



Article

CsCuAO1 Associated with CsAMADH1 Confers Drought Tolerance by Modulating GABA Levels in Tea Plants

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Supplementary Materials

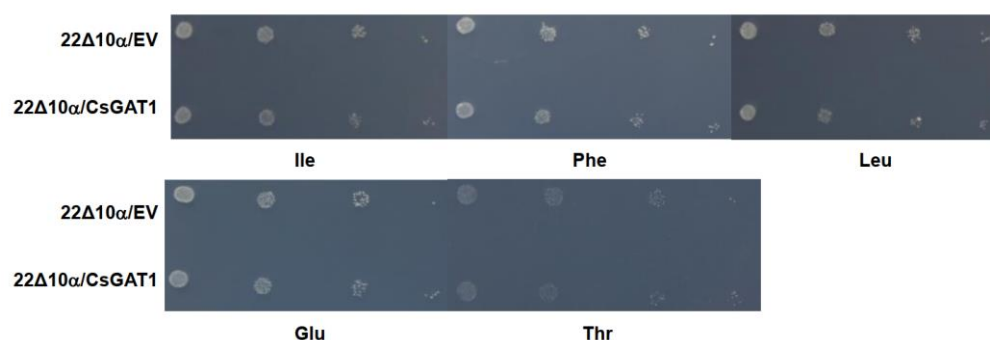


Figure S1. Phenotypes of yeast strains with CsGAT recombinant plasmids grown on solid medium, with amino acid as the only nitrogen source.

