

Supporting information for

A synthetic bacterial community derived from a desert rhizosphere confers salt stress resilience to tomato in the presence of a soil microbiome

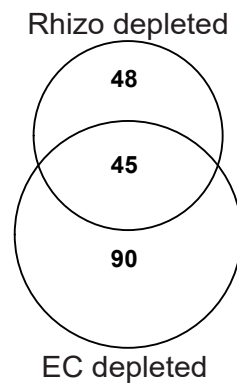
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Figures

Fig. S1: Former agricultural areas in the Jizan desert, which suffer from high salinity, are scarcely populated with indigofera.



Fig. S2: Venn diagram showing number of depleted OTUs in the Rhizo and EC of *I. argentea* in comparison to the Soil. See Dataset S2.



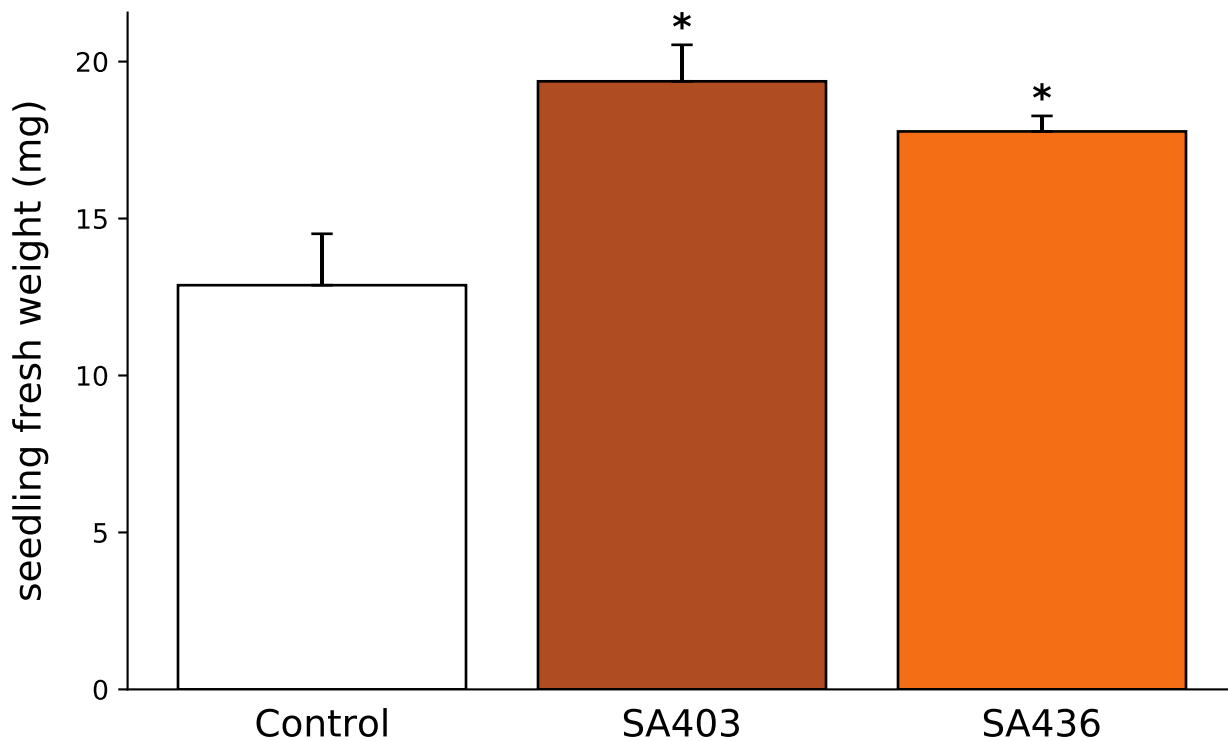
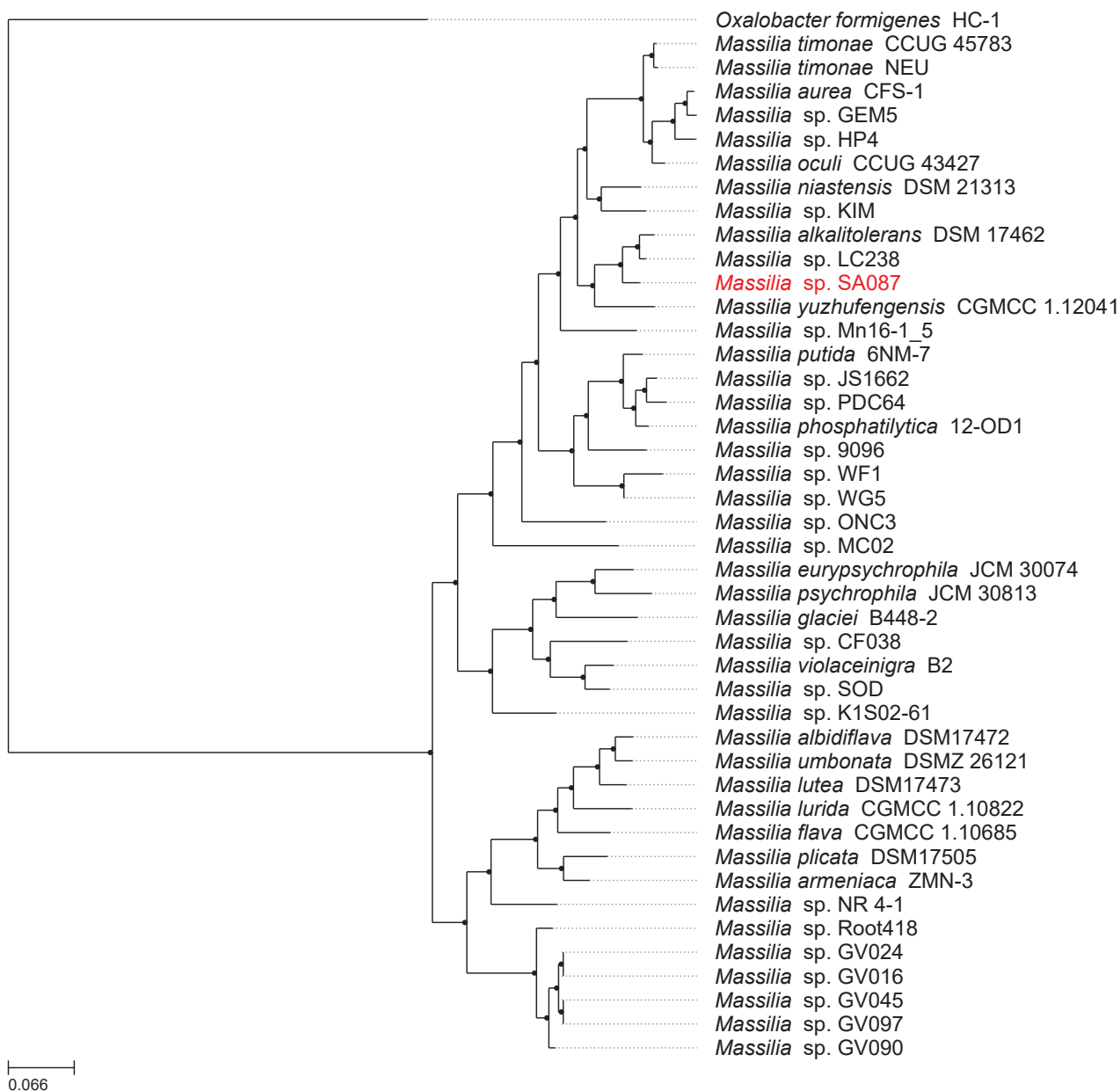


