seeds on one-half strength MS plates. (B) Kiwifruit seedlings two weeks after germination. (C) Transferring the seedlings to new one-half strength MS plates with forceps. Seedling incubation at 24° C with a light intensity of 150-200  $\mu\text{E m}^{-2} \text{sec}^{-1}$  and a 12 h light/12 h dark photoperiod. (D) Four-week-old kiwifruit seedlings for screening. (E) Preparation of Psa3 transposon mutant inoculum ( $\text{OD}_{600}=0.2$ ). (F) Dip-inoculation for two kiwifruit seedlings. (G) Transferring the seedlings to new one-half strength MS plates. (H) Disease phenotypes of kiwifruit seedlings 14 days after inoculation with Psa3 transposon mutants.





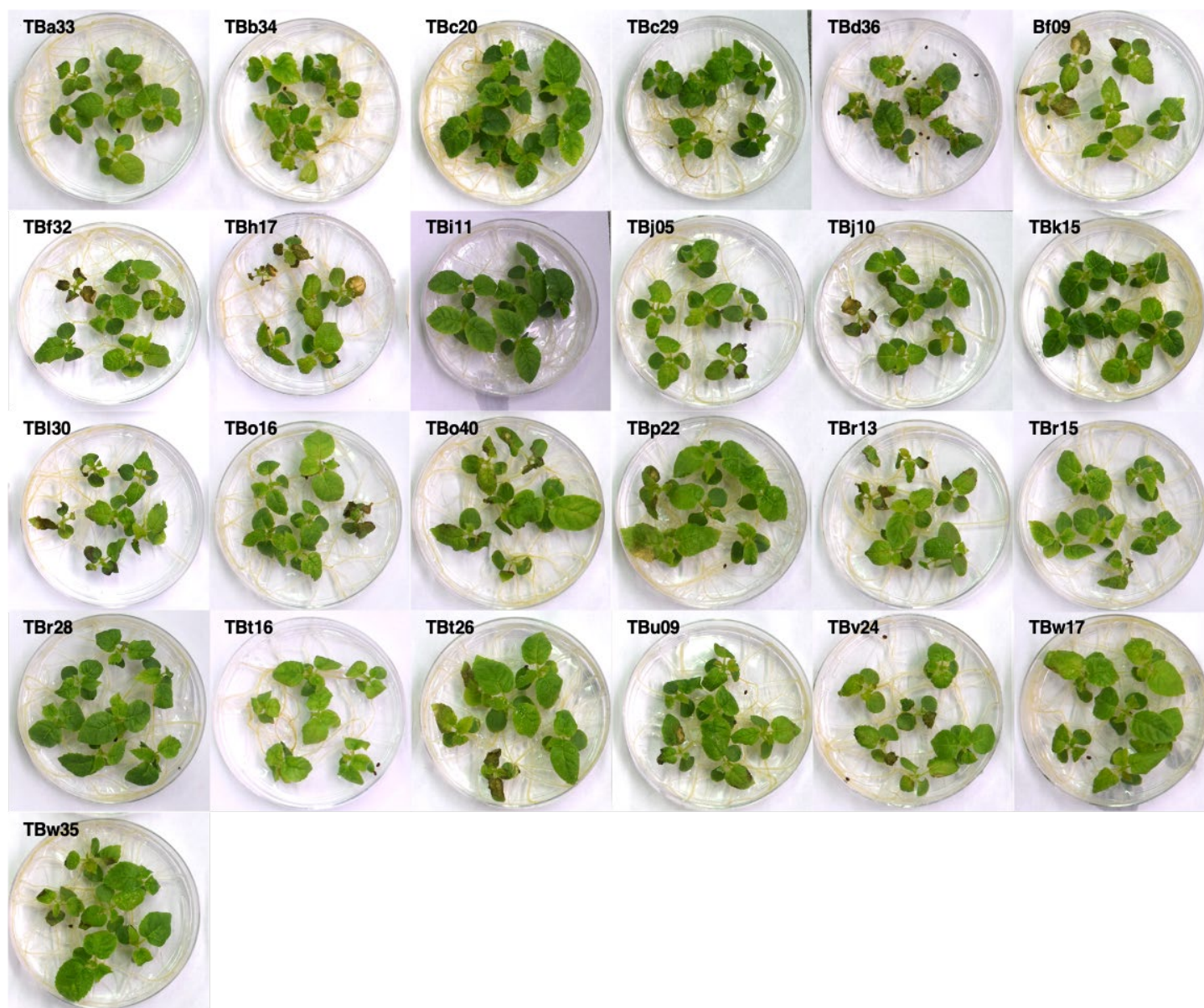
**Figure S2:** Second screening results in kiwifruit seedlings flood-inoculated with Tn5 transposon mutants (line number from 1 to 1,040). Disease symptoms in kiwifruit seedlings flood-inoculated with  $1 \times 10^8$  CFU/ml ( $OD_{600}=0.2$ ) of Psa3 WT and mutants containing 0.025% SilwetL-77.