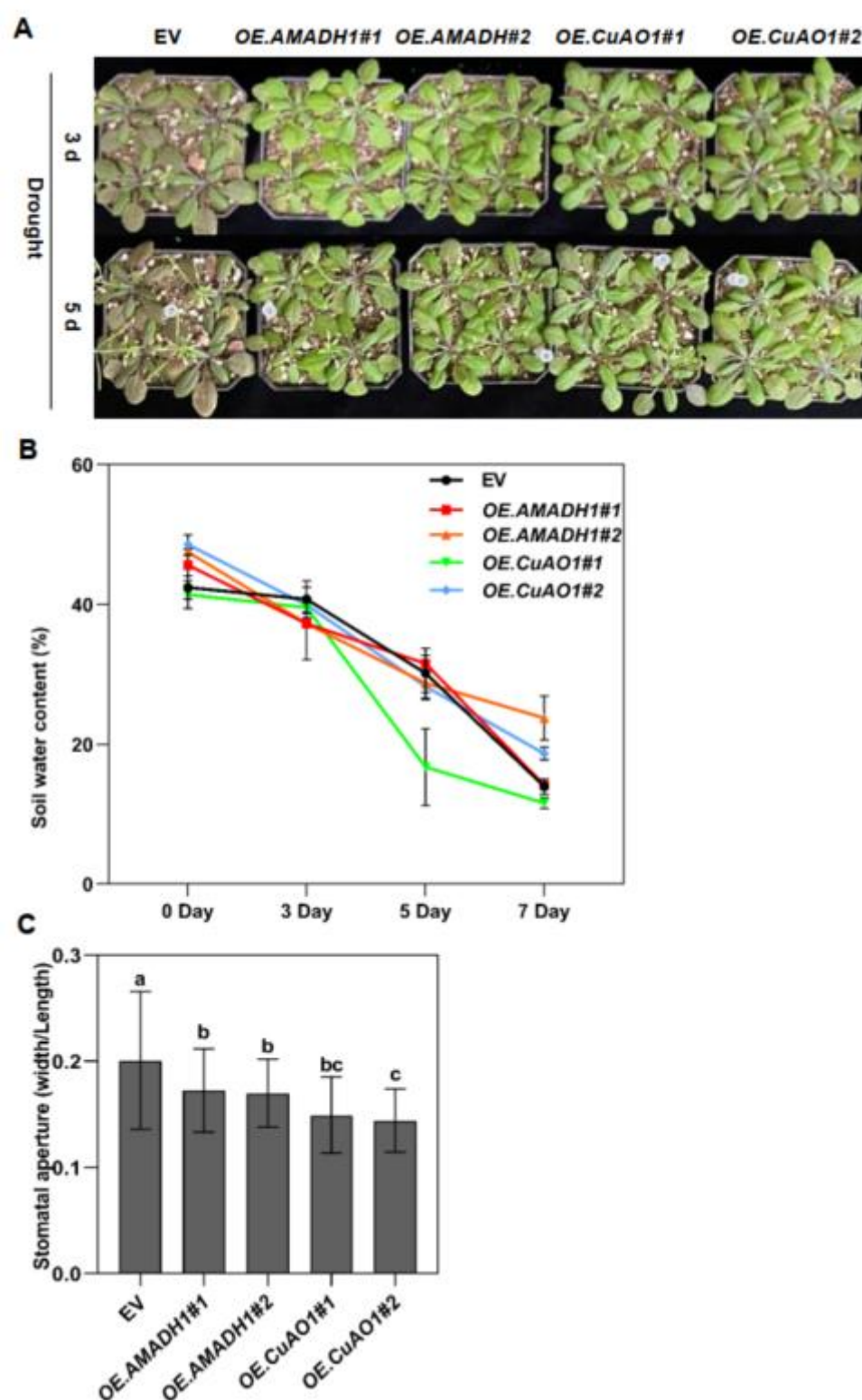


Figure S2. The overexpressing *Arabidopsis* lines exhibited high tolerance during drought treatment. (A) Phenotypes of two dependent lines of *CsAMADH1*-overexpressing and *CsCuAO1*-overexpressing four-week-old *Arabidopsis* before and after 3 and 5 days of drought treatment. The overexpressed empty vector (EV) was set as control. (B) Soil water content for each *Arabidopsis* line during drought treatment. (C) Stomatal apertures in guard cells of *Arabidopsis* leaves after 7 d of drought treatment. Different lowercase letters over columns indicate significant differences between lines ($p < 0.05$).

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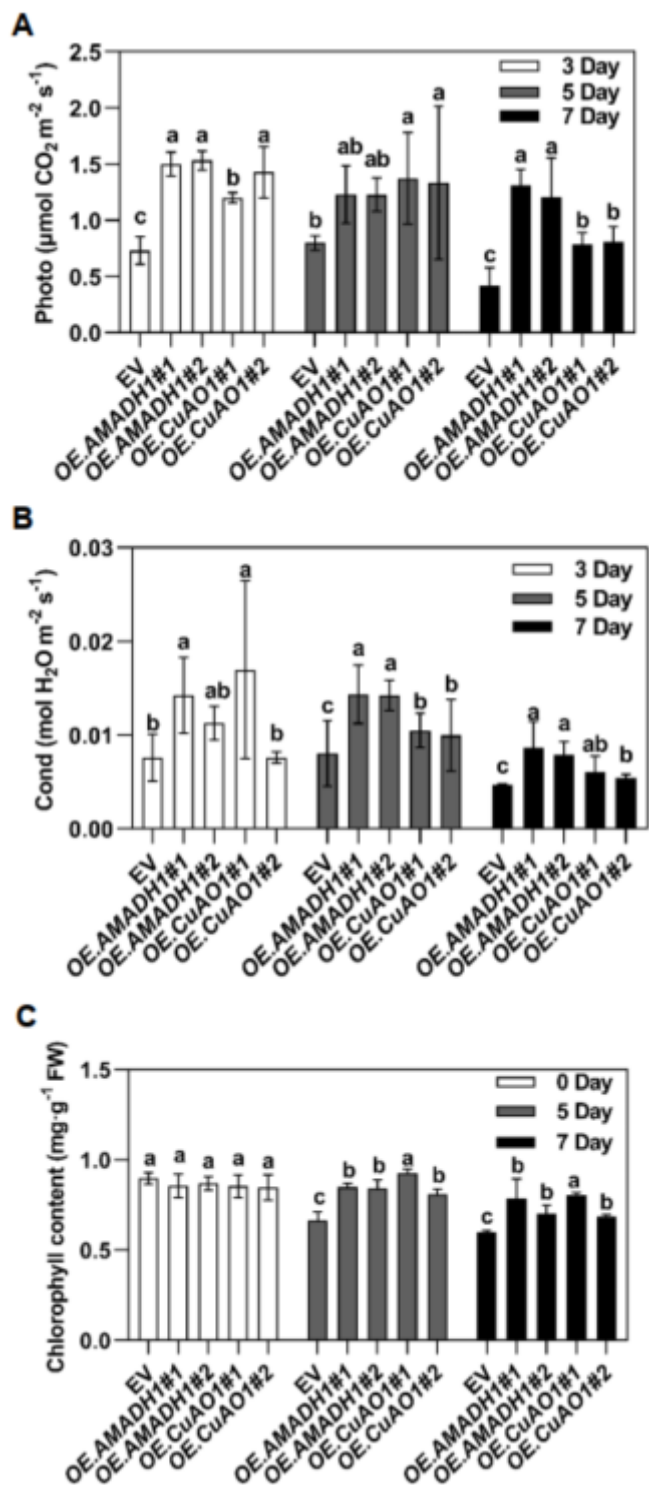


