

Supplementary Figures

Mn₃O₄ nanoparticles maintain ROS homeostasis to modulate stomatal aperture to improve cotton drought tolerance

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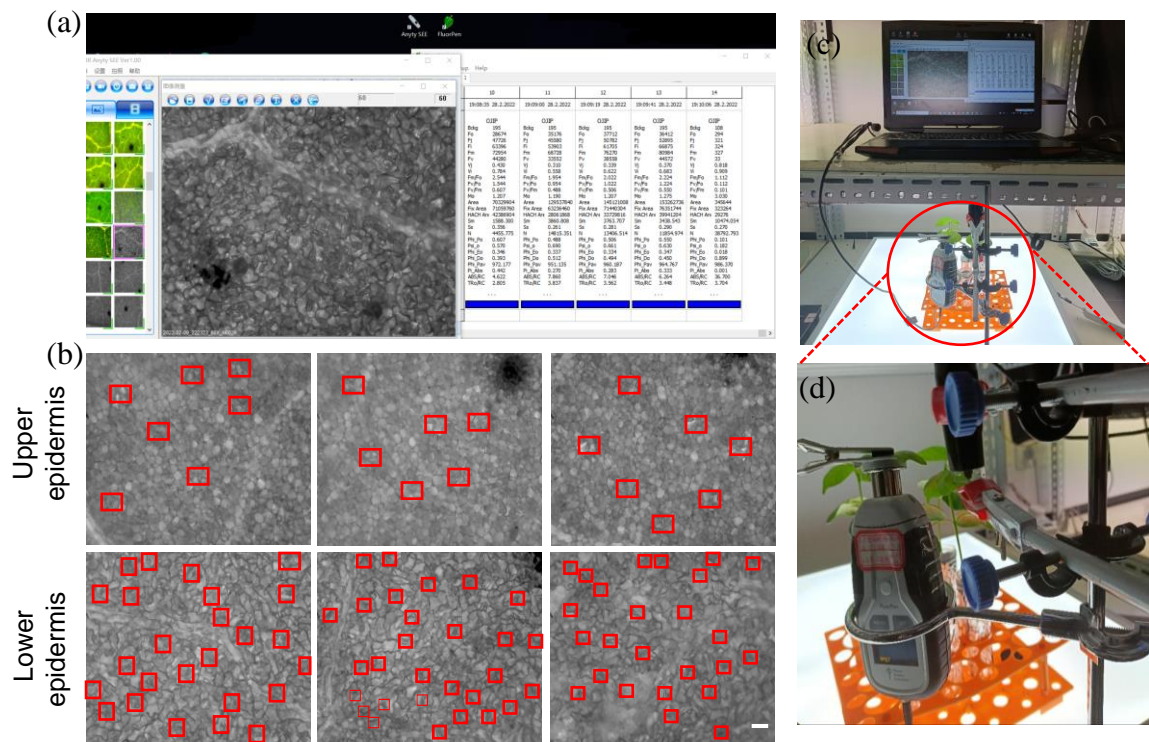


Figure S1. Rapid observation and recording device for cotton leaf stoma under drought stress. (a) The operation interface of the software. (b) Stomatal imaging of the upper and lower epidermis of cotton leaves under normal conditions. (c and d) The representative images of the portable microscope and chlorophyll fluorescence instrument.

