

Supporting information of

Root bacteria recruited by *Phragmites australis* in Constructed Wetlands have the potential to enhance azo-dye phytodepuration

Supplementary Table S1. Distribution of cultivable bacteria isolated from *P. australis* roots and rhizosphere according to the phylogenetic classification at the phylum, genus and species level.

| Phylum | Genus | E (80) | R (72) | Species | E (80) | R (72) |
|-----------------------|-------------------------|--------|--------|------------------------------|--------|--------|
| <i>Actinobacteria</i> | <i>Arthrobacter</i> | | 1 | <i>A. ureafaciens</i> | | 1 |
| | <i>Microbacterium</i> | | 4 | <i>M. kitamiense</i> | | 2 |
| | | | | <i>M. oxydans</i> | | 1 |
| | | | | <i>M. maritipicum</i> | | 1 |
| | <i>Streptomyces</i> | | 5 | <i>S. pluriclorescens</i> | | 2 |
| | | | | <i>S. althioticus</i> | | 1 |
| | | | | <i>S. acidiscabies</i> | | 1 |
| | | | | <i>S. caeruleatus</i> | | 1 |
| <i>Bacteroidetes</i> | <i>Flavobacterium</i> | 3 | 9 | <i>F. gyeonganense</i> | 2 | 3 |
| | | | | <i>F. johnsoniae</i> | 1 | 5 |
| | | | | <i>F. oncorhynchi</i> | | 1 |
| | <i>Fluvicola</i> | | 1 | <i>F. taffensis</i> | | 1 |
| <i>Firmicutes</i> | <i>Bacillus</i> | 38 | 24 | <i>B. aryabhattai</i> | 2 | |
| | | | | <i>B. horneckiae</i> | | 1 |
| | | | | <i>B. litoralis</i> | | 1 |
| | | | | <i>B. marisflavi</i> | | 1 |
| | | | | <i>B. megaterium</i> | | 1 |
| | | | | <i>B. niaci</i> | | 1 |
| | | | | <i>B. oceanisediminis</i> | | 2 |
| | | | | <i>B. pumilus</i> | 34 | |
| | | | | <i>B. thuringiensis</i> | 2 | 17 |
| | <i>Lysinibacillus</i> | | 8 | <i>L. fusiformis</i> | | 6 |
| | | | | <i>L. parviboronicapiens</i> | | 1 |
| | | | | <i>L. varians</i> | | 1 |
| | <i>Viridibacillus</i> | | 9 | <i>V. arenosi</i> | | 9 |
| <i>Proteobacteria</i> | <i>Caulobacter</i> | 1 | | <i>C. segnis</i> | | 1 |
| | <i>Cedecea</i> | | 1 | <i>C. neteri</i> | | 1 |
| | <i>Enterobacter</i> | | 4 | <i>E. cancerogenus</i> | | 2 |
| | | | | <i>E. cloacae</i> | | 1 |
| | | | | <i>E. ludwigii</i> | | 1 |
| | <i>Ochrobactrum</i> | | 1 | <i>O. anthropi</i> | | 1 |
| | <i>Pseudomonas</i> | 24 | 3 | <i>P. aeruginosa</i> | 2 | |
| | | | | <i>P. brassicacearum</i> | 5 | |
| | | | | <i>P. fluorescens</i> | | 1 |
| | | | | <i>P. plecoglossicida</i> | | 1 |
| | | | | <i>P. stutzeri</i> | | 1 |
| | | | | <i>P. thivervalensis</i> | 17 | |
| | <i>Rheinheimera</i> | | 1 | <i>R. chironomi</i> | | 1 |
| | <i>Rhizobium</i> | 6 | | <i>A. tumefaciens</i> | 6 | |
| | <i>Stenotrophomonas</i> | 8 | 1 | <i>S. rhizophila</i> | | 1 |
| | | | | <i>S. chelatiphaga</i> | 8 | |

Supplementary Table S2. Percentage distribution of plant growth promoting activities according to the bacterial genera isolated from the root system of *P. australis*. IAA = 3-indoleacetic acid production, ACC-d = ACC deaminase activity, Protease = protease production, EPS = exopolysaccharides production, SWIMMING =swimming lifestyle, SWARMING = swarming lifestyle.