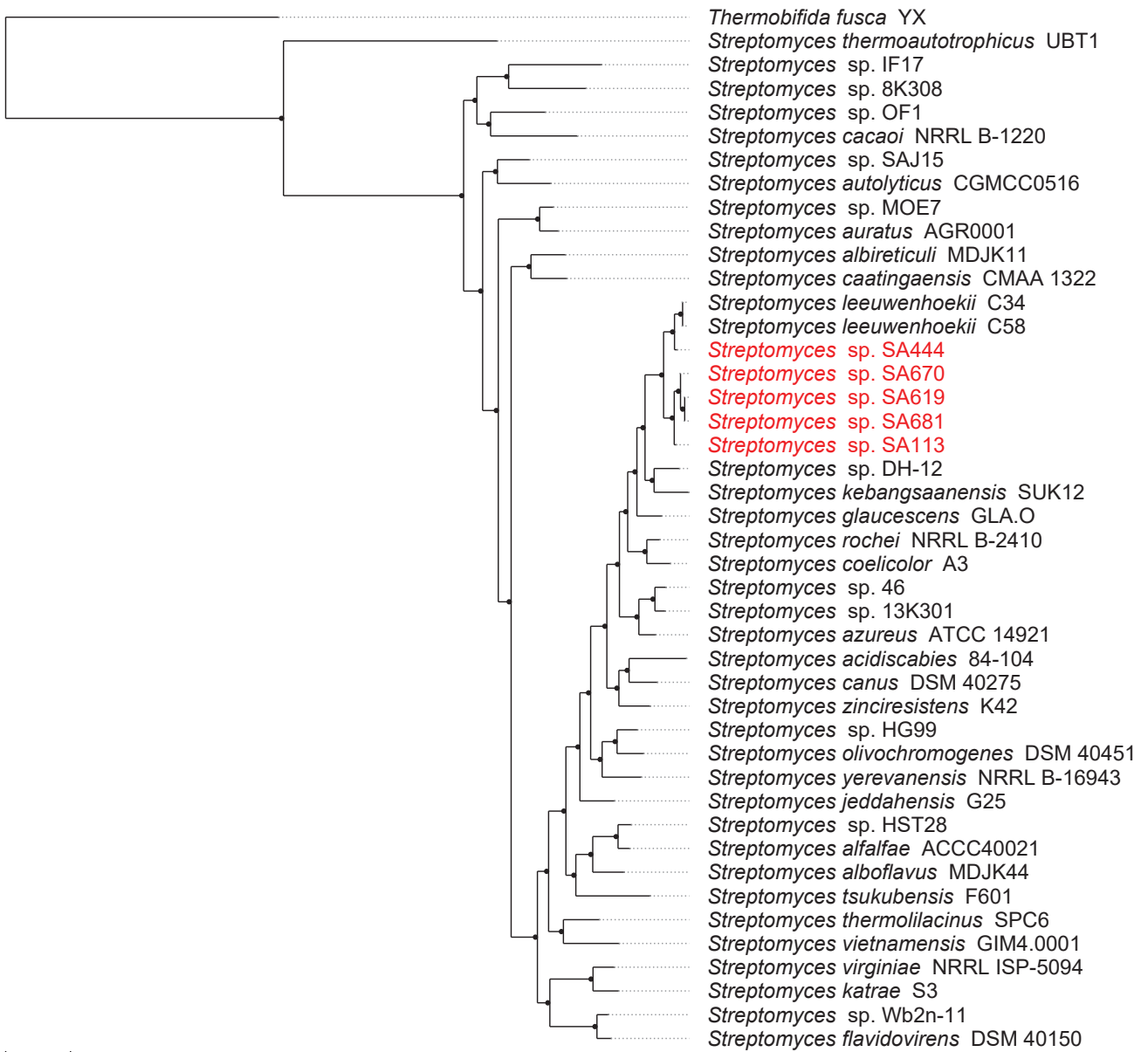
Fig. S3: Growth promotion of arabidopsis by the Jizan strains *Ensifer* sp. SA403 and *Bacillus* sp. SA436. Ten arabidopsis seeds were grown on a plate which was the experimental unit. There were four experimental units per inoculation which also included a sterile control. Seedling fresh weight was defined as the average weight per experimental unit and determined eleven days post bacterial inoculation. The y-axis shows the average seedling fresh weights with the standard deviation as error bars. An asterisk is a significant difference ($p < 0.05$) from the control per Dunnett's test. Strains SA436 and SA403 were grown in 5 mL 1/10th TSB or 5 mL YEM liquid medium, respectively, for 48 hours at 28°C. Bacterial cells were washed and resuspended in 10 mM MgSO₄ and the final suspension had an OD₆₀₀ of 1.0 (~10⁹ cfu/mL). Seeds of *Arabidopsis thaliana* Columbia-0 were surface sterilized by washing with ethanol and soaking in 1/4 strength commercial bleach for 10 minutes. Seeds were transferred to wet filter paper in Petri dishes. After incubating at 4°C for 3 days, 10 seeds were sown on plates containing 50 mL 1/2th Murashige Skoog medium. Two µL of the bacterial suspension was applied to the root tips of one-week-old seedlings. Control plants were inoculated with 2 µL of 10 mM MgSO₄.

Fig. S4: Maximum likelihood trees based on the AMPHORA gene alignments of nine different genera. (a) *Massilia*, (b) *Streptomyces*, (c) *Ensifer*, (d) *Bacillus*, (e) *Enterobacter*, (f) *Acinetobacter*, (g) *Ochrobactrum*, (h) *Pseudomonas*, (i) *Ralstonia*. Each tree contains at least one of the strains found in the Jizan SynCom, representative genomes and plant-associated strains for that genus. All trees are rooted on a suitable outgroup and the Jizan strains are colored in red. The distance scale indicates the number of differences between sequences.

