

Exploring bacterial and fungal endophytic microbiomes of *Salicornia europaea*

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Table S1 Potential role of *Salicornia* bacterial and fungal endophytic strains reported in this study.

A

Bacterial genus	NCBI accession no.	Group	Source of isolation	Potential function	References
<i>Aestuariicella</i>	KF982858	ND	sea tidal flat	hydrocarbon-degrading	Naysim et al. 2015
<i>Arcobacter</i>	JF521610	endophyte	Bryopsis specimens	ND	Hollants et al. 2011
<i>Flavimarina</i>	KX664463	endophyte	<i>Salicornia herbacea</i>	hydrolyse gelatin	Cho et al. 2017
<i>Halomonas</i>	LWGO00000000	endophyte	roots of <i>Cyperus conglomeratus</i>	PGP	Lafi et al. 2016
<i>Idiomarina</i>	KP706811	ND	coastal plain	ND	You et al. 2015
<i>Kushneria</i>	LN651144	endophyte	leaves of Salicornioideae	ND	Mora-Ruiz et al. 2015
<i>Leeuwenhoekiella</i>	DQ294290 and DQ294291	ND	seawater	degradation of organic matter during and following algal blooms	Pinhassi et al. 2006
<i>Lewinella</i>	KX499306	epiphytes	<i>Caulerpa cylindracea</i>	ND	Stabili et al. 2017
<i>Marinimicrobium</i>	HQ882175	ND	marine solar saltern	inulinase activity	Li et al. 2012
<i>Marinobacter</i>	PRJEB8726	endophyte / rhizosphere	roots of <i>Suaeda salsa</i>	PGP	Yuan et al. 2016
<i>Marivirga</i>	JX500546	ND	rhizosphere	ND	Haldar et al. 2015
<i>Massilia</i>	ND	endophyte	stalk of maize plant	IAA producer	Celador-Lera et al. 2016
<i>Nocardioides</i>	AY148081	endophyte	wheat roots	induce defence	Conn et al. 2008
<i>Planomicrobium</i>	KP209393	endophyte	<i>Lavandula angustifolia</i>	antimicrobial activity	Beiranvand et al. 2017
<i>Pseudoalteromonas</i>	AOPM00000000	ND	saltern	enzyme activity	Haba et al. 2013
<i>Psychrobacter</i>	AJ748266	endophyte	<i>Arthrocnemum macrostachyum</i>	ND	Mora-Ruiz et al. 2016
<i>Saccharospirillum</i>	KY310592	endophyte	<i>Halimione portulacoides</i>	ND	Fidalgo et al. 2017

<i>Salegentibacter</i>	ND	ND	water and sediment samples	cytotoxic effects in mammals	Díaz-Cárdenas et al. 2017
<i>Salinicola</i>	KT036407 S	endophyte	<i>Spartina maritima</i>	PGP	Mesa et al. 2015
<i>Thalassospira</i>	KU587165	endophyte	roots of Caliph medic (<i>Medicago truncatula</i>)	ND	Yaish et al. 2016
<i>Vibrio</i>	KT036401	endophyte	<i>Spartina maritima</i>	PGP	Mesa et al. 2015

B

Fungal genus	NCBI accession no.	Group	Source of isolation	Potential function	References
<i>Plenodomus</i>	KY064055	pathogen	Chrysanthemum sp.	ND	Marin-Felix et al. 2017
<i>Paradendryphiella</i>	KT207744	ND	Marine brown algae, Sargassum sp.	antifungal and antioxidant activity	Hong et al. 2015
	AB975313	endophyte	<i>Salicornia europaea</i>	ND	Okane et al. 2015
<i>Neodevriesia</i>	NR_132905	ND	leaves of <i>Afrocarpus falcatus</i>	saprobic or plant pathogenic	Crous et al. 2014
<i>Neocamarosporium</i>	KX817217	ND	saline lakes of Iran	ND	Papizadeh et al. 2017
<i>Didymella</i>	AB975296	endophyte	<i>Salicornia europaea</i>	ND	Okane et al. 2015
<i>Alternaria</i>	EU143251	endophyte	leaves of <i>Polygonum senegalense</i>	production of natural cytotoxic metabolites	Aly et al. 2008
	FJ465171	endophyte	leaves of <i>Sonneratia alba</i>	secondary metabolites	Kjer et al. 2009