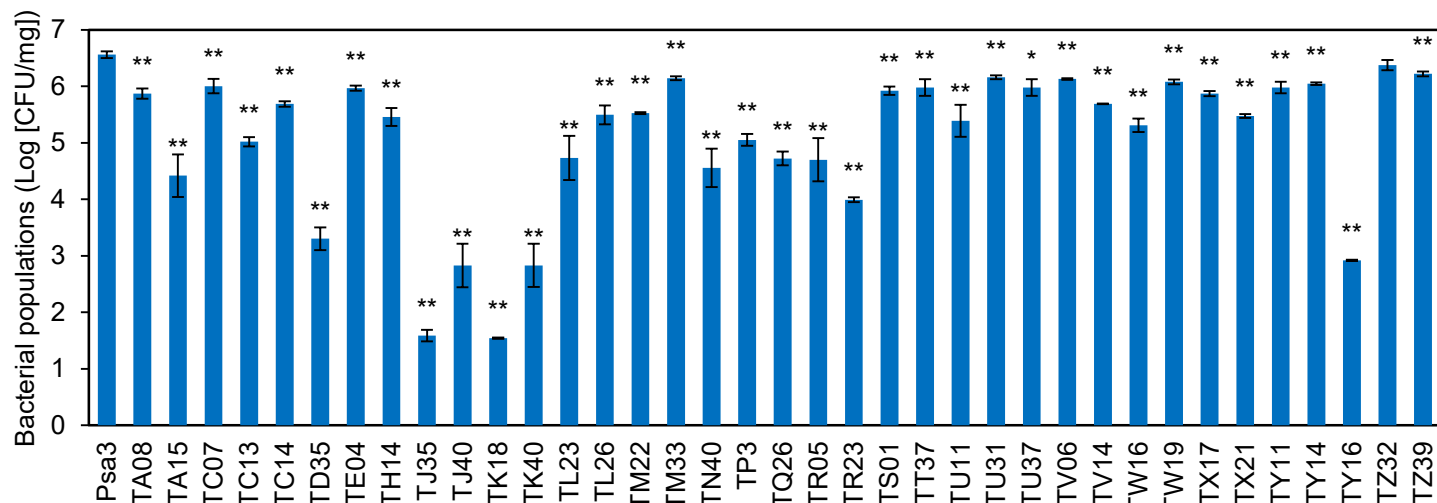
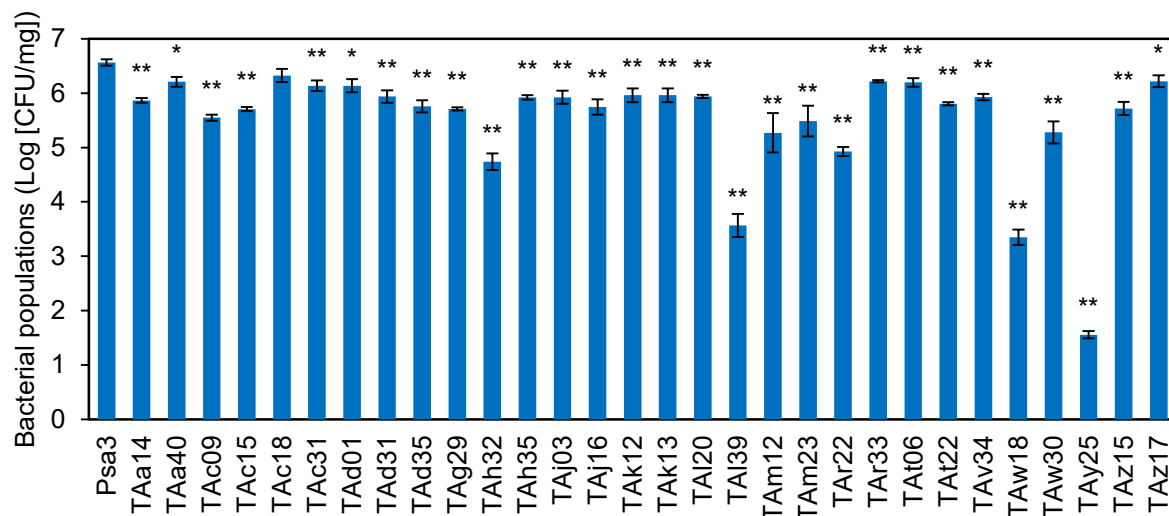