a

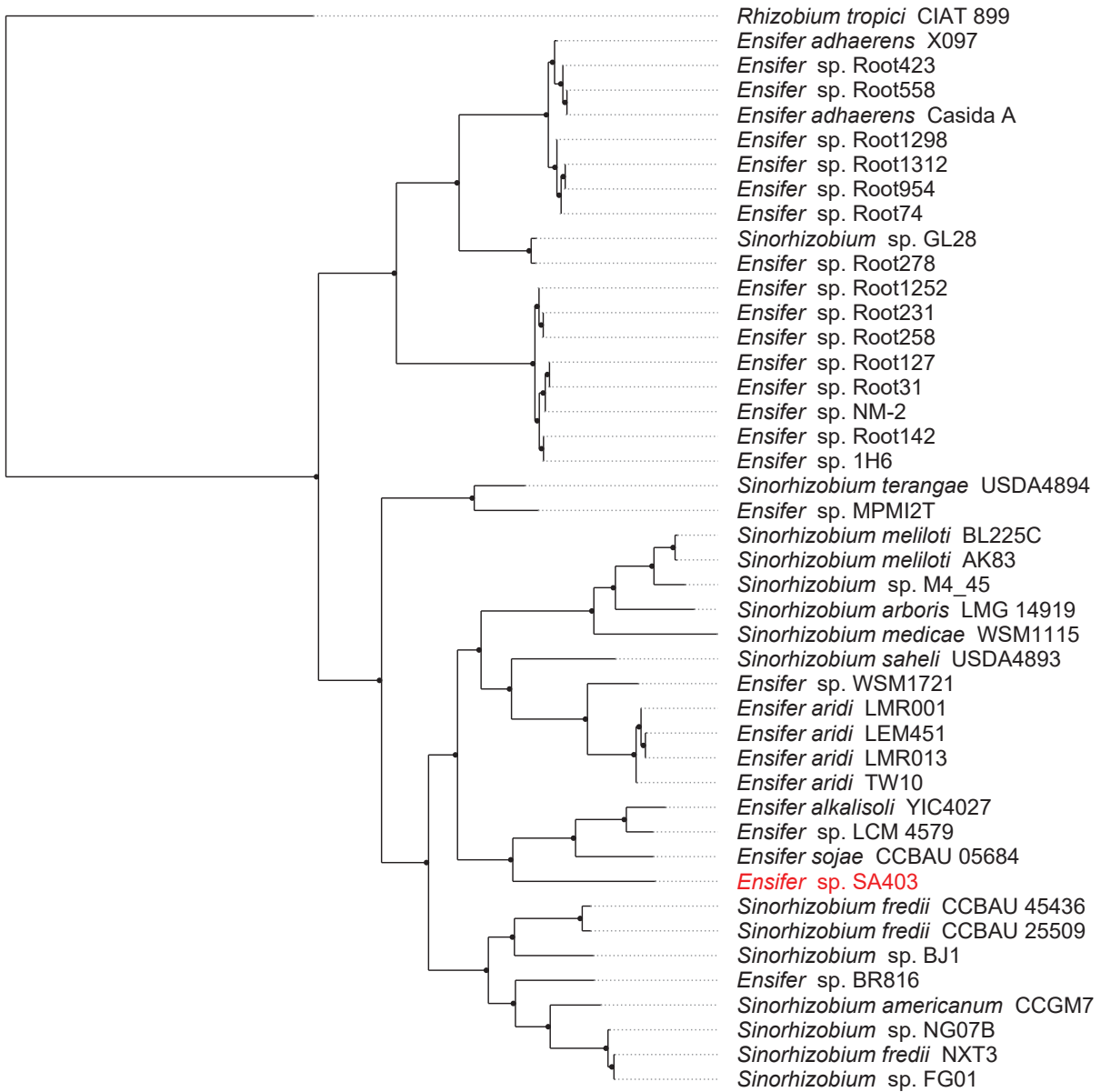


b

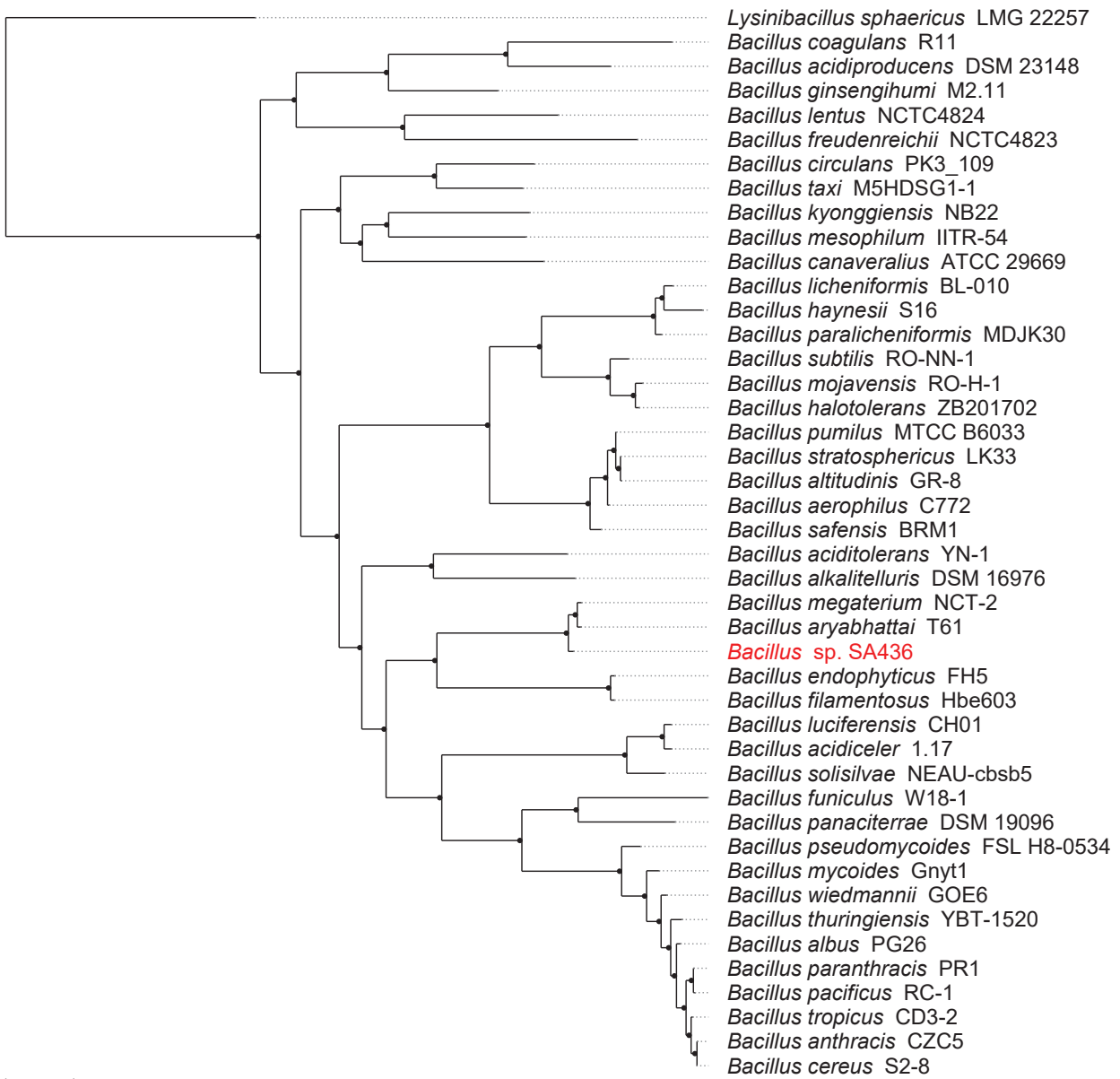


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C