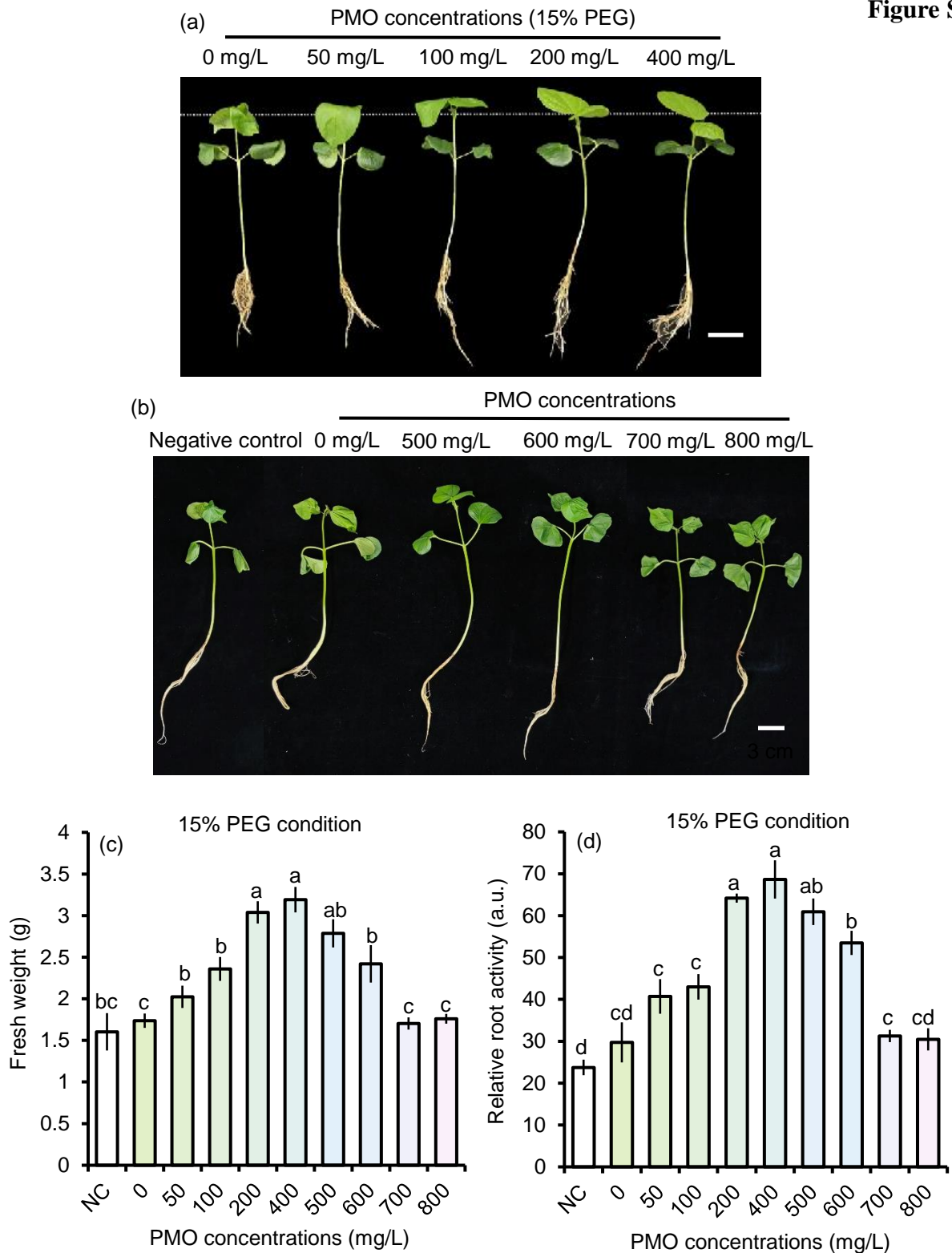


Figure S2. Screening of optimal PMO concentration in improving cotton drought tolerance. (a and b) Phenotypic performance of 0, 50, 100, 200, 400, 500, 600, 700, 800 mg/L PMO and negative control treated cotton seedlings after 24 h of drought stress (15% PEG). (c-d) Fresh weight (c) and relative root activity (d) of cotton seedlings treated with different concentration PMO and negative control (NC) under drought stress. Mean \pm SE ($n = 8$). Different lowercase letters represent significance at 0.05 level. The data were analyzed by one-way ANOVA based on Duncan's multiple range test (two-tailed).

