

Supporting information

Hydroxyl radical scavenging by cerium oxide nanoparticles improves *Arabidopsis* salinity tolerance by enhancing leaf mesophyll potassium retention

Honghong Wu¹, Lana Shabala², Sergey Shabala², Juan Pablo Giraldo^{1*}

¹ Department of Botany and Plant Sciences, University of California, Riverside, CA, U.S. 92521

² School of Land and Food, University of Tasmania, Hobart, Tas, Australia, 7001

* Corresponding author: juanpablo.giraldo@ucr.edu, +1 9518273583

Fig. S1

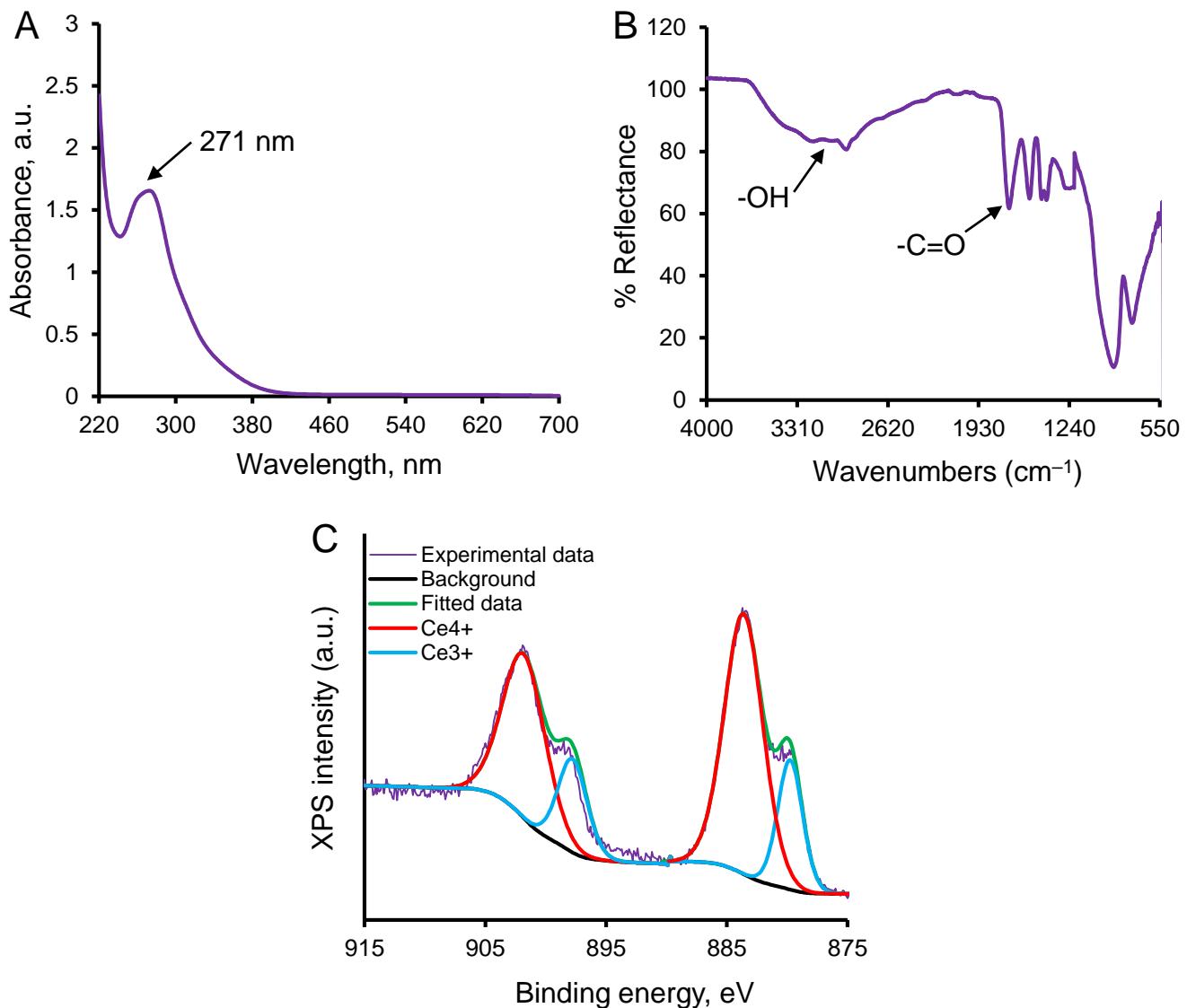


Fig. S1 PNC absorbance and Fourier Transform Infrared (FTIR) spectra, and X-ray photoelectron spectroscopy (XPS). **(A)** PNC has a peak of absorbance at 271 nm. **(B)** FTIR experiment confirmed the presence of --COOH group in PNC. **(C)** XPS analysis demonstrated the PNC has low $\text{Ce}^{3+}/\text{Ce}^{4+}$ ratio ($35 \pm 2.2\%$).