| Genus | nº strain | IAA | ACC-d | Protease | EPS | Swimming | Swarming |
|-------------------------|-----------|-----|-------|----------|-----|----------|----------|
| <i>Arthrobacter</i> | 1 | 0 | 100 | 100 | 0 | 0 | 0 |
| <i>Microbacterium</i> | 4 | 50 | 100 | 25 | 25 | 0 | 0 |
| <i>Streptomyces</i> | 4 | 50 | 50 | 0 | 0 | 0 | 0 |
| <i>Flavobacterium</i> | 10 | 20 | 50 | 20 | 20 | 0 | 0 |
| <i>Fluviicola</i> | 1 | 0 | 0 | 0 | 0 | 100 | 100 |
| <i>Bacillus</i> | 12 | 83 | 58 | 75 | 8 | 17 | 33 |
| <i>Lysinibacillus</i> | 7 | 100 | 57 | 57 | 0 | 86 | 86 |
| <i>Viridibacillus</i> | 2 | 100 | 100 | 0 | 0 | 100 | 100 |
| <i>Caulobacter</i> | 1 | 100 | 100 | 0 | 0 | 0 | 0 |
| <i>Cedecea</i> | 1 | 100 | 100 | 0 | 0 | 0 | 0 |
| <i>Enterobacter</i> | 4 | 100 | 100 | 0 | 0 | 75 | 25 |
| <i>Ochrobactrum</i> | 1 | 100 | 100 | 0 | 0 | 0 | 0 |
| <i>Pseudomonas</i> | 6 | 100 | 83 | 50 | 50 | 17 | 0 |
| <i>Rheinheimera</i> | 1 | 100 | 100 | 100 | 0 | 0 | 0 |
| <i>Stenotrophomonas</i> | 3 | 100 | 33 | 100 | 33 | 0 | 0 |

Supplementary Table S3. Metal-tolerance phenotypes. The code of the isolates is simplified and includes only the fraction of origin (R = rhizosphere; E = endosphere) and the progressive number. The percentage in the last column is referred to the strains belonging to each detected phenotype. '+': refers to the strain tolerance to the metal at the indicated concentration.

| Phenotype | Code of the isolates | Metal Tolerance | | | | | | | | | |
|-----------|--|------------------------|---|---|------------------------|---|---|------------------------|-----|---|---------------|
| | | ZnCl ₂ (mM) | | | NiCl ₂ (mM) | | | CdCl ₂ (mM) | | | |
| | | 0.5 | 1 | 2 | 0.5 | 1 | 2 | 0.05 | 0.5 | 1 | % of isolates |
| 1 | R8, R25, R31, R32, R34, R67 | + | + | + | + | + | + | + | + | + | 12 |
| 2 | R17, R20, R78 | + | + | + | + | + | + | + | + | + | 6 |
| 3 | R3, R6, R50 | + | + | + | + | + | + | + | + | + | 6 |
| 4 | R15 | + | + | + | + | + | + | + | + | + | 2 |
| 5 | R22, R68 | + | + | + | + | + | + | + | + | + | 4 |
| 6 | E2, E6, E8, E14, E16, E21, E28, E33, E73, R4, R28, R65, R71, R75 | + | + | + | + | + | + | + | + | + | 28 |
| 7 | E13, R76, R79 | + | + | + | + | + | + | + | + | + | 6 |
| 8 | R26, R69 | + | + | + | + | + | + | + | + | + | 4 |
| 9 | R2 | + | + | + | + | + | + | + | + | + | 2 |
| 10 | R16, R23, R38, R39, R47, R49, R52, R64 | + | + | + | + | + | + | + | + | + | 16 |
| 11 | E15, R10, R40 | + | + | + | + | + | + | + | + | + | 6 |
| 12 | R33 | + | + | + | + | + | + | + | + | + | 2 |
| 13 | R7 | + | | | + | + | + | + | + | + | 2 |
| 14 | E27 | + | + | + | + | + | + | + | + | + | 2 |
| 15 | R77 | + | | | | | | | + | + | 2 |

Supplementary Table S4 (next page). Characterization of the bacterial collection.

In vitro plant growth promotion (PGP) tests: IAA = IAA production; ACC-d = ACC deaminase activity; Prot. = protease production. *In vivo* PGP test under hydroponic condition with *L. esculentum* plants: % germin. = germination percentage; root-l = root length; shoot-l = shoot length; 2ary roots = production of secondary roots; SVI = seedling vigor index; root dw = root dry weight. Ability to bind the root apparatus: Swimming = swimming lifestyle; Swarming = swarming lifestyle; EPS exopolysaccharides release. BPA tolerance. In vitro decolorization ability of RB5 = Reactive Black 5.

