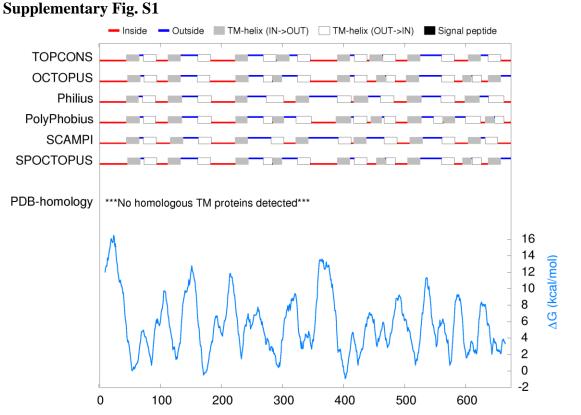
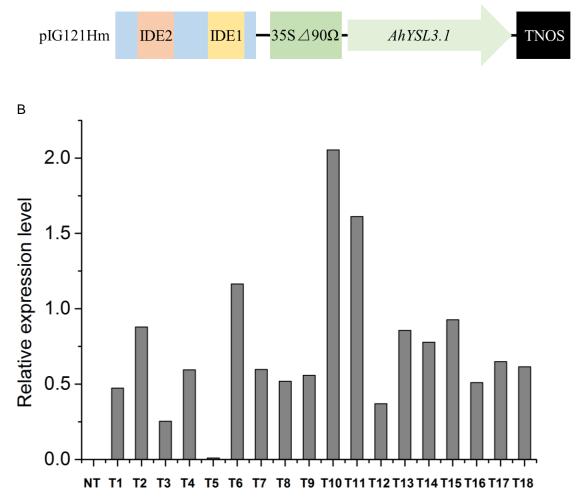




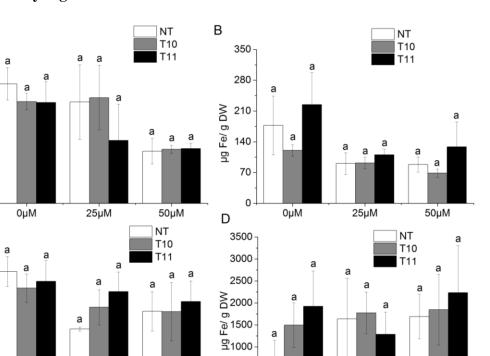
#### **Supplementary data**



Supplementary Fig. S1 The predicted membrane protein topology and signal peptide of AhYSL3.1 based on different methods.



Supplementary Fig. S2 Generation of *AhYSL3.1*-induced transgenic plants. (A) Schematic diagram of the binary vector for induction of AhIRT1 expression. Expression of the AhYSL3.1 gene was driven by an artificial promoter containing IDE1 and IDE2, which are cis-acting elements conferring Fe-deficiency-specific expression in tobacco roots, fused to the -90/+8 region of the cauliflower mosaic virus 35S promoter and a 5' leader ( $\Omega$ ) sequence of the tobacco mosaic virus to enhance basal expression (Kobayashi et al., 2003; Kobayashi et al., 2004). The pIG121Hm vector was used as a backbone. TNOS is the terminator of the nopaline synthase gene. (B) *AhYSL3.1* expression levels in shoots of the NT and transgenic lines. NT, non-transformed plants; T1–T18, transgenic lines.



а

0µM

25µM

50µM

# **Supplementary Fig. S3**

А

µg Fe/ g DW

С

160

120

80

40

0

60

40

0

0μ̈́M

а

25µM

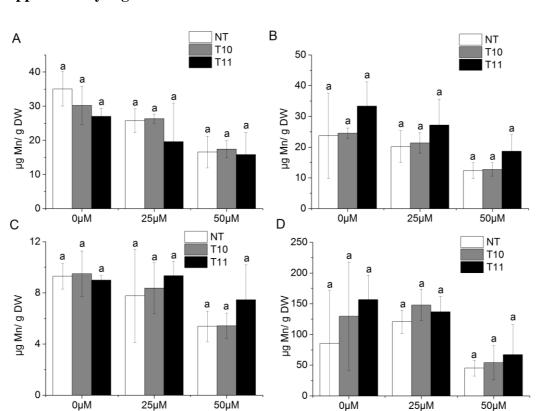
40 20 20

Supplementary Fig. S3 Fe concentrations in the NT and transgenic lines treated with various concentrations of Cu. (A) Young leaves, (B) old leaves, (C) stems, and (D) roots.