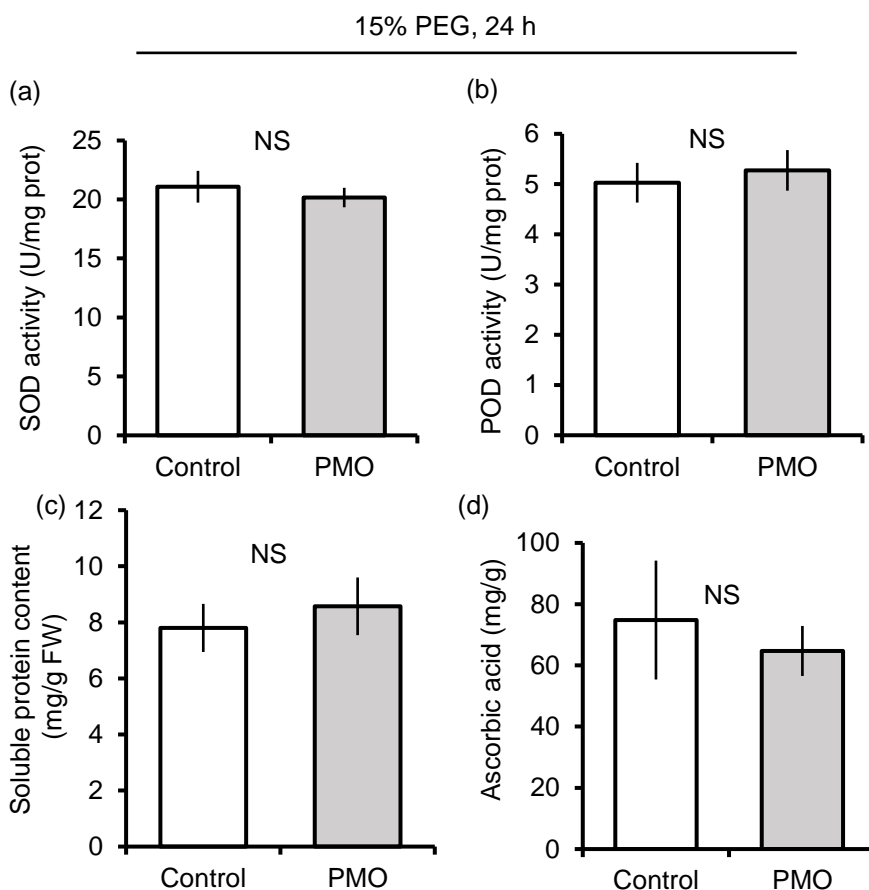


Figure S3. PMO regulates antioxidant enzyme and non-enzymatic systems activities and osmolyte content in cotton under 15% PEG. (a and b) SOD and POD activities of cotton leaves treated with 200 mg/L PMO or 10 mM TES buffer under drought stress. (c) The content of soluble protein content of leaves from drought stressed cotton seedlings treated with 200 mg/L PMO or 10 mM TES buffer. (d) The ascorbic acid content in cotton leaves treated with 200 mg/L PMO or 10 mM TES buffer under drought stress. Mean \pm SE ($n = 8$). *, $P < 0.05$. NS means no significant difference. The data were analyzed by independent samples t-test (two-tailed).

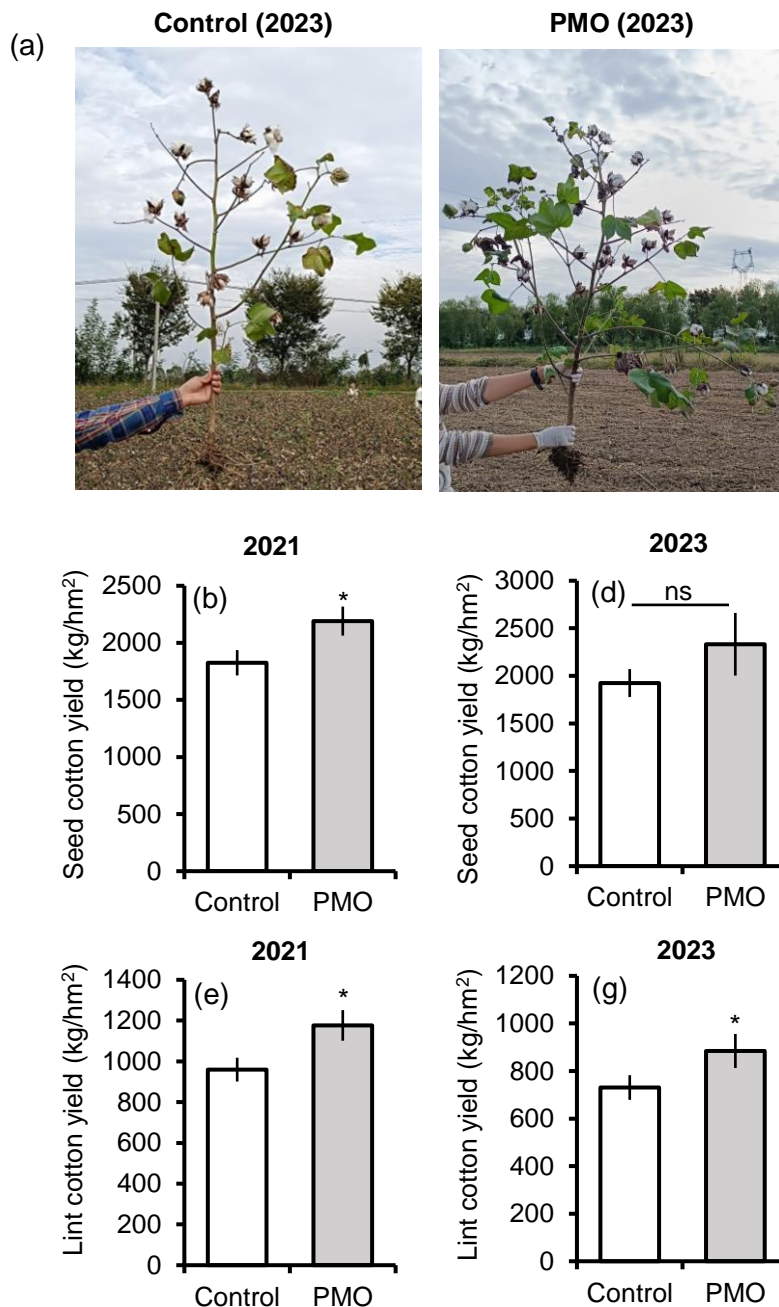


Figure S4. PMO improve cotton yield under field drought condition. (a) Phenotypic performance of 50 mg/L PMO and control buffer treated cotton plants under field drought condition. (b-d) The seed cotton yield of PMO or control buffer-treated cotton plants under field drought conditions in 2021 and 2023. (e-g) The lint cotton yield of PMO or control buffer-treated cotton plants under field drought conditions in 2021 and 2023. Mean \pm SE ($n = 3$). *, $P < 0.05$. The data were analyzed by independent samples t-test (two-tailed), ns means no significant difference.