The + indicates that the bacterium is positive to the test. The stars indicate statistically significant differences (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$). R indicates the resistance to the antibiotic.

| Strain | Closest Described Relative | IAA | ACC-d | Prot. | Hydroponic Experiment | | | | | Swimming | Swarming | EPS | BPA | RB5 |
|-----------|--------------------------------------|-----|-------|-------|-----------------------|--------|---------|------------|-----|----------|----------|-----|-----|-----|
| | | | | | % Germin. | Root-l | Shoot-l | 2ary Roots | SVI | | | | | |
| CWMP-8E2 | <i>Pseudomonas thivervalensis</i> | + | + | + | | | | | | | | | + | + |
| CWMP-8E6 | <i>Flavobacterium johnsoniae</i> | + | + | + | | | | | | | | | + | + |
| CWMP-8E8 | <i>Stenotrophomonas chelatiphaga</i> | + | + | + | | | | | | | | | + | + |
| CWMP-8E13 | <i>Bacillus thuringiensis</i> | | | + | | | | | | | | | + | + |
| CWMP-8E14 | <i>Flavobacterium gyeonganense</i> | + | | | * | | | | ** | | | | | + |
| CWMP-8E15 | <i>Bacillus aryabhattachai</i> | + | + | + | | | | | | | | | | |
| CWMP-8E16 | <i>Flavobacterium gyeonganense</i> | | | | | | | | | | | | | + |
| CWMP-8E21 | <i>Pseudomonas thivervalensis</i> | + | + | + | | | | | ** | | | | + | + |
| CWMP-8E27 | <i>Bacillus aryabhattachai</i> | + | + | + | | | | | | | | | + | + |
| CWMP-8E28 | <i>Pseudomonas brassicacearum</i> | + | + | + | | | | | | | | | + | + |
| CWMP-8E33 | <i>Bacillus pumilus</i> | + | | + | | | | | | | | | + | |
| CWMP-8E42 | <i>Caulobacter segnis</i> | + | + | | | | | | ** | | | | + | |
| CWMP-8E73 | <i>Stenotrophomonas chelatiphaga</i> | + | | + | | | | | | | | | | |
| CWMP-8R1 | <i>Bacillus niacini</i> | + | | | | | | | | | | | + | |
| CWMP-8R2 | <i>Microbacterium kitamiense</i> | | + | | | | | | ** | * | * | | | + |
| CWMP-8R3 | <i>Bacillus thuringiensis</i> | + | + | + | | | | | | | | + | | 24% |
| CWMP-8R4 | <i>Flavobacterium oncorhynchii</i> | + | | | | | | | | | * | | | 17% |
| CWMP-8R6 | <i>Flavobacterium johnsoniae</i> | + | | | | | | | | | ** | | | 7% |
| CWMP-8R7 | <i>Bacillus megaterium</i> | + | + | + | ** | * | *** | | *** | * | | | | 14% |
| CWMP-8R8 | <i>Enterobacter cancerogenus</i> | + | + | | | * | *** | ** | ** | * | | + | | 32% |
| CWMP-8R9 | <i>Streptomyces althioticus</i> | | | | | | | *** | | | | | | + |

