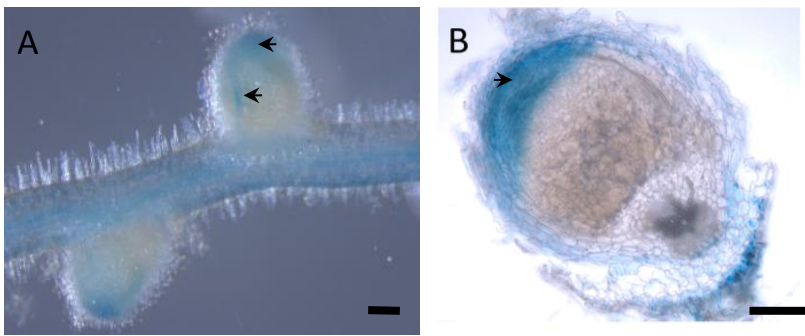


### Supplementary Figure 1. The induction of *CEP7* expression by rhizobium relies on NFP

Expression analysis of the *MtCEP7* gene in Wild-Type (WT) or *nfp* (*nod factor perception*) mutant roots 24 hours post rhizobium inoculation (hpi). qRT-PCR was used to measure gene expression levels in treated roots, normalized relative to untreated roots. Center lines show the medians; box limits indicate the 25<sup>th</sup> and 75<sup>th</sup> percentiles as determined by R software; whiskers extend 1.5 times the interquartile range from the 25<sup>th</sup> and 75<sup>th</sup> percentiles, outliers are represented by dots; crosses represent sample means; and data points from three biological replicates are plotted as open circles (n=9).



**Supplementary Figure 2. Rhizobia inoculation induces a *pMtCEP7:GUS* transcriptional fusion in vascular bundles and the apex of mature nodules**

**A-B.** The *pCEP7:GUS* activity was analyzed in mature nodules 14 days post rhizobium inoculation (dpi). A is a whole mount image, and B is a sagittal root and nodule section.

A minimum of 8 independent roots were analyzed for each condition from five independent experiments. Images show the GUS staining as a blue signal in bright field microscopy, indicated with arrows. Scale bars=200  $\mu$ m.

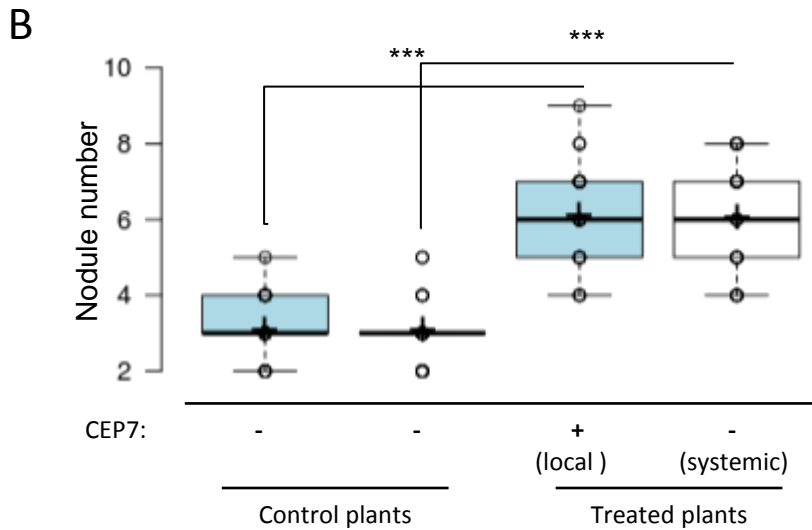
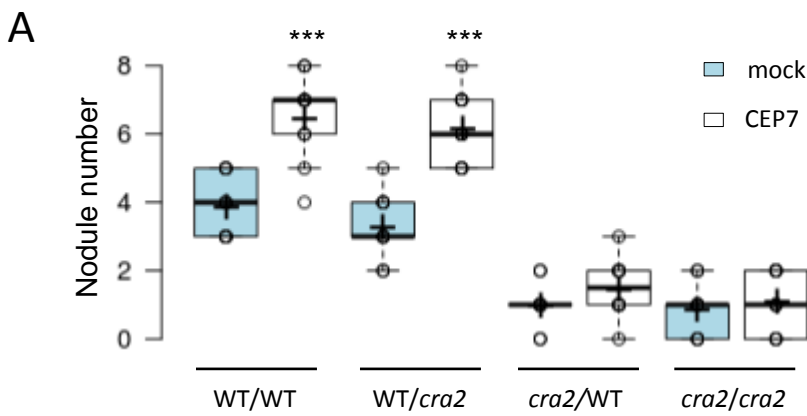
**A** MGLFQVTTKYLIVILALSIVYNSFQITQARPIKPLNQSSLNTQDSGAIHTN **SFRPTTPGSSPGVGH** RNFVVGDKNTR  
TMVVVQSPDVEVFVTNKRSDD **GFKPTNPSHSPGVGH** GYHTKIRHLN\*