Note: In 2022, the heat wave exerted a super high temperature stress in field cotton plants. The field trial was failed.

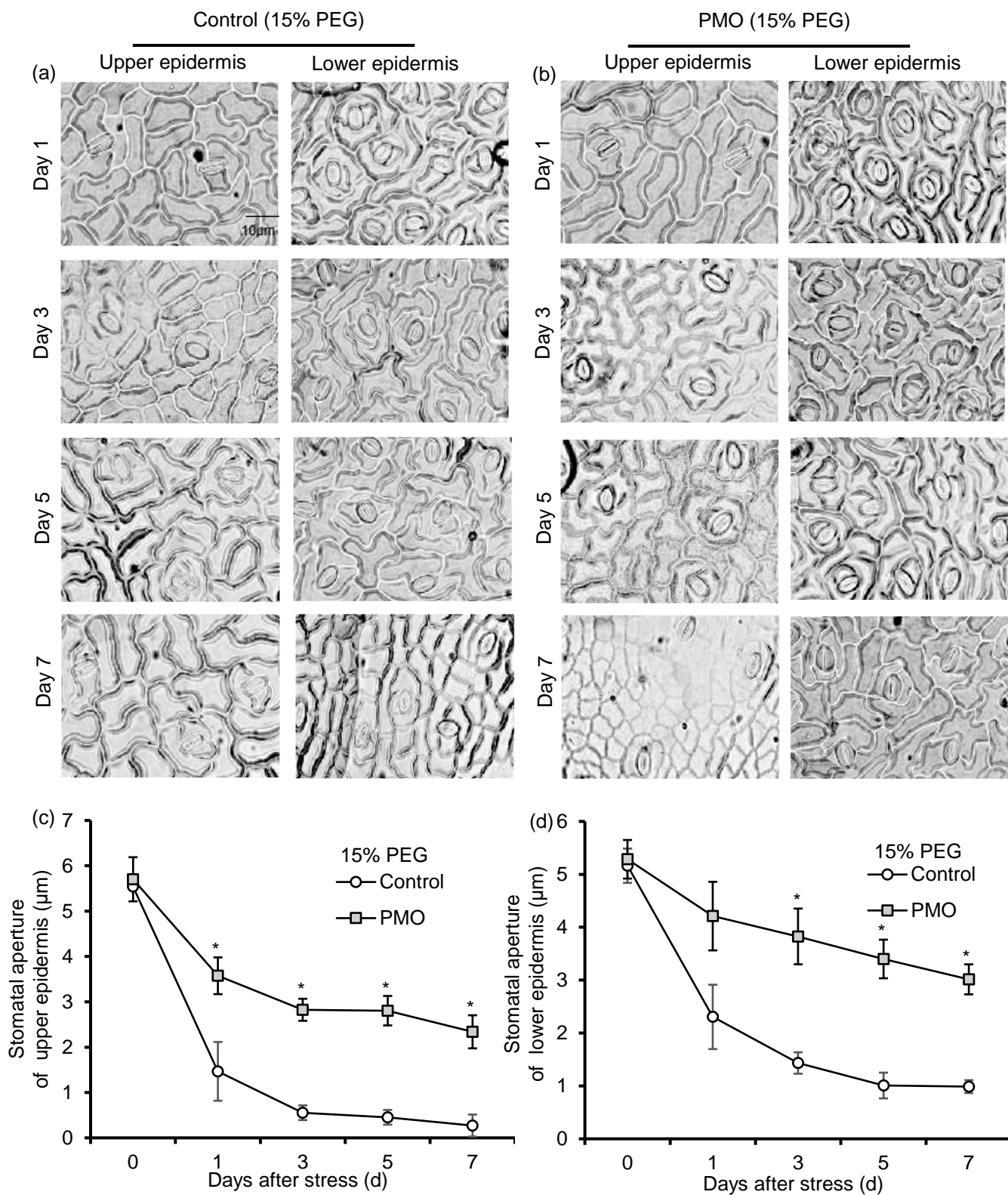


Figure S5. Long-term (7 days) observation on stomata aperture of cotton leaf under drought stress. (a and b) Stomatal imaging of upper and lower epidermis of cotton leaves treated with 200 mg/L PMO or 10 mM TES buffer under drought stress. (c and d) The calculation of stomatal aperture of leaf under drought stress. Mean \pm SE ($n = 4$). *, $P < 0.05$. The data were analyzed by independent samples t-test (two-tailed).