

## Supporting Information for

### **m<sup>6</sup>A modification plays an integral role in mRNA stability and translation during pattern-triggered immunity**

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#### **This PDF file includes:**

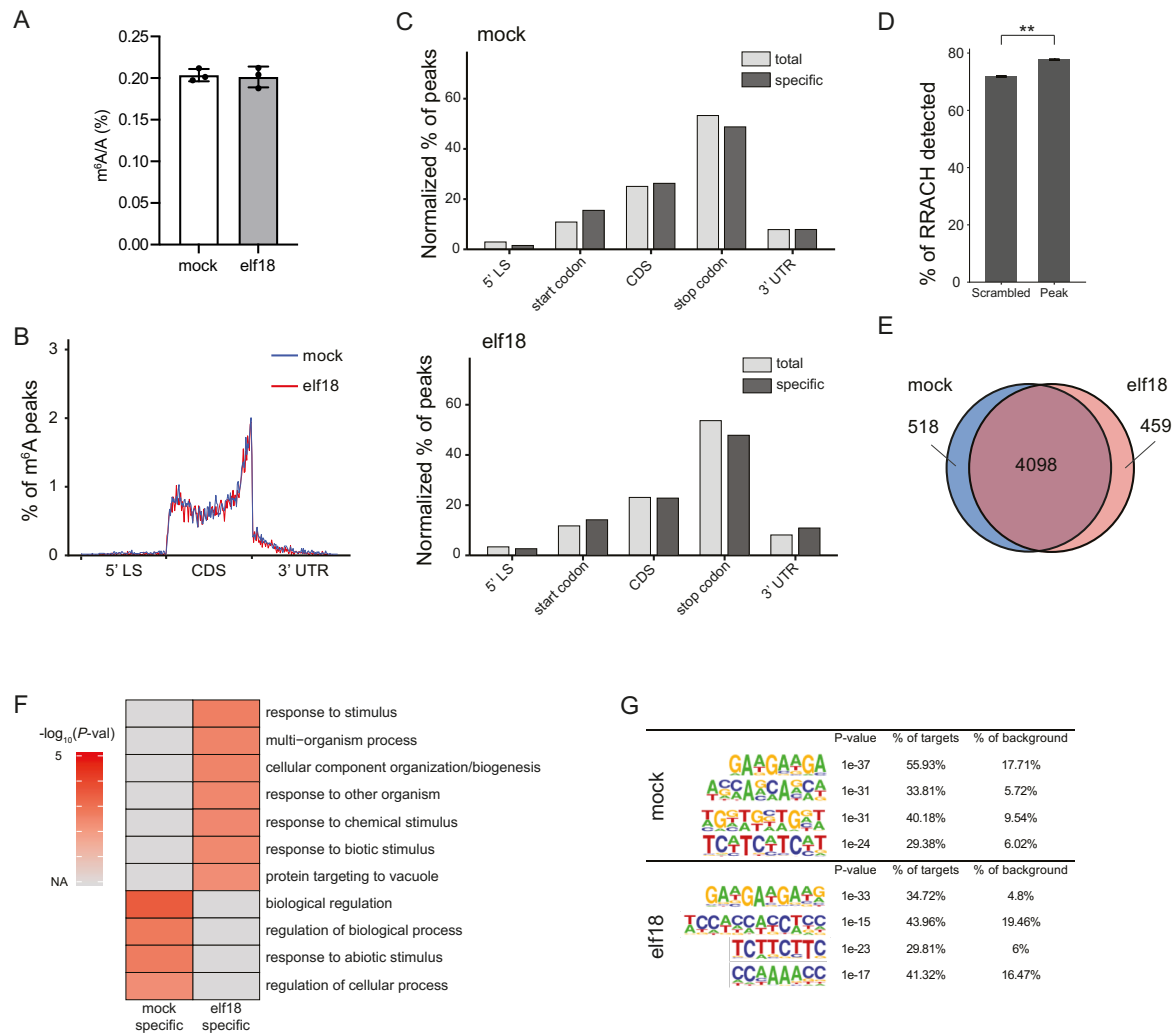
Figures S1 to S5

#### **Other supporting materials for this manuscript include the following:**

Dataset S1 to S6

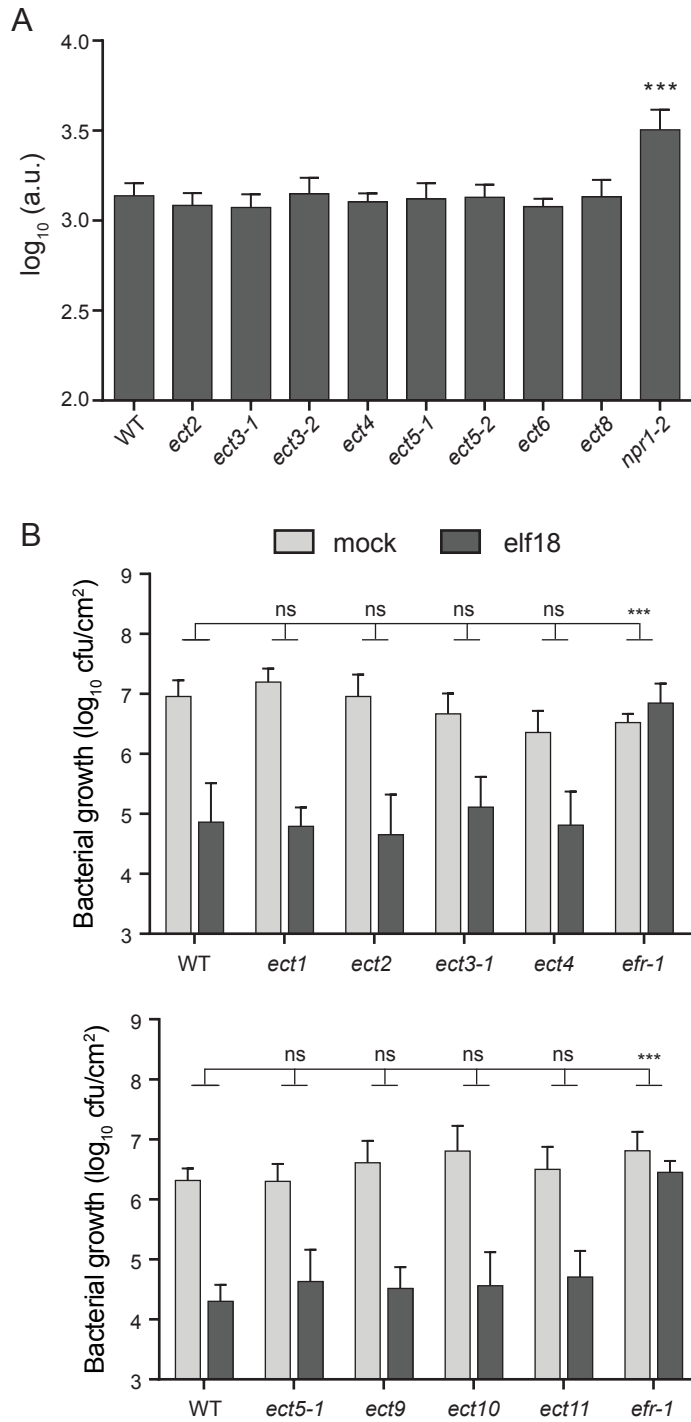
SI Appendix

Figures



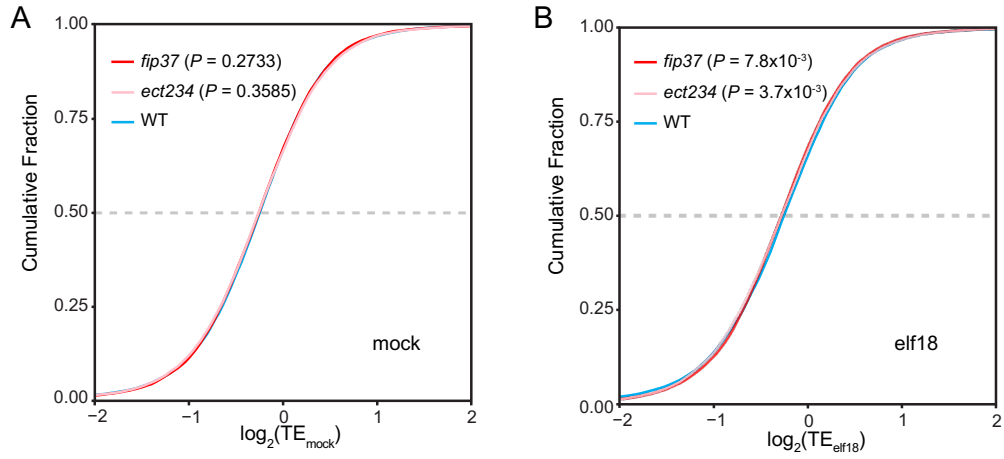
**Fig. S1.** Global analyses of m<sup>6</sup>A modification in response to elf18 treatment. (A) Quantification of global m<sup>6</sup>A modification with and without elf18 induction using liquid chromatography-tandem mass spectrometry. The m<sup>6</sup>A level was calculated as the ratio of m<sup>6</sup>A to A. (B) Global distribution of m<sup>6</sup>A modification in transcripts under mock- or elf18-treated condition (percentage to total). Position of m<sup>6</sup>A modification in the 5' leader sequence (5'LS), the coding sequence (CDS), and