**B** *GGGCATTGAACTCAATACAAAGATTATAAATAGCATAAGACCCCAGAGCTTGTGTTATAATTTCTTTCCATTG  
CCAATATATAGCTTAATCGTTCTCTCTAGTTTTAACCTACATATATCCTTTCACCTCACTTACCTTTGTTGTATAG  
TCATGGGTTTGTTC AAGTCACAACAAAATATTTGATCGTTATTCTAGCACTAAGTATTGTATACAATTCCTTTCA  
AATAACTCAAGCCAGGCCAATTAACCATTGAATCAACAATCTTCATTAACACACAAGACTCGGGTGCAATCC  
ACACTAACTCTTTTCGGCCGACAACACCAGGAAGTAGTCTGGTGTGGCCACCGAAATTTGTTGTAGG **AGAT**  
**AAGAACACGAGAACAAT** TGGTGGTTGTT CAGAGCCCGGATGTTGAGGTTTTTG TGACGAATAAGAGATCCGAT  
GATGGTTTCAAACCTACAAATCCTAGTCATAGTCTGGAGTTGCCATGGTTACCATACCAAAATTAGACATTT  
AAATTAGAACGTGATTGAATTCATTGTAGATTTCAAACCATCTTAATTAGAAGCTGTGTGTACCTACAATAATG  
TCCTTAGTTTCATTCTTCAAATTGCATGCCAT ATCATATGCATGTAGCACATGCACCTGGACTTTTGTTATTTGTTT  
AAGAGGCTTGGCTTTTGGTTATTGTTTGGGGGTGCATAATCAAAGTATGTAAAGTTTACTCTATTTATTTGTA  
ATTGTTTGGTTAATATGATACAAATCATAGCAAATAAATAAAAGATGTTTTAG*

**Supplementary Figure 3. CEP7 protein and genomic sequences**

**A.** MtCEP7 precursor sequence highlighting the CEP domains 1 (CEP7 D1, in blue) and 2 (CEP7 D2, in green).

**B.** CEP7 genomic sequence highlighting regions used for RNA interference (in blue) and artificial microRNA (in yellow) strategies. Italics indicate predicted 5' and 3' untranslated regions (UTRs), and initiation and stop codons are underlined.