Figure S3. (A) Net photosynthetic rate and (B) stomatal conductance of *Arabidopsis* leaves after 7 d of drought treatment. (C) Chlorophyll content of *Arabidopsis* leaves after 7 d of drought treatment. Different lowercase letters over columns indicate significant differences between lines ($p < 0.05$).

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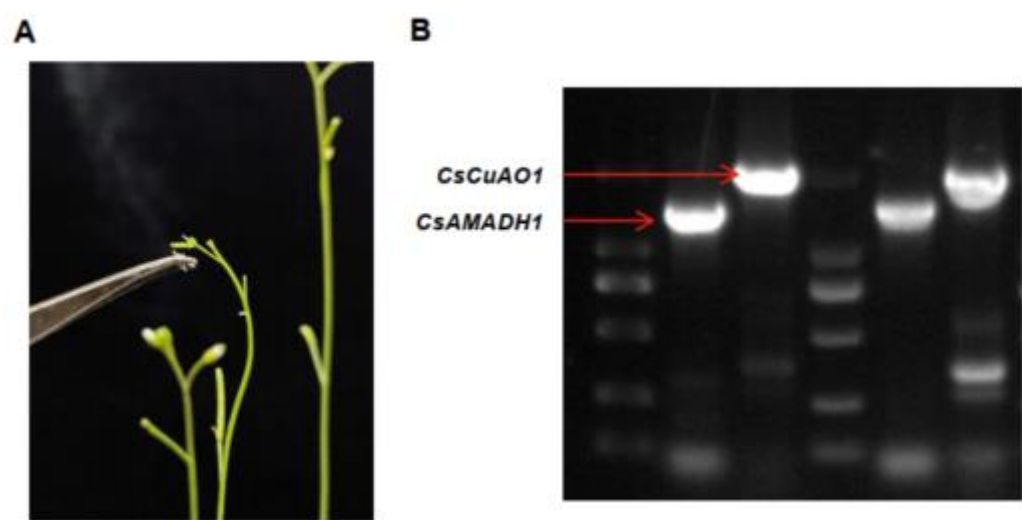


Figure S4. The generation of transgenic co-overexpressing lines of *CsCuAO1*–*CsAMADH1*. (A) *CuAO1*-overexpressing and *AMADH1*-overexpressing transgenic *Arabidopsis* were used as male and female parents, respectively, for pollen hybridization. (B) Photograph of electrophoresis analysis of co-overexpressing line.

Table S1. Genes used in this study.


Gene name		
		
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Gene name	Gene ID	Template size
<i>CsCuAO1</i>	TEA029860.1	2172bp
<i>CsCuAO3</i>	TEA015253.1	2040bp
<i>CsAMADH1</i>	TEA025182.1	1515bp

Table S2. Primers used in this study.