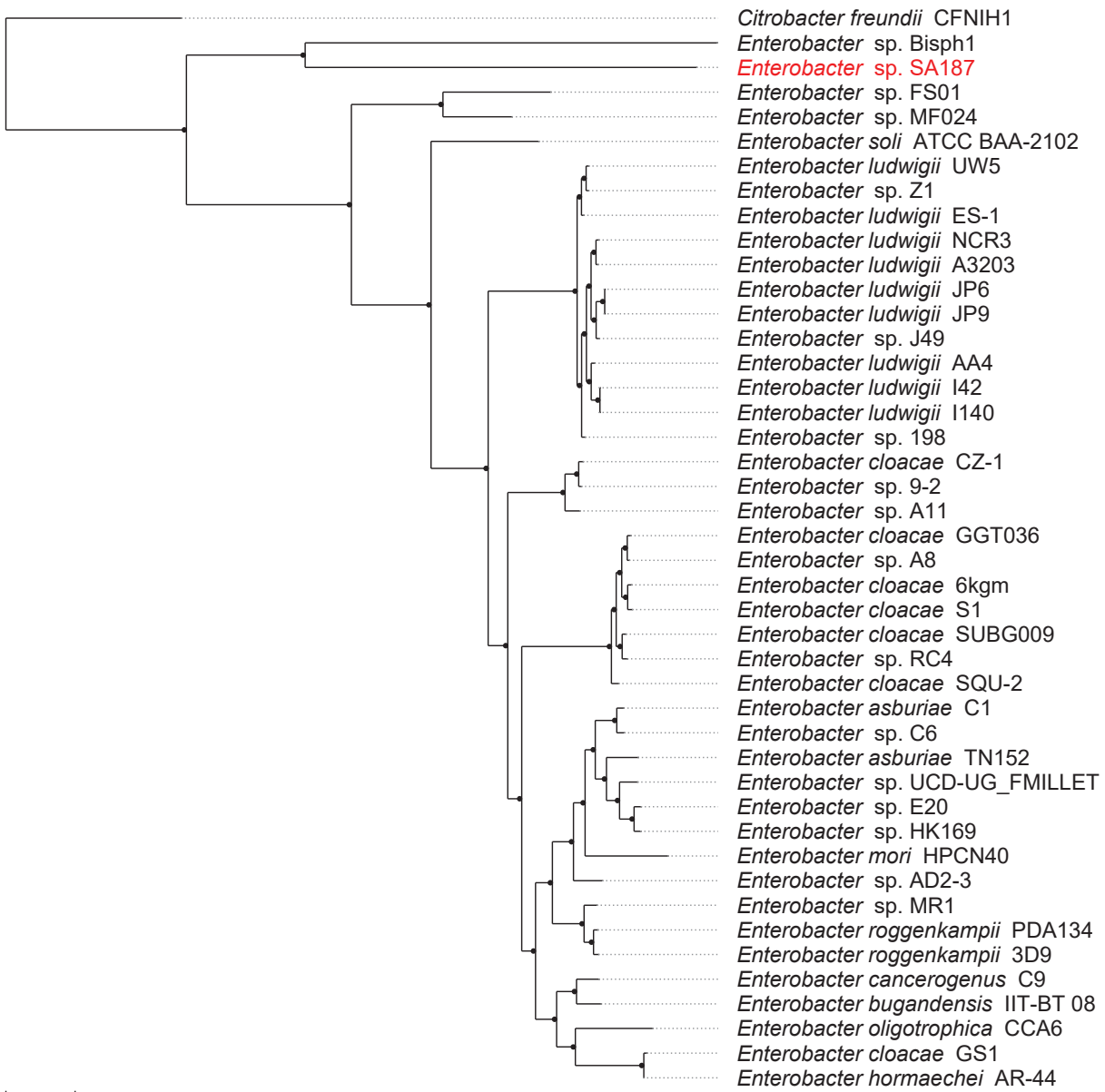


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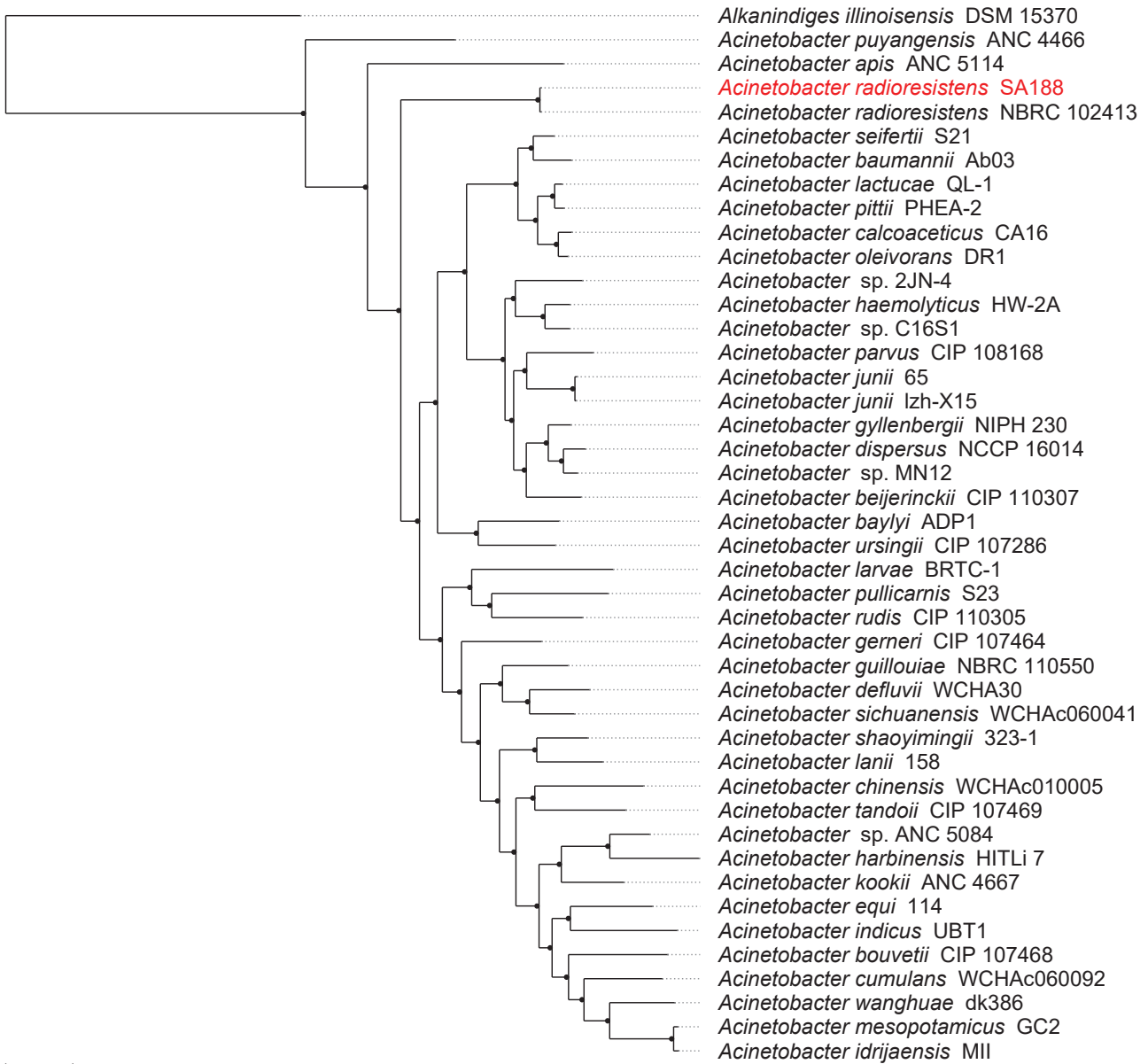
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