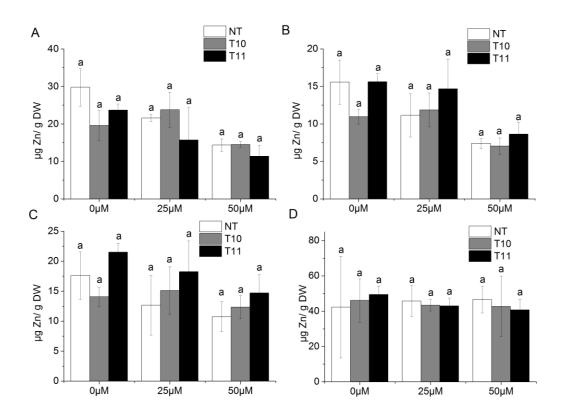
50µM

500

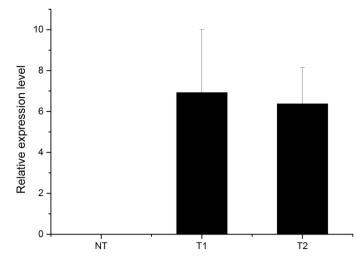
0



Supplementary Fig. S4 Mn concentrations in the NT and transgenic lines treated with various concentrations of Cu. (A) Young leaves, (B) old leaves, (C) stems, and (D) roots.

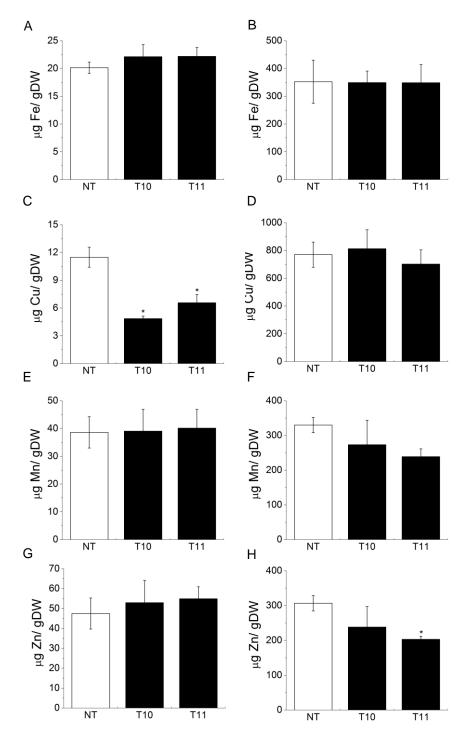


Supplementary Fig. S5 Zn concentrations in the NT and transgenic lines treated with various concentrations of Cu. (A) Young leaves, (B) old leaves, (C) stems, and (D) roots.

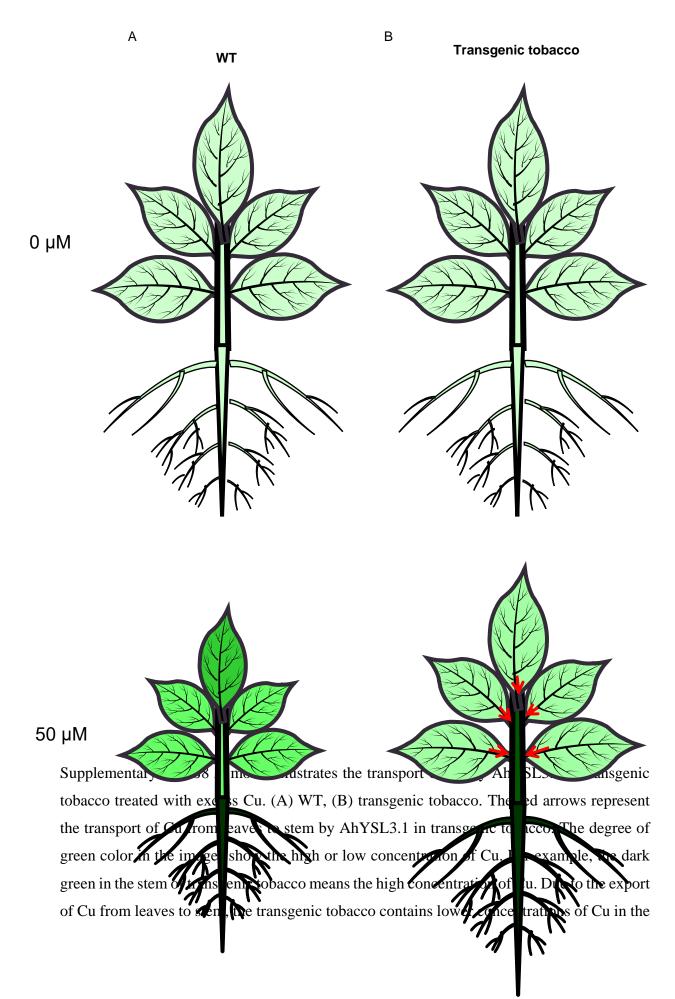


Supplementary Fig. S6 Relative expression level of *AhYSL3.1* in young leaves of NT and transgenic rice plants under excess Cu conditions.





Supplementary Fig. S7 Metal concentrations in young leaves and roots of transgenic and NT tobacco plants under Fe-deficient conditions. The tobacco plants were Fe-deprived for 9 days. (A, B) Fe, (C, D) Cu, (E, F) Mn, and (G, H) Zn concentrations in new leaves (A, C, E, G) and roots (B, D, F, H) of NT and transgenic plants. Results are presented as means  $\pm$  SD of triplicate samples. Significant differences from NT were determined by Student's *t*-test, \**P* < 0.05.



leaves and higher concentrations of Cu in the stem compared to the WT in the excess Cu condition. Thus, the transgenic plants are tolerant to excess Cu.