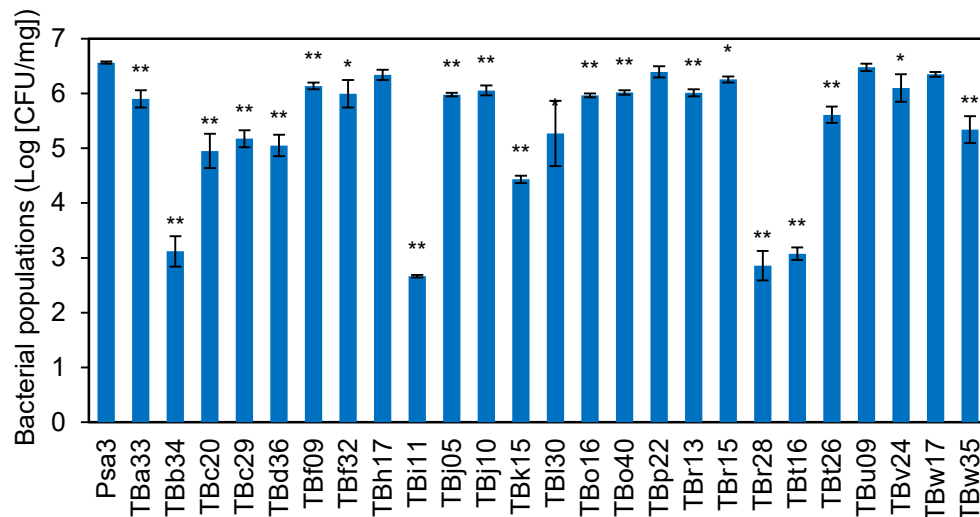