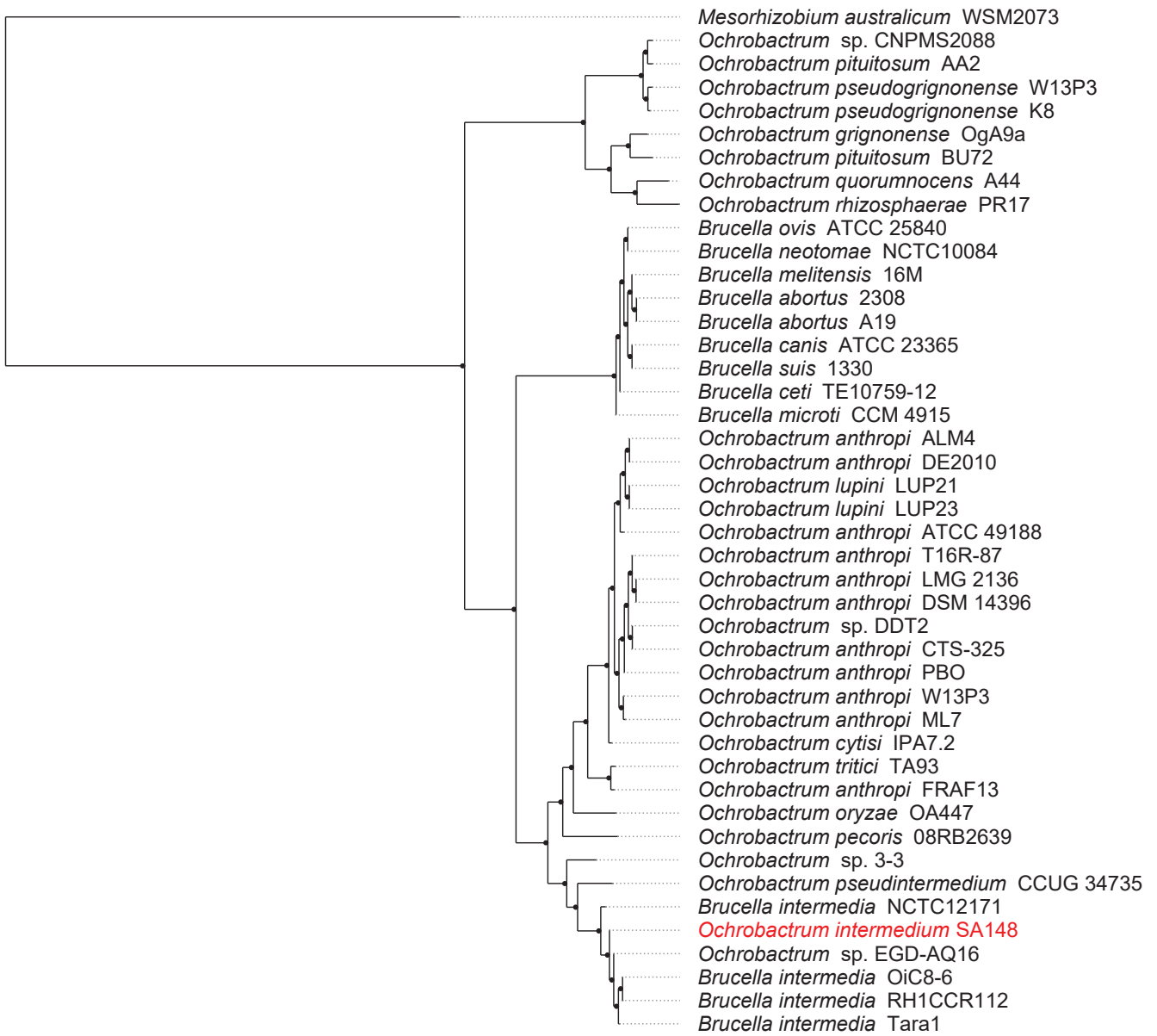
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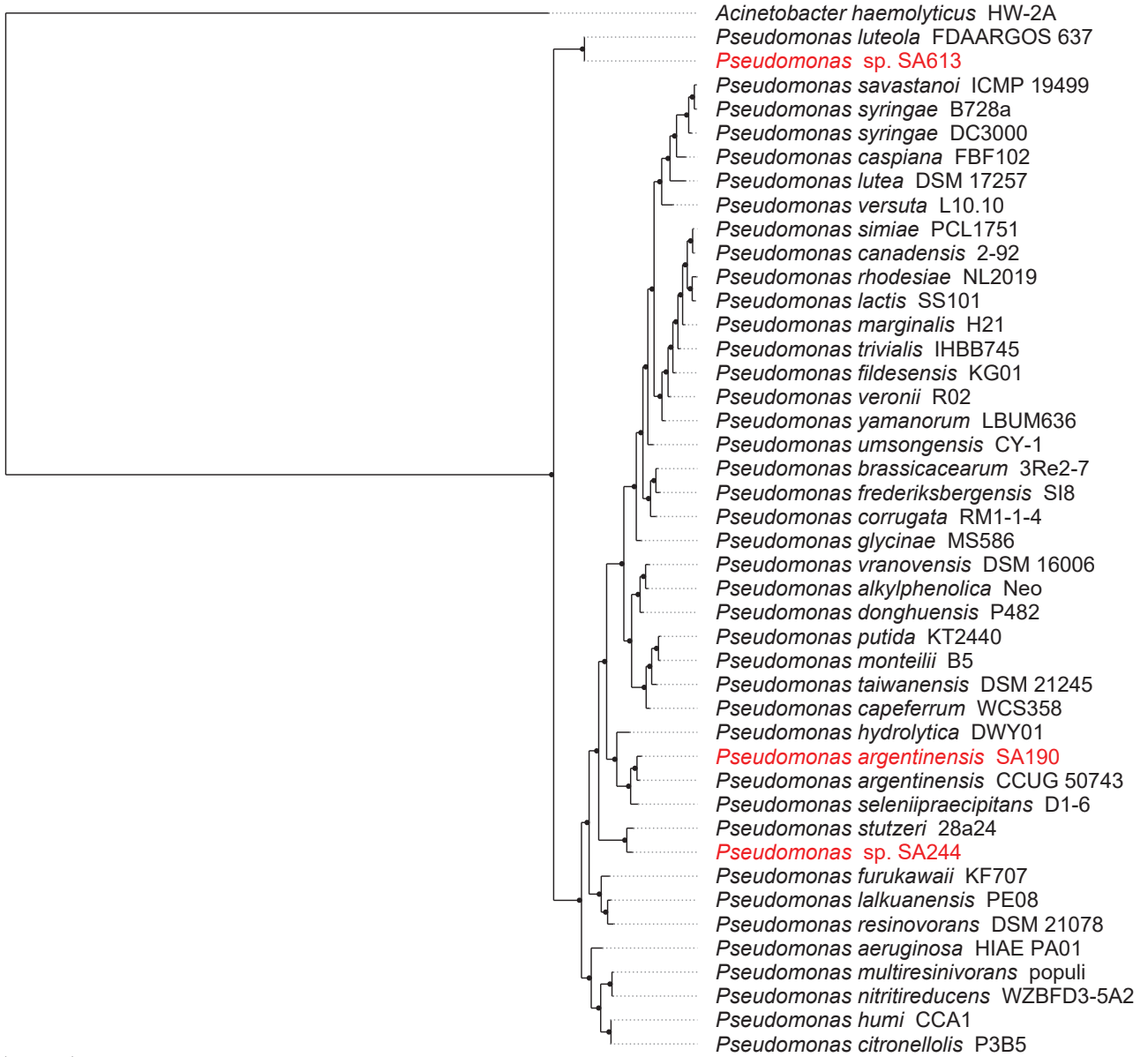
0.015