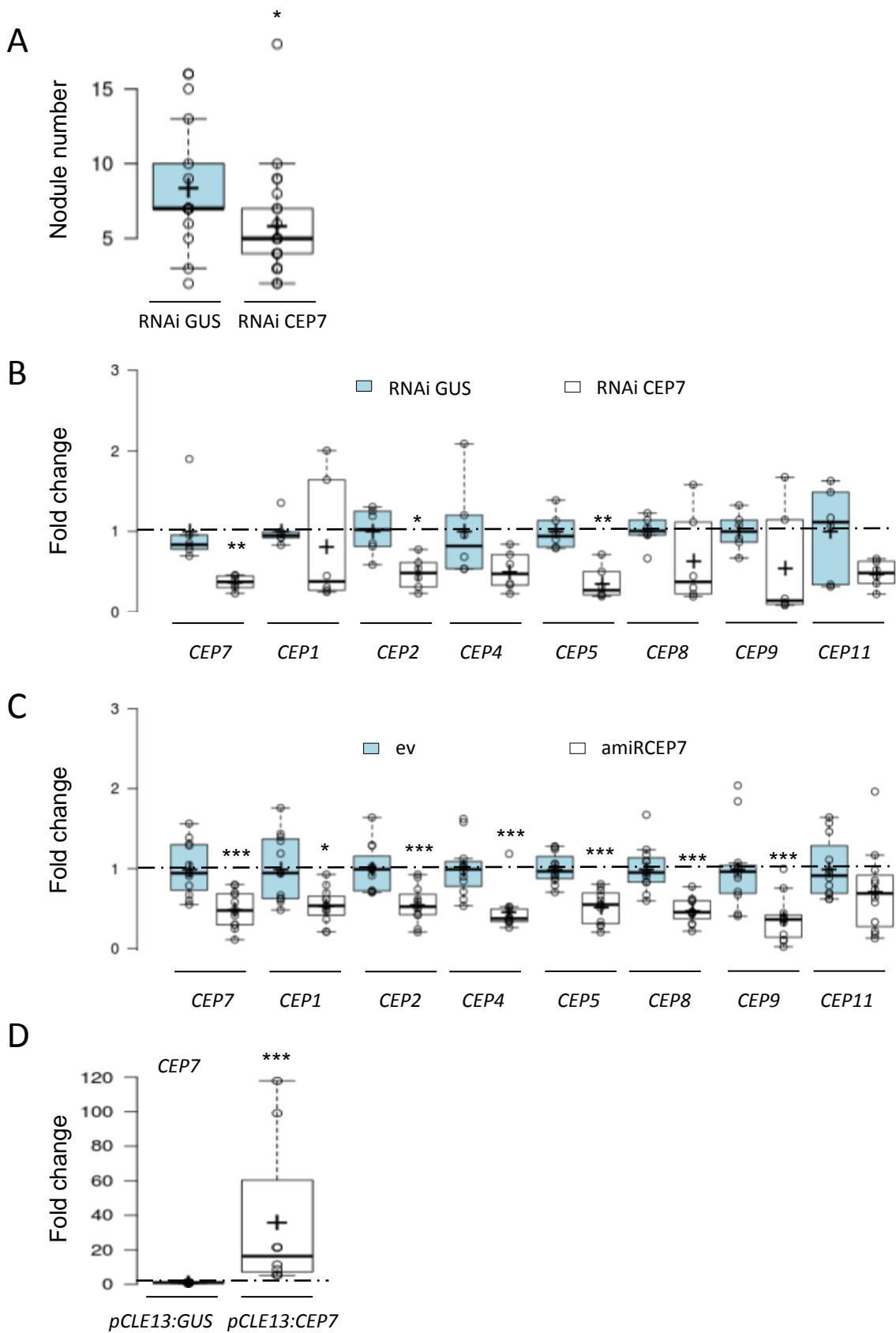


#### Supplementary Figure 4. CEP7 promotes nodulation systemically

**A.** Nodulation phenotype of grafts between Wild-Type (WT) and *cra2* mutants treated or not with synthetic MtCEP7 peptides. Quantification of the nodule number in different combinations of WT and *cra2* mutant grafted plants, grown *in vitro* on a nitrogen-free Fahraeus medium supplemented or not with 1  $\mu$ M CEP7 Domain1 (CEP7 D1) peptides. Nodule number was scored at 14 days post rhizobium inoculation (dpi). Data points from one representative biological experiment out of two are plotted as open circles ( $n \geq 9$ ). A Mann-Whitney test was used for each graft combination to assess significant differences between peptide-treated plants and the control, as indicated by asterisks (\*\*\*)  $\alpha < 0.001$ .