**Figure S3:** Second screening results in kiwifruit seedlings flood-inoculated with Tn5 transposon mutants (line number from 1,041 to 2,080). Disease symptoms in kiwifruit seedlings flood-inoculated with  $1 \times 10^8$  CFU/ml ( $OD_{600}=0.2$ ) of Psa3 WT and mutants containing 0.025% SilwetL-77.

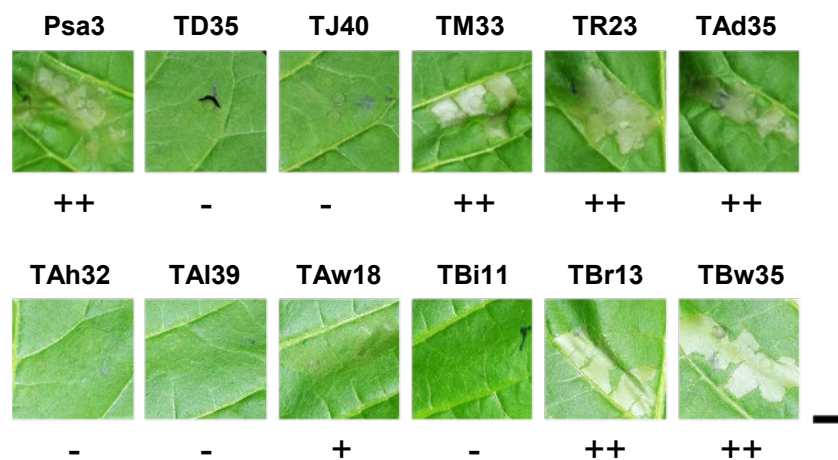




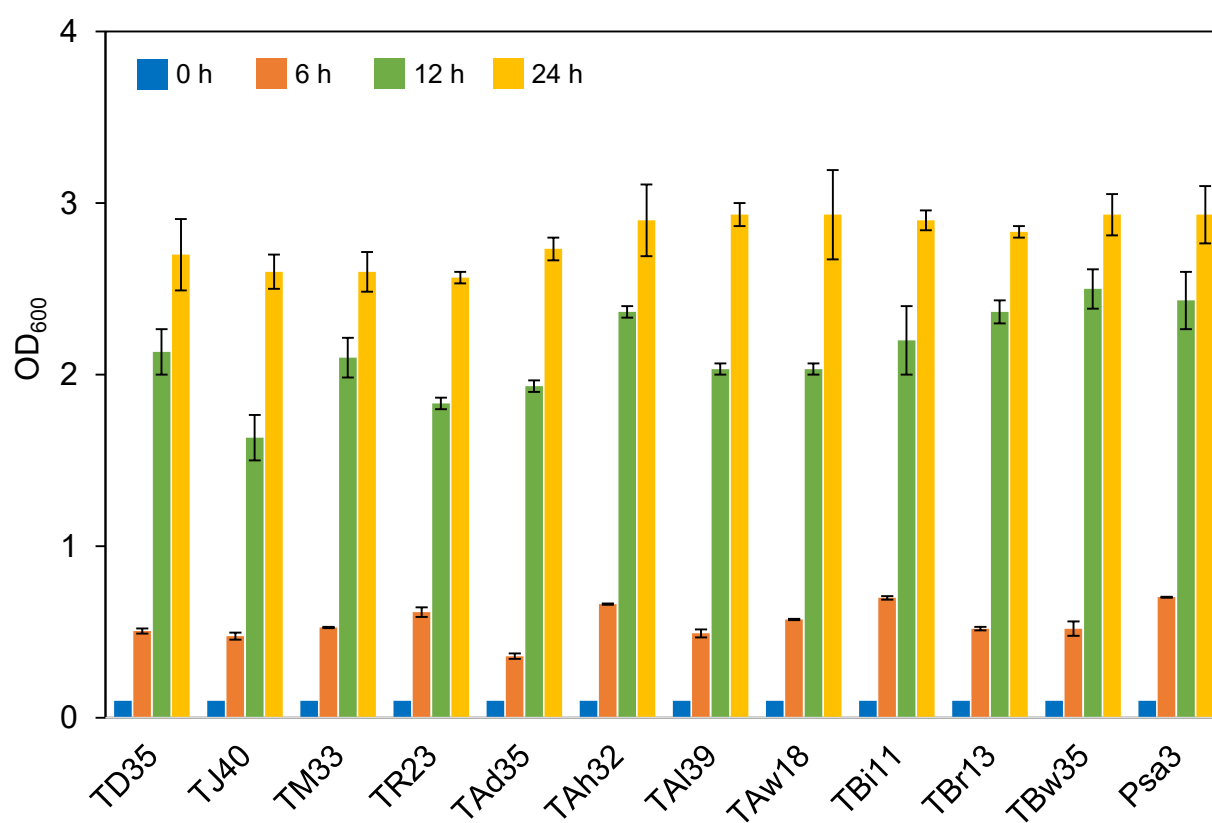
**Figure S4:** Second screening results in kiwifruit seedlings flood-inoculated with Tn5 transposon mutants (line number from 2,081 to 3,000). Disease symptoms in kiwifruit seedlings flood-inoculated with  $1 \times 10^8$  CFU/ml ( $OD_{600}=0.2$ ) of Psa3 WT and mutants containing 0.025% SilwetL-77.