the 3' untranslated region (UTR) of mRNA was normalized for each region. (C) Distribution of m<sup>6</sup>A modification in 5'LS, start codon  $\pm$  50 nt (start), CDS, stop codon  $\pm$  50 nt (stop), and 3'UTR. Upper panel, mock total and mock-specific (specific) m<sup>6</sup>A modification distribution; lower panel, elfl8 total and elfl8-specific m<sup>6</sup>A modification distribution. (D) FIMO (Find Individual Motif Occurrences) analysis of all m<sup>6</sup>A modifications for the presence of the RRACH-consensus compared to scrambled background sequences with equivalent length and nucleotide composition of modification site sequences. chi-squared test, \*\*  $P < 0.01$ . (E) Venn diagram of overlap between m<sup>6</sup>A modified mRNAs in mock- and elfl8-treated samples. (F) Heatmap of Gene Ontology enriched terms for the m<sup>6</sup>A-modified mRNAs present under mock (mock-specific) or elfl8 (elfl8-specific) condition. (G) Enrichment of *de novo* consensus in regions 150 nt flanking either end of the m<sup>6</sup>A modification site. Top panel, consensus in mock-specific transcripts. Bottom panel, consensus in elfl8-specific transcripts. Consensus enrichment performed with HOMER against a randomized background of equivalent length and nucleotide composition.

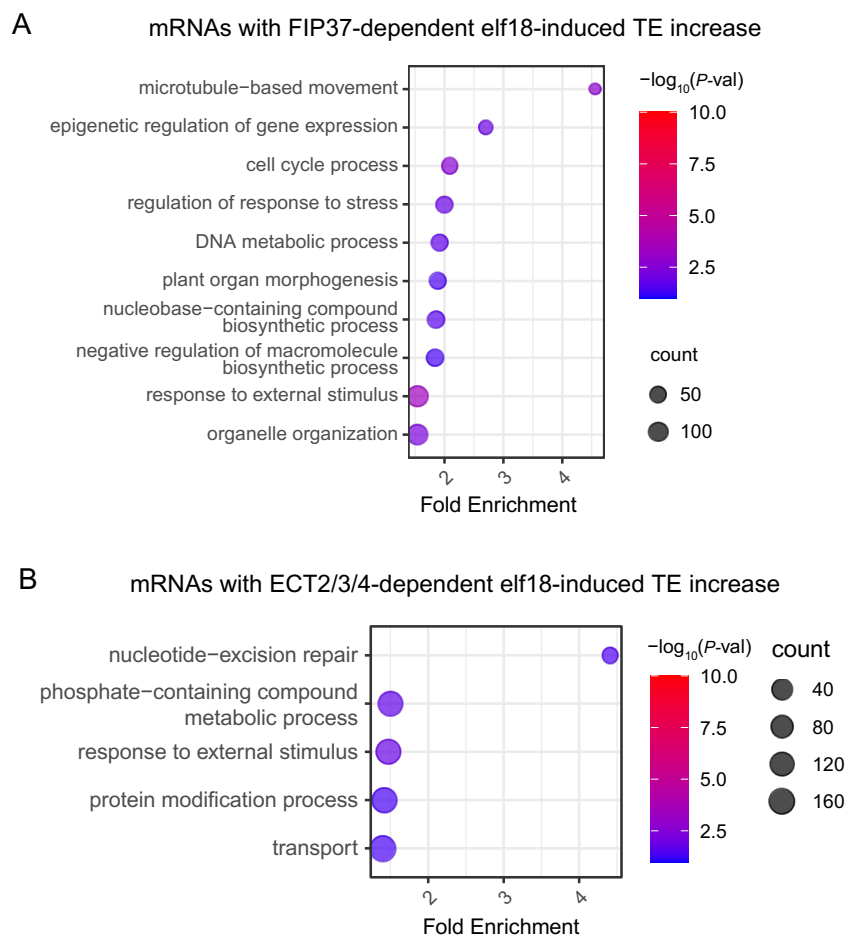


**Fig. S2.** Immune phenotypes of the m<sup>6</sup>A reader single mutants. (A) Basal resistance to a bacterial pathogen. Leaves from 3.5-week-old plants were infiltrated with *Pseudomonas syringae* pv *tomato* DC3000-LUX (*Pst-LUX*) (OD<sub>600nm</sub> = 0.0001). Bacterial growth was