Fig. S2

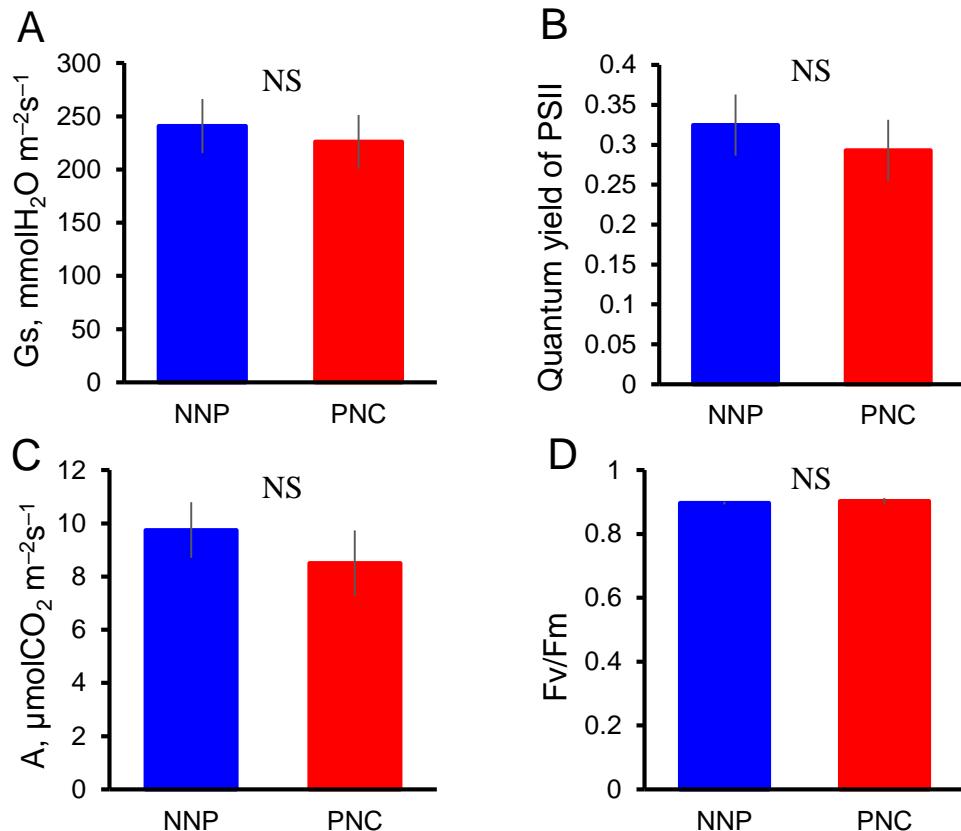


Fig. S2 Effect of nanoceria on leaf gas exchange and photosynthetic parameters. No significant differences were observed in stomatal conductance (G_s) (**A**), quantum yield (**B**), carbon assimilation rate (A) (**C**), and maximum PSII efficiency (F_v/F_m) (**D**) between NNP-leaves and PNC-leaves under control conditions. Mean \pm SE (n = 8). NS, no significant difference.

Fig. S3

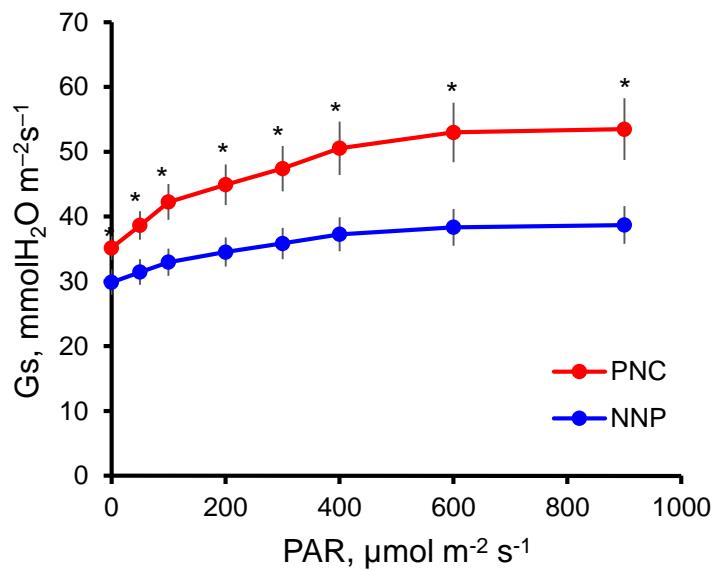


Fig. S3 Higher stomatal conductance in leaves infiltrated with nanoceria relative to controls in plants under salt stress (100 mM NaCl, 3 days). Stomatal conductance (Gs) was measured with a gas exchange analyzer (GFS-3000, Walz) in leaves infiltrated with PNC (PNC-Leaves) and buffer with no nanoparticles (NNP-Leaves) after 3 days of exposure to 100 mM NaCl. Mean \pm SE ($n = 15-16$). *, $P < 0.05$.