f





h



0.409

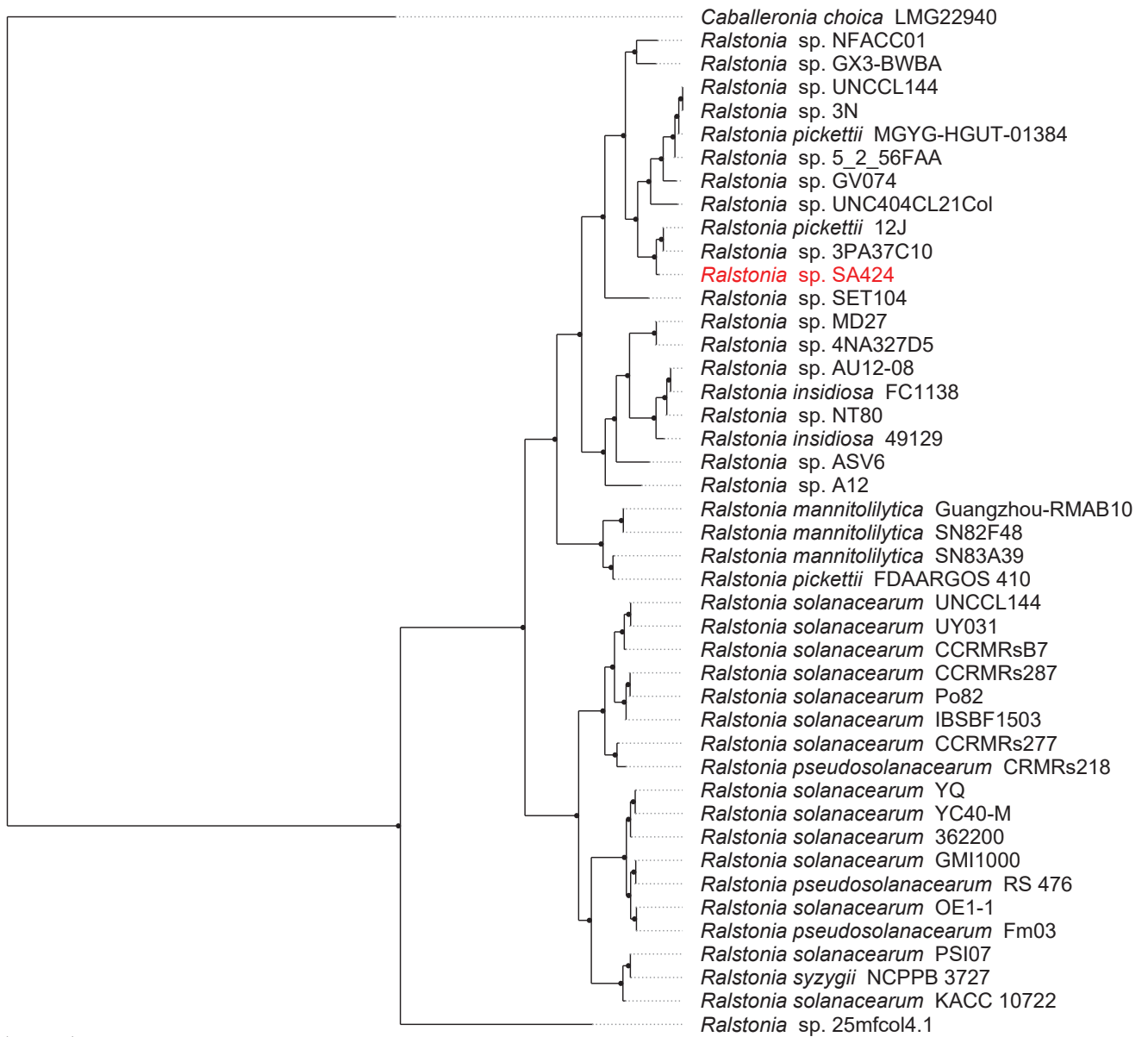


Fig. S5: Tomato salt stress assay. The salt stress assay was adapted from a protocol by Awlia et al., 2016. Pots with plants are soaked in a saline solution for about 30 minutes. As the WHC is always maintained at 60%, there is a certain volume of water already present in the substrate. When the saline solution enters through the bottom of the cups during the soaking, the salt concentration lowers as the solution is diluted by the water present in the substrate. The volume of saline solution that enters the pot is equal to the remaining 40% WHC plus extra air. This extra air volume is a result of the supersaturation by the soaking method which exceeds the amount predicted by 100% WHC. As the WHC returns to 60% due to evaporation and transpiration, the diluted saline solution will concentrate to a target concentration in the substrate.