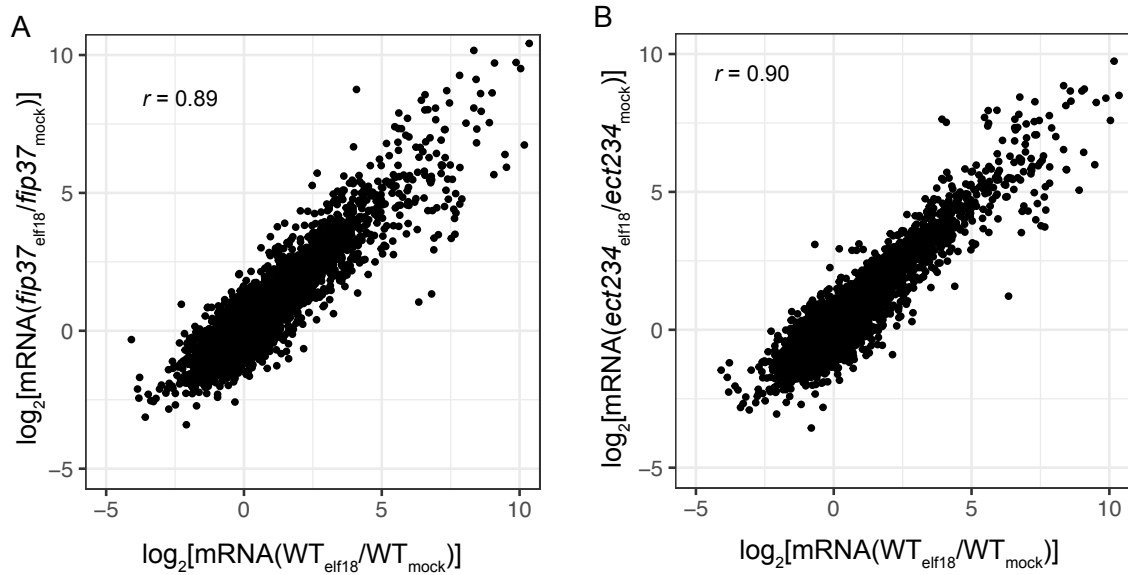
measured by luminescence after 3 days ( $n = 6$ ). *npr1*, *nonexpressor of pathogenesis-related 1* mutant known to have enhanced disease susceptibility. (B) elf18-induced resistance to a bacterial pathogen. Mature leaves were infiltrated with 1  $\mu$ M elf18 or mock ( $H_2O$ ). After 1 day, the same leaves were infiltrated with *Psm* ES4326 ( $OD_{600nm} = 0.001$ ) and bacterial growth was scored 2 days later ( $n = 8$ ). *efr-1*, an elf18-receptor mutant. All error bars represent 95% confidence intervals. Data were analyzed by the Student's t-test (A) and two-way ANOVA (B) with Bonferroni post hoc test. \*\*\*,  $P < 0.001$ ; ns, not significant.



**Fig. S3.** Global translation efficiency (TE) in the m<sup>6</sup>A writer and reader mutants. (*A* and *B*) Cumulative distribution of TE in WT, *fip37-4*, and *ect2/3/4* plants under mock (*A*) or elf18 (*B*) condition. *P* values were calculated by the Mann-Whitney test.



**Fig. S4.** Gene Ontology term enrichment for transcripts whose elf18-induced TE are dependent on FIP37 and ECT2/3/4. The 10% most affected mRNAs in *fip37-4* (A) or *ect2/3/4* (B) compared to WT were analyzed.



**Fig. S5.** The transcriptional changes induced upon *elf18* treatment were not defective in the m<sup>6</sup>A mutants. The scatterplots illustrate the correlation of transcriptional induction by *elf18* treatment between WT and *fip37-4* (A) or *ect2/3/4* (B) mutant.



**Datasets (separate files)**

**Dataset S1.** m<sup>6</sup>A peaks under mock- and elf18-treatments

**Dataset S2.** ECT2-binding peaks under mock- and elf18-treatments

**Dataset S3.** RNA decay rates under mock- and elf18-treatments

**Dataset S4.** Translation efficiency under mock- and elf18-treatments

**Dataset S5.** *ect* mutants used in this study

**Dataset S6.** Primers used in this study