| | | | | | | | | | | | | | | | |
|-----------|--|---|---|---|---|-----|-----|-----|-----|----|---|---|---|---|-----|
| CWMP-8R10 | <i>Viridibacillus arenosi</i> | + | + | | * | ** | *** | | *** | ** | + | + | + | + | 9% |
| CWMP-8R12 | <i>Streptomyces acidiscabies</i> | + | | | | *** | *** | | *** | | | | | + | |
| CWMP-8R15 | <i>Bacillus horneckiae</i> | + | | + | | | ** | | * | * | + | + | | | 29% |
| CWMP-8R16 | <i>Lysinibacillus fusiformis</i> | + | + | | | | ** | | | | + | + | | | 11% |
| CWMP-8R17 | <i>Enterobacter cloacae</i> | + | + | | | | | * | * | | + | + | | | 37% |
| CWMP-8R19 | <i>Bacillus oceanisediminis</i> | + | + | + | | | | * | * | | | | | | |
| CWMP-8R20 | <i>Cedecea neteri</i> | + | + | | | * | * | | * | | | | | | 10% |
| CWMP-8R22 | <i>Enterobacter cancerogenus</i> | + | + | | | | | | | | | | | | 35% |
| CWMP-8R23 | <i>Lysinibacillus fusiformis</i> | + | | + | | *** | *** | ** | * | | + | + | | | 18% |
| CWMP-8R26 | <i>Flavobacterium gyeonganense</i> | + | + | | * | * | | | | | | | | | 13% |
| CWMP-8R28 | <i>Flavobacterium johnsoniae</i> | | | | | | | * | | | | | | | 5% |
| CWMP-8R31 | <i>Microbacterium kitamiense</i> | | | | | | | | * | | | | | | 6% |
| CWMP-8R32 | <i>Ochrobactrum anthrop</i> | + | + | | | | | | * | * | | | | | 27% |
| CWMP-8R33 | <i>Arthrobacter ureafaciens</i> | + | | + | | | | | | | | | | | 50% |
| CWMP-8R38 | <i>Lysinibacillus varians</i> | + | + | + | | | | | | | | | | | 22% |
| CWMP-8R39 | <i>Lysinibacillus parviboronicapiens</i> | + | | + | | ** | | | * | | + | + | | | 19% |
| CWMP-8R40 | <i>Viridibacillus arenosi</i> | + | + | | | ** | *** | | ** | | + | + | | | 7% |
| CWMP-8R47 | <i>Fluviicola taffensis</i> | | | | | | | *** | | | | | | | 20% |
| CWMP-8R49 | <i>Lysinibacillus fusiformis</i> | + | + | + | | | *** | | * | | + | + | | | 18% |
| CWMP-8R50 | <i>Pseudomonas plecoglossicida</i> | + | + | + | | | | | | | | | | | 12% |
| CWMP-8R52 | <i>Streptomyces pluricolorescens</i> | + | | | * | | *** | | *** | | | | | | |
| CWMP-8R57 | <i>Rheinheimera chironomi</i> | + | + | + | | *** | *** | | *** | | | | | | |
| CWMP-8R64 | <i>Lysinibacillus fusiformis</i> | + | + | | | | | *** | | | | + | + | | 74% |
| CWMP-8R65 | <i>Flavobacterium johnsoniae</i> | | | | | | | | *** | | | | | | 22% |
| CWMP-8R68 | <i>Bacillus marisflavi</i> | + | | + | | * | *** | | *** | | | | | | 10% |
| CWMP-8R69 | <i>Pseudomonas stutzeri</i> | + | | | | * | *** | | ** | | | | | | 30% |
| CWMP-8R72 | <i>Bacillus litoralis</i> | + | + | | | | | | | | | | | | |
| CWMP-8R76 | <i>Flavobacterium gyeonganense</i> | | | + | | *** | *** | | *** | | | | | | 16% |
| CWMP-8R77 | <i>Bacillus oceanisediminis</i> | + | | | | * | *** | | *** | | | | | | |
| CWMP-8R79 | <i>Stenotrophomonas rhizophila</i> | + | | + | | ** | *** | | ** | | | | | | 32% |
| CWMP-8R80 | <i>Streptomyces caeruleatus</i> | + | + | | | | ** | | * | | | | | | |

Supplementary Table S5. Effect of bacterial consortium on *Juncus acutus* plants grown in soil. The stars indicate statistically significant differences in comparison to the non bacterized plants (* $0.01 \leq p < 0.05$).

| Treatment | Root | | | Shoot | | | |
|-----------|----------------|---------------------|-------------------|----------------|---------------------|-------------------|-------------------------------|
| | Length (cm) | Fresh Weight (g) | Dry Weight (g) | Length (cm) | Fresh Weight (g) | Dry Weight (g) | Chlorophyll (mg/g of leaf) |
| NC | 22 | 0,78 | 0,24 | 35 | 3,30 | 0,85 | 0,23 |
| MIX | 23 | 1,37 * | 0,32 * | 36 | 4,45 | 1,10 | 0,36 * |

NC: negative control

Supplementary Figure S1. Difference in root apparatus development of (a) bacterized and (b) non bacterized tomato seedlings.