Fig. S4

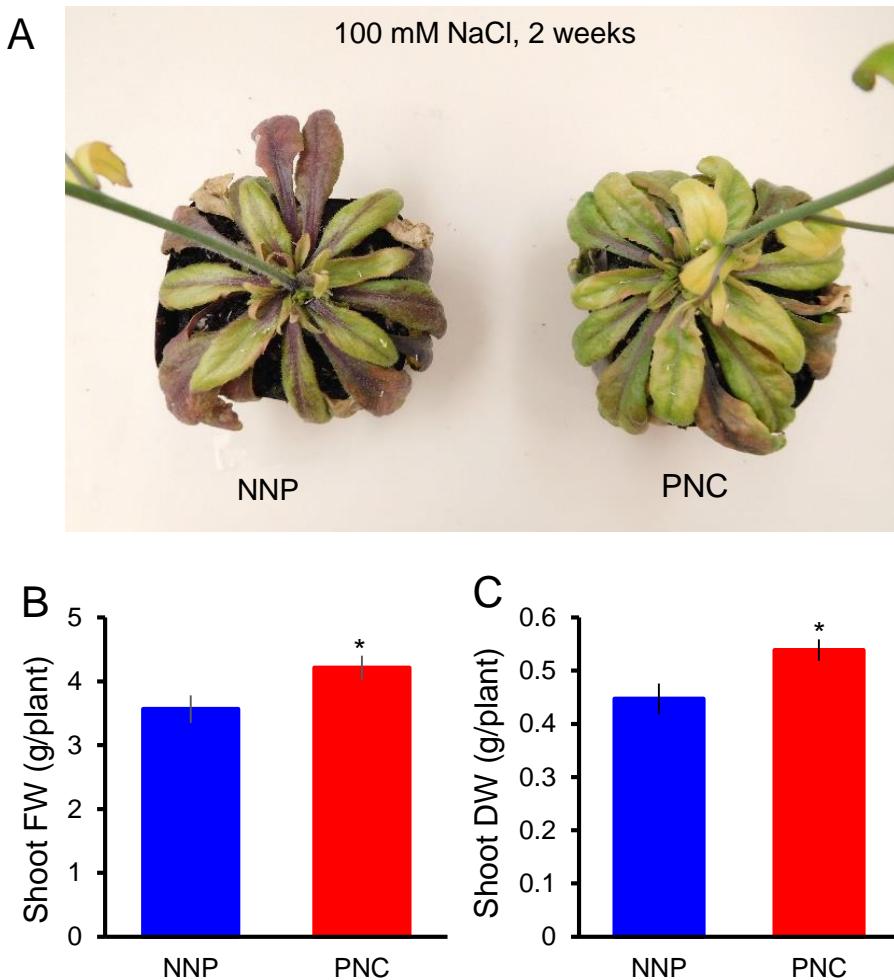


Fig. S4 Nanoceria improve *Arabidopsis* long term salt stress tolerance (100 mM NaCl, 2 weeks). **(A)** PNC-Leaves in comparison with NNP-Leaves under long term salt stress. **(B)** and **C**) Plants infiltrated with PNC have significantly higher shoot fresh weight (FW) and dry weight (DW) than those infiltrated with buffer without nanoparticles. Mean \pm SE ($n = 8$). *, $P < 0.05$.