**A****B****C**

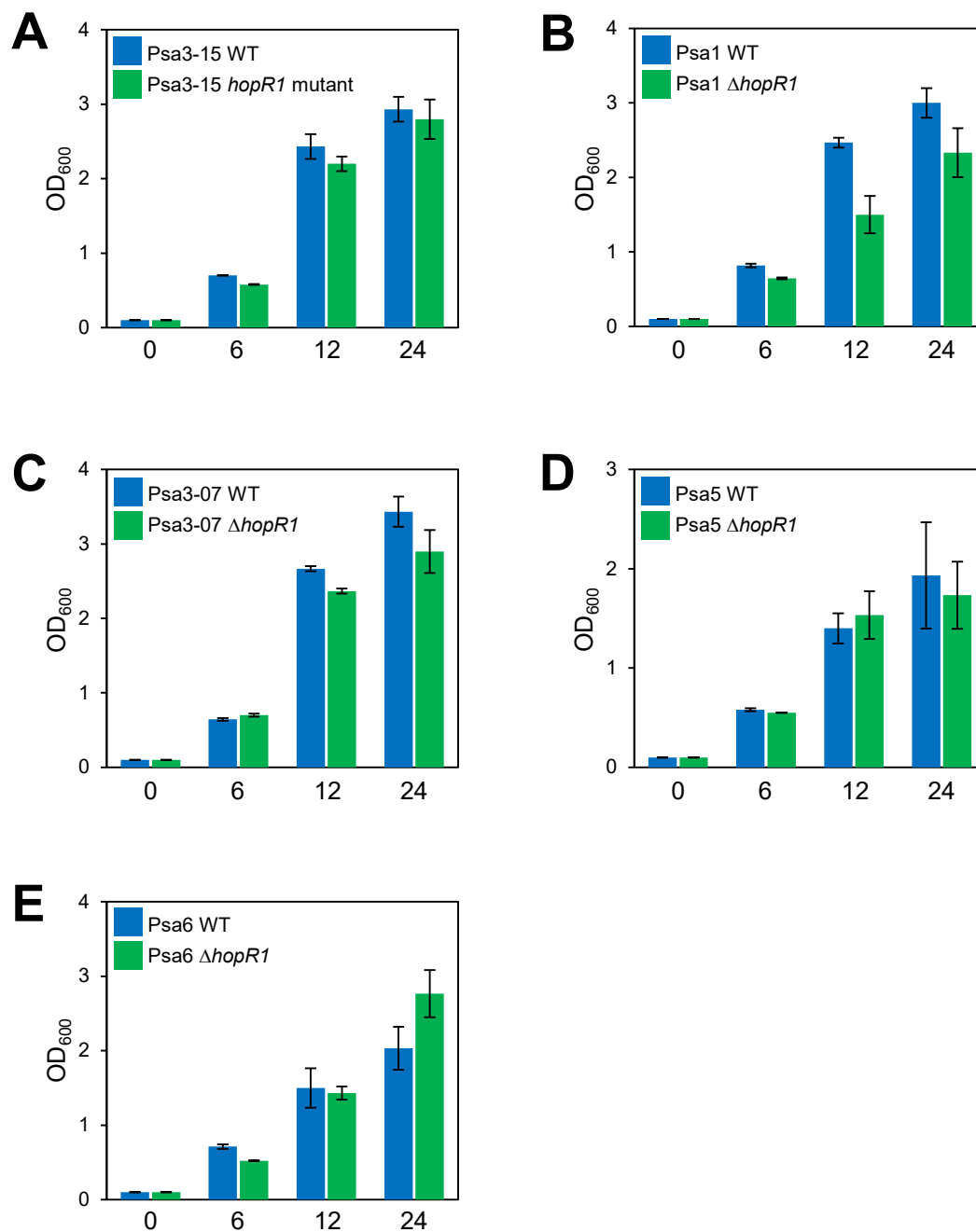
**Figure S5:** Bacterial population dynamics in kiwifruit seedlings flood-inoculated with Psa3 WT and mutants selected from the second screening. Bacterial populations in kiwifruit seedlings flood-inoculated with  $1 \times 10^8$  CFU/ml ( $OD_{600}=0.2$ ) of Psa3 WT and mutants from (A) line number from 1 to 1,040, (B) line number from 1,041 to 2,080 and (C) line number from 2,081 to 3,000 containing 0.025% SilwetL-77. The bacterial populations were obtained by homogenizing the inoculated leaves after surface-sterilization and plating dilutions to selective media at 7 days post-inoculation (dpi). Vertical bars indicate the standard error for three independent experiments. Asterisks indicate a significant difference from the WT and each mutant in a *t* test ( $*p < 0.05$ ,  $**p < 0.01$ ).



**Figure S6:** HR cell death assay with the type III secretion mutants in tobacco leaves. The leaf areas were infiltrated with type III secretion mutants indicated at  $5 \times 10^7$  CFU/ml, and photographed 1 day post-inoculation (dpi). ++, extensive necrosis; +, reduced necrosis; -, no symptoms.



**Figure S7:** Growth of type III secretion mutants in LB medium. Absorbance at 600 nm was measured in 3 replicates per strain for 6, 12, and 24 hours. Vertical lines indicate the standard deviation.



**Figure S8:** Growth of *hopR1* mutants in each Psa biovar in LB medium. Absorbance at 600 nm was measured in 3 replicates per strain for 6, 12, and 24 hours. (A) Psa3-15 WT and TR23. (B) Psa1 WT and  $\Delta hopR1$  mutant (C) Psa3-07 WT and  $\Delta hopR1$  mutant. (D) Psa5 WT and  $\Delta hopR1$  mutant. (E) Psa6 WT (blue) and  $\Delta hopR1$  mutant.