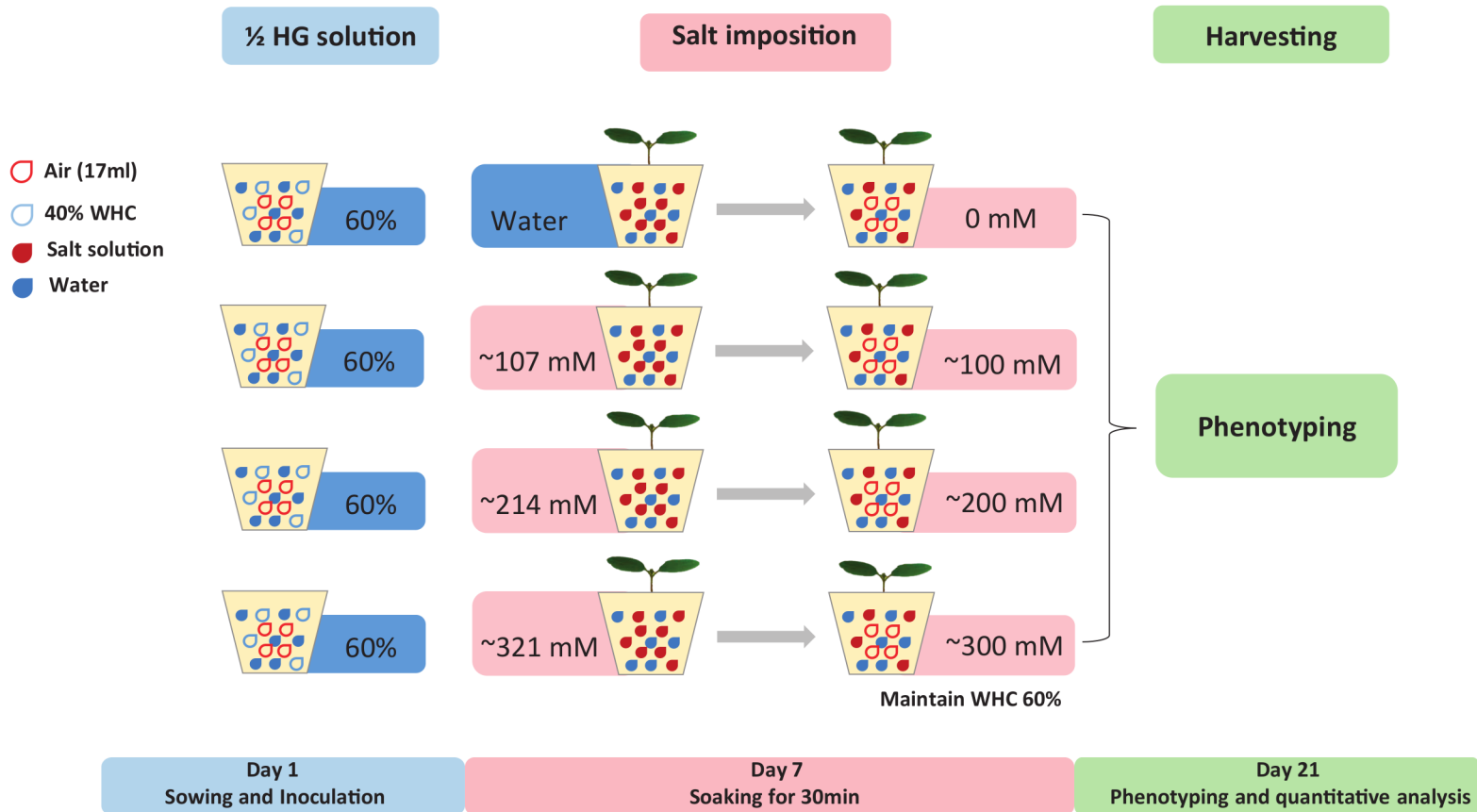
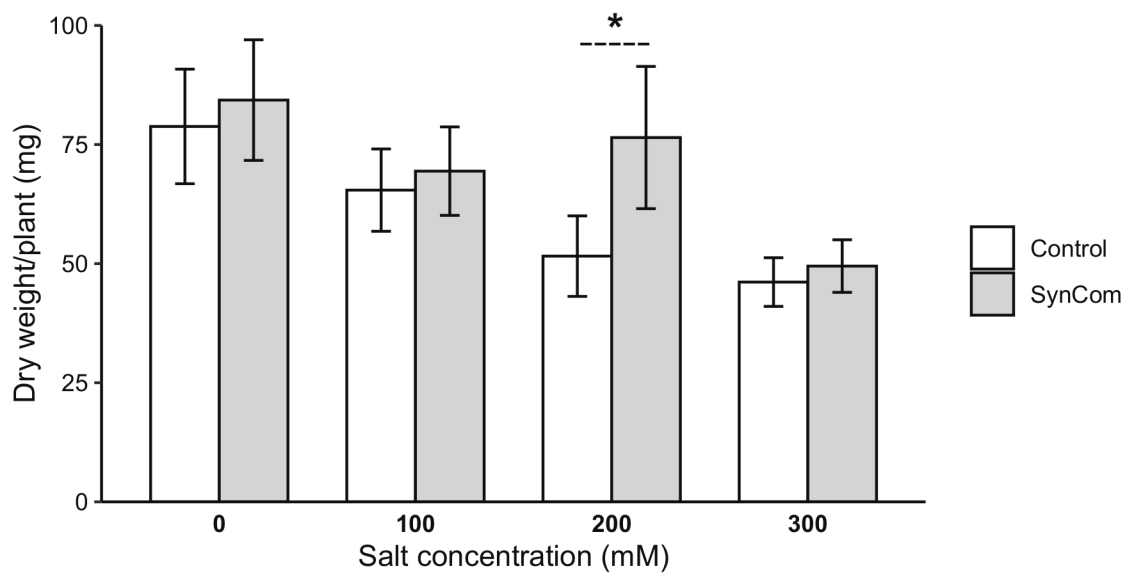
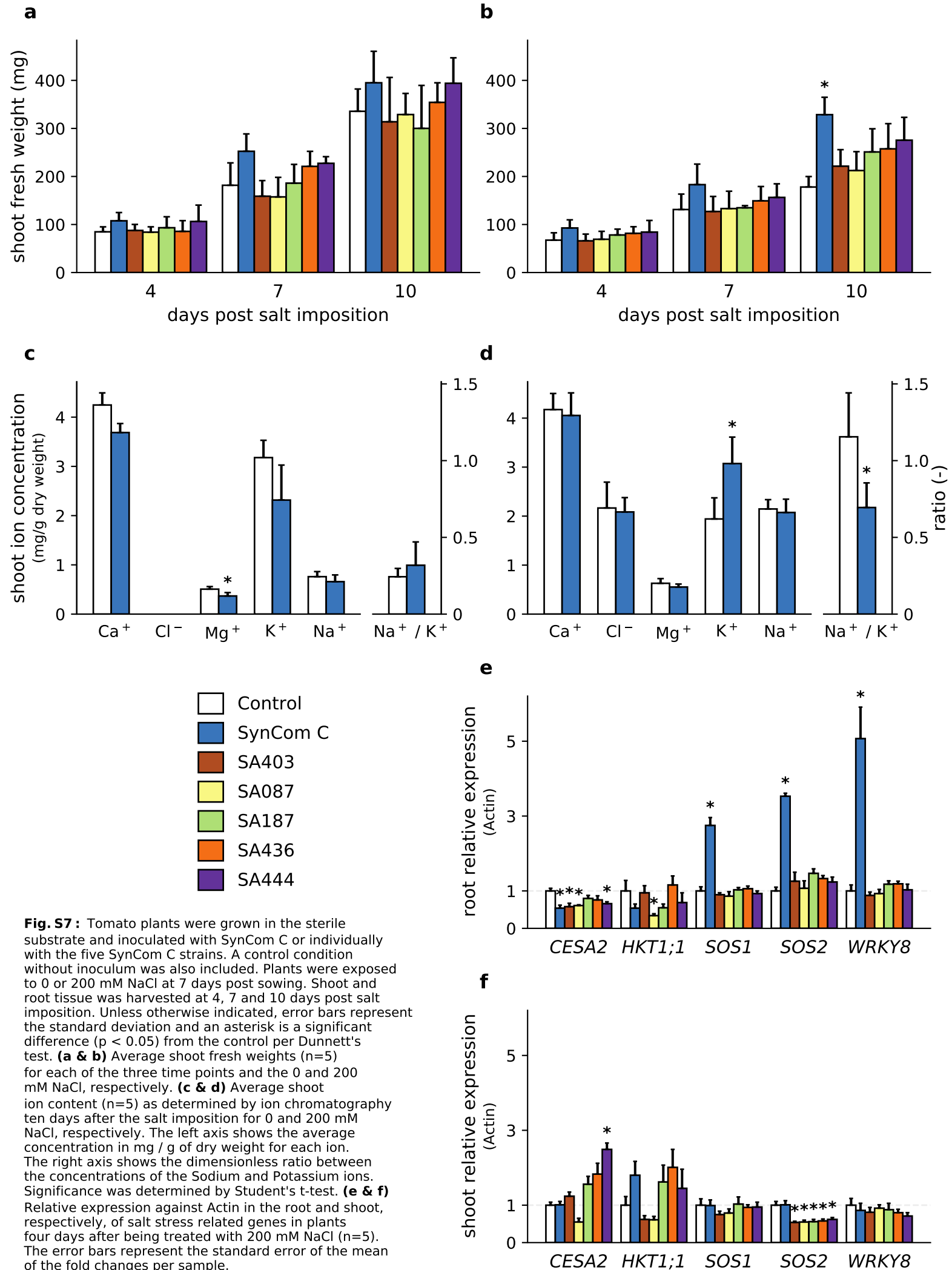


Fig. S6: Salt tolerance test of tomato plants grown with or without SynCom inoculation. Shoot dry weight of 21-day-old tomato plantlets exposed to 0, 100, 200, 300 mM NaCl (for 14 days) and inoculated with or without SynCom C. Plants were grown in the non-sterile substrate. An asterisk represents a statistically significant ($p < 0.05$) difference between the two inoculations per Student's t-test.