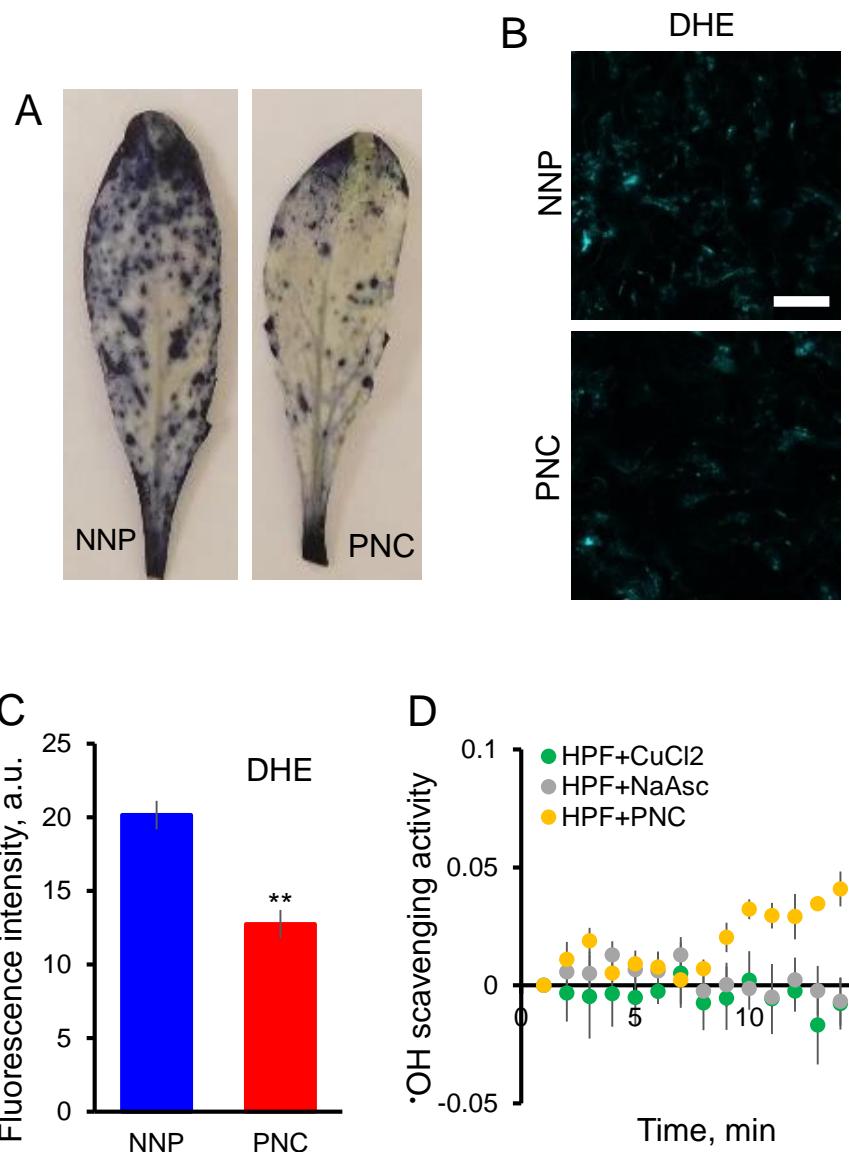


Fig. S5 Catalytic scavenging of ROS by nanoceria in *Arabidopsis* leaf mesophyll cells under salinity stress (100 mM NaCl, 3 days). **(A)** Histochemical staining with NBT (superoxide anion, blue spots) of leaves infiltrated with PNC (PNC-Leaves) and buffer with no nanoparticles (NNP-Leaves) under salt stress. **(B)** Confocal images of leaf spongy mesophyll cells showing the intensity of superoxide anion (DHE). **(C)** Comparison of DHE dye intensity between PNC-Leaves and NNP-Leaves under salt stress. Mean \pm SE ($n = 3$). Scar bar represents 40 μ m. **, $P < 0.01$. **(D)** The background signal of HPF dye in the presence of PNC, CuCl₂, and NaAsc separately. Mean \pm SE ($n = 3-6$).

Table S1

Table S1 Primers used.

Target genes	Primer sequences
<i>At-Gork</i>	F: 5'-TTGCGTGAATTACAAGAGGA-3' R: 5'-TGCTTTCTACTACGCTCTTC-3'
<i>At-HAK5</i>	F: 5'-AAGAGGAACCAAATGCTGAGACA-3' R: 5'-GCCCGATGAAGGGACAT-3'
<i>At-NHX1</i>	F: 5'-CAGCACAGTGGTGTGTTGGTATGCTG-3' R: 5'-GTATGGATTTGGGGTGTGTCATCA-3'
<i>At-Aha1</i>	F: 5'-GACATTGAAAGTTGCCATTGGTAT-3' R: 5'-CTTCCTCCAATACCGTAATCTTCTTGGTT-3'
<i>At-Avp1</i>	F: 5'-GGAACACCACTGCTGCTATTGGAA-3' R: 5'-GGGTCAAAACATCTACGGTGTGGAT-3'
<i>At-GAPDH-A</i>	F: 5'-TGGTTGATCTCGTTGTGCAGGTCTC-3' R: 5'-GTCAGCCAAGTCAACAACTCTCTG-3'
<i>At-Actin2</i>	F: 5'-ACCTTGCTGGACGTGACCTTACTGAT-3' R: 5'-GTTGTCTCGTGGATTCCAGCAGCTT-3'