**B.** Nodulation phenotype of local and systemic roots from plants grown in a split-root experimental system. Quantification of the nodule number in split-root WT plants, grown *in vitro* on a nitrogen-free Fahraeus medium, where half of the root system was treated, or not, with 1  $\mu$ M CEP7 Domain1 (CEP7 D1) peptides. Rhizobium inoculation was performed on the whole root system. Nodule number was scored at 14 days post-rhizobium inoculation (dpi). Data points from one representative biological experiment out of two are plotted as open circles ( $n \geq 10$ ). A Mann-Whitney test was used for each split root combination to assess significant differences between local and systemic roots for peptide-treated plants and the control, as indicated by asterisks (\*\*\*)  $\alpha < 0.001$ .

In A-B, center lines show the medians; box limits indicate the 25<sup>th</sup> and 75<sup>th</sup> percentiles as determined by R software; whiskers extend 1.5 times the interquartile range from the 25<sup>th</sup> and 75<sup>th</sup> percentiles, outliers are represented by dots; and crosses represent sample means.



**Supplementary Figure 5. Efficiency and specificity of RNA interference, artificial microRNA and CEP7 ectopic expression**

**A.** Nodulation phenotype of plants where *MtCEP7* expression was silenced by RNA interference (RNAi).

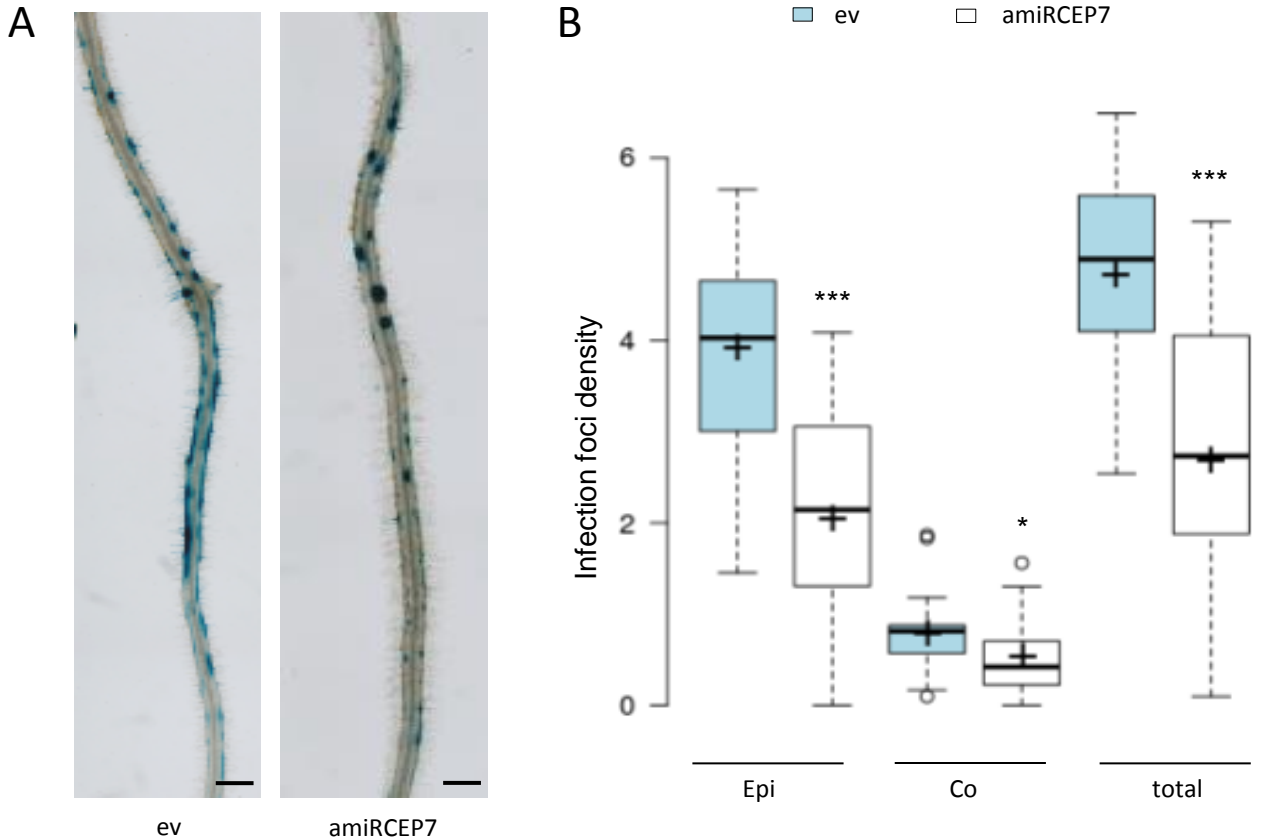
Quantification of the nodule number of Wild-Type (WT) plants expressing a RNAi construct driven by the 35S:CaMV promoter and targeting either *GUS* as a negative control or the *MtCEP7* gene. Plants were grown *in vitro* on a nitrogen-free Fahraeus medium and the nodules were scored at 14 days post rhizobium inoculation (dpi). Data points from one representative biological experiment out of two are plotted as open circles ( $n \geq 13$ ).