0 mM NaCl

200 mM NaCl



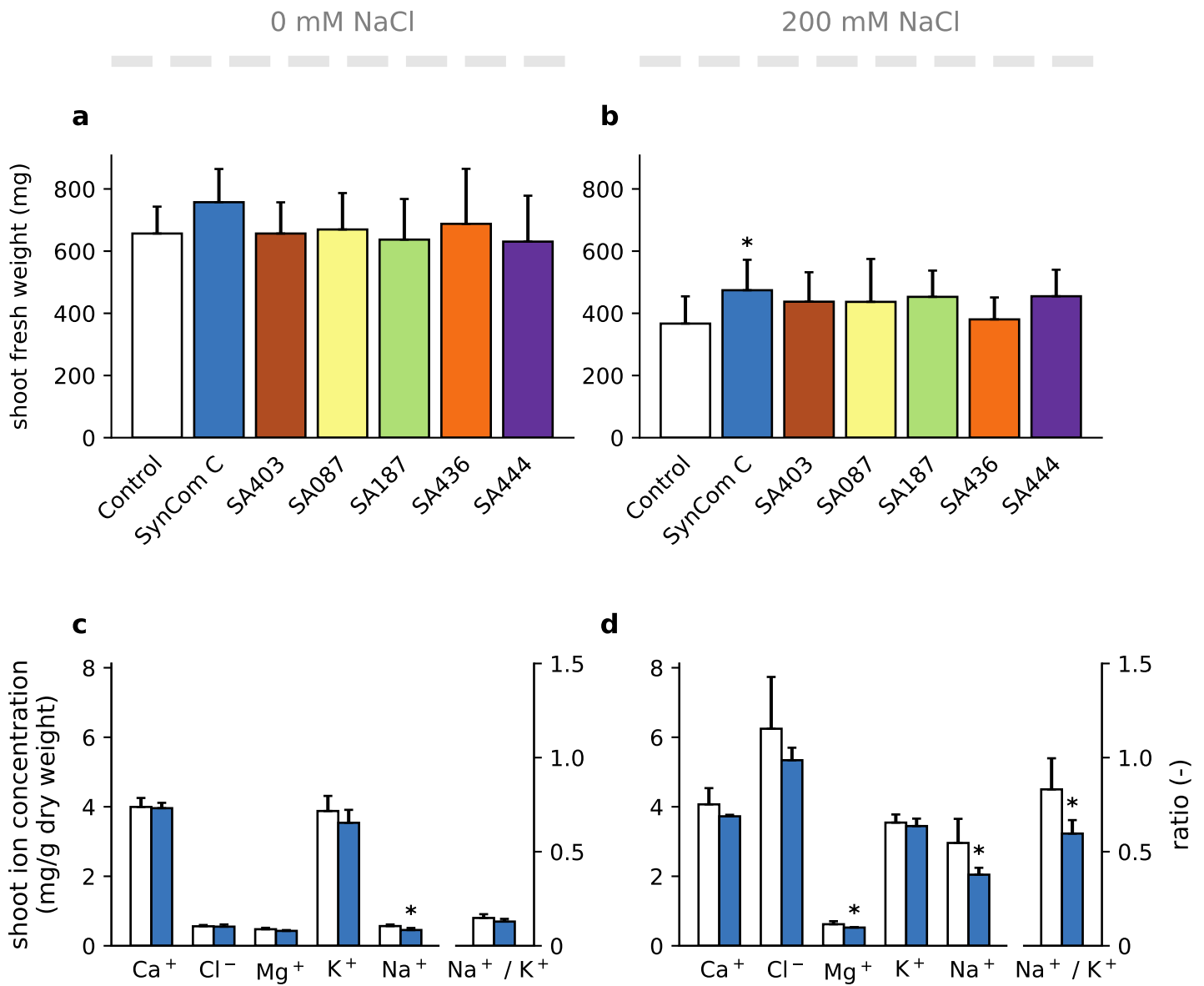
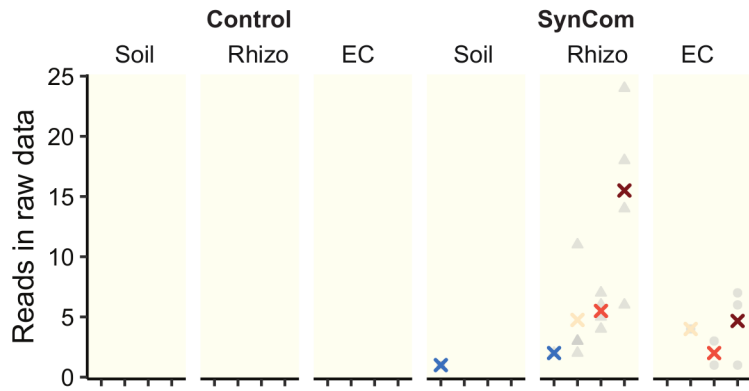


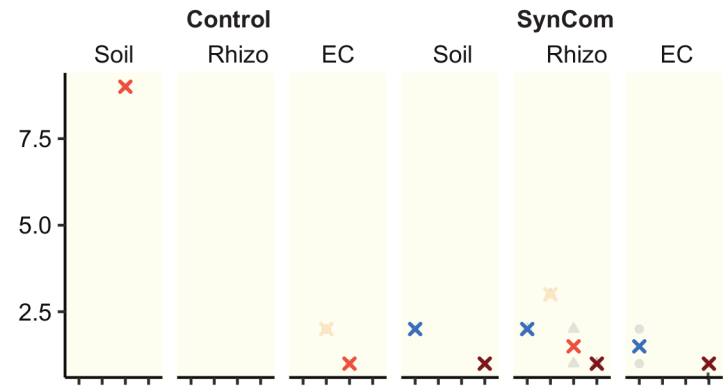
Fig. S8: Tomato plants were grown in the non-sterile substrate and inoculated with SynCom C or individually with the five SynCom C strains. A control condition without inoculum was also included. Plants were exposed to 0 or 200 mM NaCl one week after sowing. Shoot tissue was harvested at ten days post salt imposition for fresh weight and ion content analysis. The standard deviation is shown by the error bars. **(a & b)** Average shoot fresh weights (n=15) for the 0 & 200 mM NaCl treated plants, respectively. An asterisk indicates a significant difference ($p < 0.05$) from the control per Dunnett's test. **(c & d)** Average shoot ion content as determined by ion chromatography for the 0 & 200 mM NaCl salt levels, respectively. Five random plants were selected from the control and SynCom C inoculated plants. The left axis shows the concentration in mg / g of dry weight for each ion. The right axis shows the dimensionless ratio between the concentrations of the Sodium and Potassium ions. An asterisk is a significant difference ($p < 0.05$) per Student's t-test.

Fig. S9: ASVs of Jizan strains which did not pass the detection criteria for the root colonization of tomato.

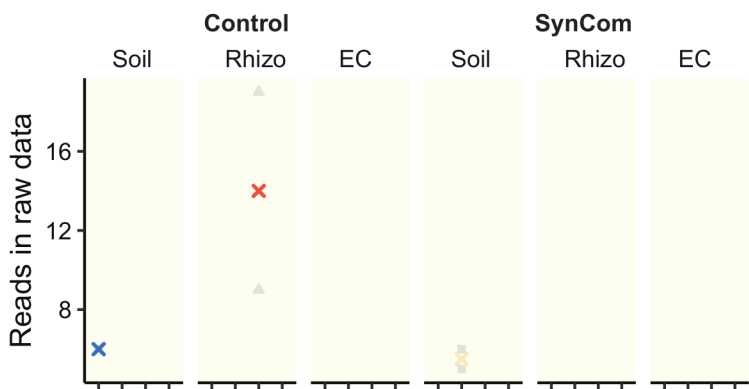
***Ensifer* sp. SA403 (ASV91)**



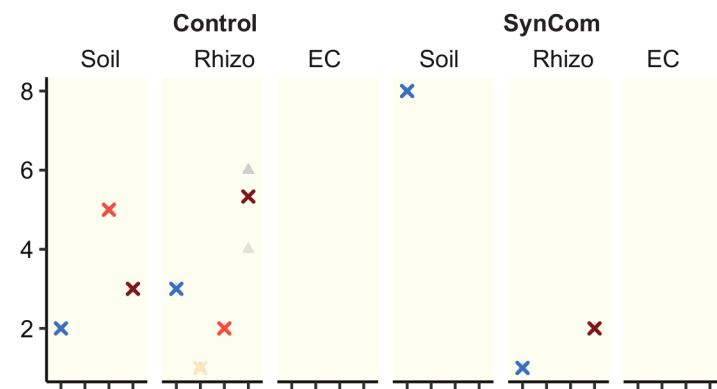
***Acinetobacter* sp. SA188 (ASV10)**



***Streptomyces* sp. SA444 (ASV783)**



***Streptomyces* sp. (ASV4047)**



Salt level (mM) x 0 x 100 x 200 x 300

Fig. S10: Positive correlation between bacterial ASVs and salt concentration. Log₂-transformed normalized read counts of ASVs found in the Rhizo (a) and EC (b) of tomato growing in the non-sterile substrate and inoculated with the Jizan SynCom. The ASVs that were enriched in any of the salt levels and with a positive correlation to the salt gradient are shown here. Crosses indicate mean log₂-transformed normalized read counts of each ASV under each salt level. Blue crosses indicate ASVs matching a SynCom strain detected in this analysis.