Primers	Sequence (5'-3')	Remarks
<i>Csβ-actin-F</i>	GCCATCTTTGATTGGAATGG	Internal reference
<i>Csβ-actin-R</i>	GGTGCCACAACCTTGATCTT	Internal reference
<i>CsGAT1-F</i>	GCAGTAGTGACAGTGACTT	qPCR
<i>CsGAT1-R</i>	GATGACGACAGACAGTGAT	qPCR
<i>CsGAD1-F</i>	AGTGACATCCAGAAAGTCTTGCT	qPCR
<i>CsGAD1-R</i>	CACCATTAGTCTTCTTCCTACTGAG	qPCR
<i>CsGAD2-F</i>	TTCGACATCTGCAAGGTGCTCCA	qPCR
<i>CsGAD2-R</i>	ACTTGTTGTTTCCTAGCCAAGAC	qPCR
<i>CsGAD3-F</i>	TTTCACATAACAAATGCAACGTC	qPCR
<i>CsGAD3-R</i>	CTCCCTTTGTCTTACCACCCAT	qPCR
<i>CsCuAO1-F</i>	AAACGCCCAACAACAAAT	qPCR
<i>CsCuAO1-R</i>	CGAACAACTCCGAATAAAC	qPCR
<i>CsCuAO3-F</i>	TATTGAGATTGCTGGACAC	qPCR
<i>CsCuAO3-R</i>	AACCCAAATTCGCCTA	qPCR
<i>CsAMADH1-F</i>	AGCCTGTGAAGAAGAAACG	qPCR

<i>CsAMADH1-R</i>	CCAGCAATAGAACGCAAA	qPCR
<i>CsCuAO1-F</i>	ATGGAAGAAAAGAGCTCCTC	PCR
<i>CsCuAO1-R</i>	GGCAGAAGCAGAAGCCTT	PCR
<i>CsCuAO3-F</i>	ATGGAAGCTTCAACATTGAG	PCR
<i>CsCuAO3-R</i>	AGGTTTGGCAGTGCAATT	PCR
<i>CsAMADH1-F</i>	ATGGCGATCCCAATACCTACTC	PCR
<i>CsAMADH1-R</i>	AAGCTTTGGCTTTGAAGGAGATGGG	PCR
<i>CsCuAO1-X-F</i>	ACACGGGGGACTCTAGAATGGAAGAAAA- GAGCCTCCTC	PCR
<i>CsCuAO1-B-R</i>	TGACCACCCGGGGATCCGGCAGAAGCAGAAGCCTT	PCR
<i>CsCuAO3-X-F</i>	ACACGGGGGACTCTAGAATGGAAGCTTCAACATT- GAG	PCR
<i>CsCuAO3-B-R</i>	TGACCACCCGGGGATCCAGGTTTGGCAGTGCAATT	PCR
<i>CsAMADH1-X-F</i>	ACACGGGGGACTCTAGAATGGCGATCCCAATAC- CTACTC	PCR
<i>CsAMADH1-B-R</i>	TGACCACCCGGGGATCCAAGCTTTGGCTTT- GAAGGAGATGGG	PCR
<i>AsODN-CsA-MADH1-1</i>	GGCGAGTAGGTATTGGGATC	Gene suppression
<i>AsODN-CsA-MADH1-2</i>	GAGCTGCAGCAGTCATGATC	Gene suppression
<i>AsODN-CsA-MADH1-3</i>	ATGGCTGTCGTTTGCCAGTT	Gene suppression
<i>AsODN-CsCuAO1-1</i>	CATGTGGAGGTGCAGTCGAG	Gene suppression
<i>AsODN-CsCuAO1-2</i>	TTGGGGATTGGTATGTTTCCT	Gene suppression
<i>AsODN-CsCuAO1-3</i>	GATTATTTGTGAATGCGCCT	Gene suppression

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