**B.** Expression analysis by qRT-PCR of *CEP* genes in roots expressing a *MtCEP7* RNAi or a *GUS* RNAi construct, as a negative control, grown as described in A. The expression data were normalized relative to the *GUS* control. The dotted line corresponds to a ratio of 1 to highlight fold changes. Data points from three biological replicates are plotted as open circles ( $n=6$ ).

**C.** Expression analysis by qRT-PCR of *CEP* genes in roots transformed either with an empty vector (*ev*) or expressing an *amiRCEP7* construct, grown as described in Figure 6D-F. Expression levels in *amiRCEP7* roots were normalized relative to control roots. The dotted line corresponds to a ratio of 1 to highlight fold changes. Data points from six biological replicates are plotted as open circles ( $n=12$ ).

**D.** Expression analysis by qRT-PCR of *MtCEP7* in roots transformed either with a *pCLE13:GUS* or a *pCLE13:CEP7* construct, grown as described in Figure 6G-I. Expression levels were normalized relative to *pCLE13:GUS* control roots. The dotted line corresponds to a ratio of 1 to highlight fold changes. Data points from four biological replicates are plotted as open circles ( $n=8$ ).

In A-D, center lines show the medians; box limits indicate the 25<sup>th</sup> and 75<sup>th</sup> percentiles as determined by R software; whiskers extend 1.5 times the interquartile range from the 25<sup>th</sup> and 75<sup>th</sup> percentiles, outliers are represented by dots; and crosses represent sample means. A Mann-Whitney test was used to assess significant differences, as indicated by asterisks (\*  $\alpha < 0.05$ ; \*\*  $\alpha < 0.01$ ; \*\*\*  $\alpha < 0.001$ ).



**Supplementary Figure 6. The amiRCEP7 construct decreases rhizobial infections**

**A.** Histochemical GUS staining of *pENOD11:GUS* roots five days post rhizobium inoculation (dpi), expressing either the empty vector (ev) or the amiRCEP7 construct. Bars=500  $\mu$ m.

**B.** Quantification using the *pENOD11:GUS* reporter shown in A of the density of rhizobial infection foci localized either in the root epidermis (Epi) or in the cortex (Co). The number of foci is normalized relative to the length (in mm) of the root region containing blue-stained foci. Center lines show the medians; box limits indicate the 25<sup>th</sup> and 75<sup>th</sup> percentiles as determined by R software; whiskers extend 1.5 times the interquartile range from the 25<sup>th</sup> and 75<sup>th</sup> percentiles, outliers are represented by dots; crosses represent sample means; and data points from two biological replicates are plotted as open circles ( $n \geq 22$ ). A Mann-Whitney test was used for each category to assess significant differences between amiRCEP7 roots and the control, as indicated by asterisks (\*  $\alpha < 0.05$ ; \*\*\*  $\alpha < 0.001$ ).