Abbreviations: **PGP**- plant growth promoting, **ND**- not defined

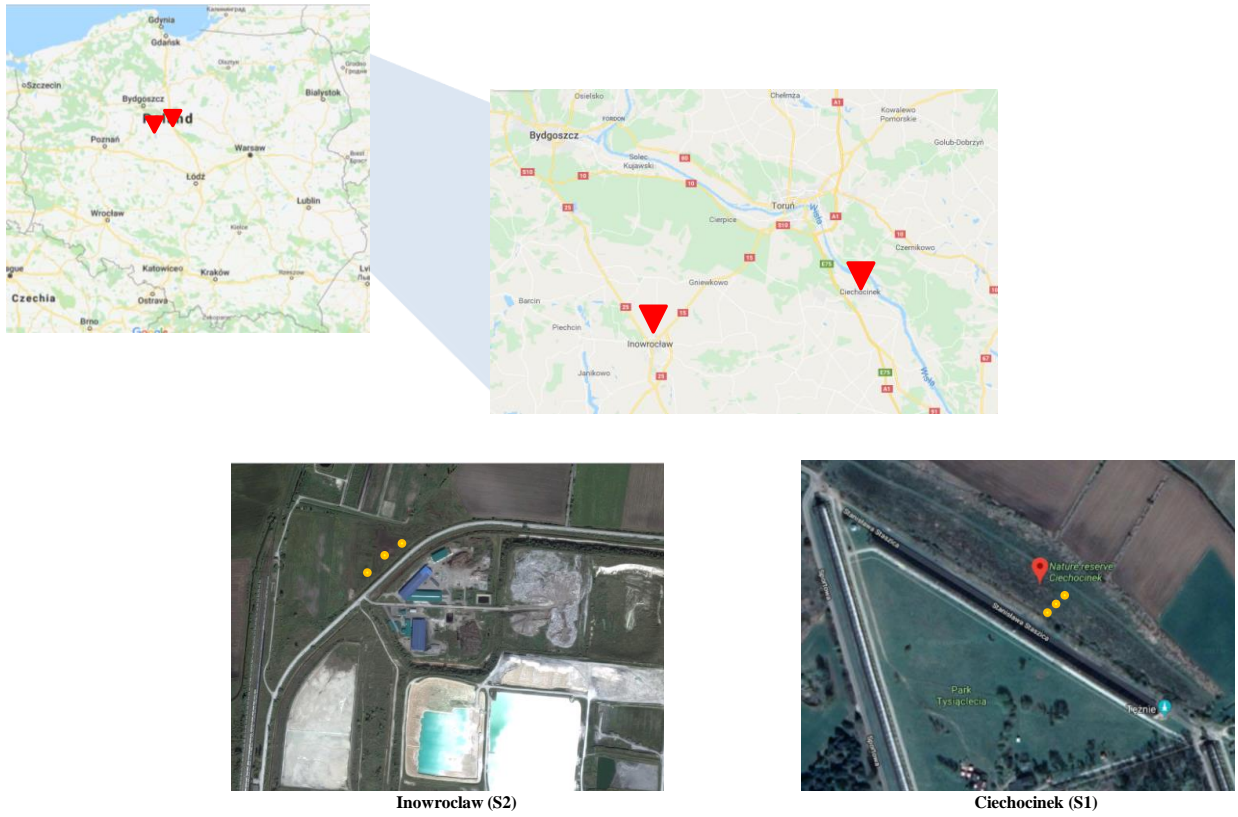


Fig. S1 Two sample collection sites in Central Poland: The map highlights the two salt affected sites S1- Ciechocinek and S2- Inowrocław and the three plots of which the three plant biological replicates were collected.

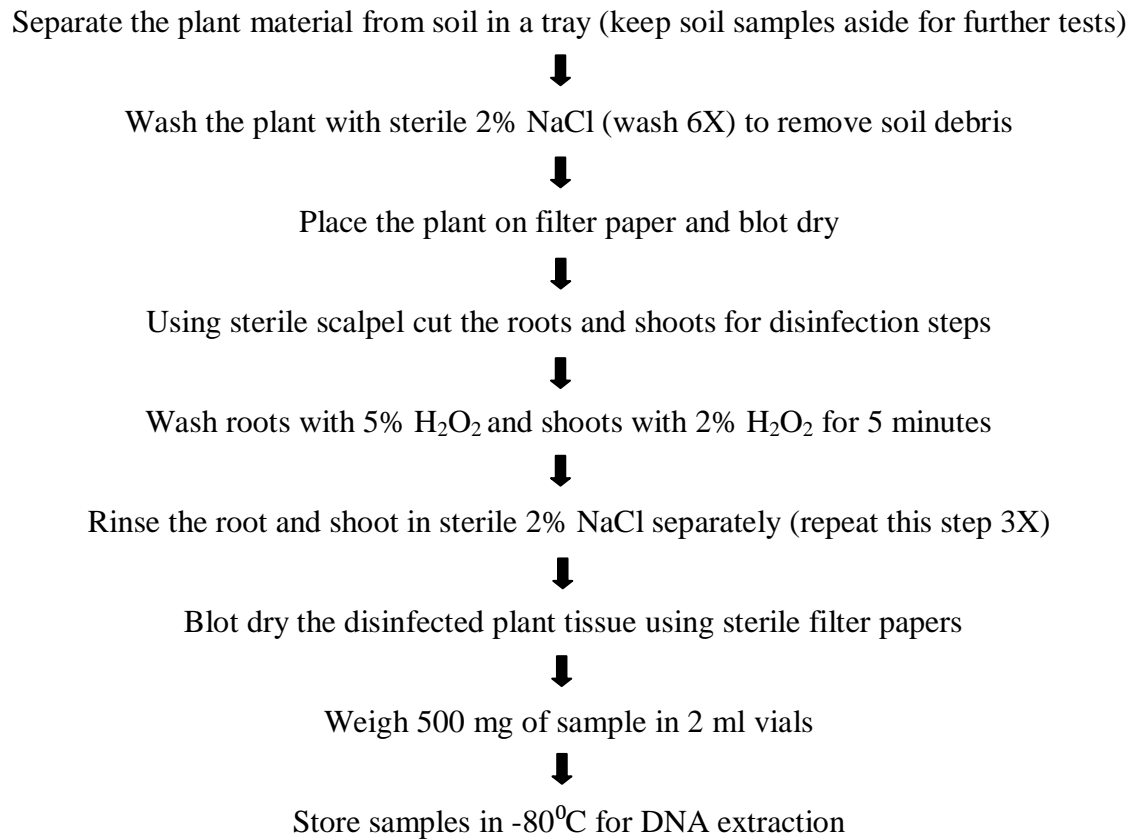


Fig. S2 Summary of plant surface sterilization technique.

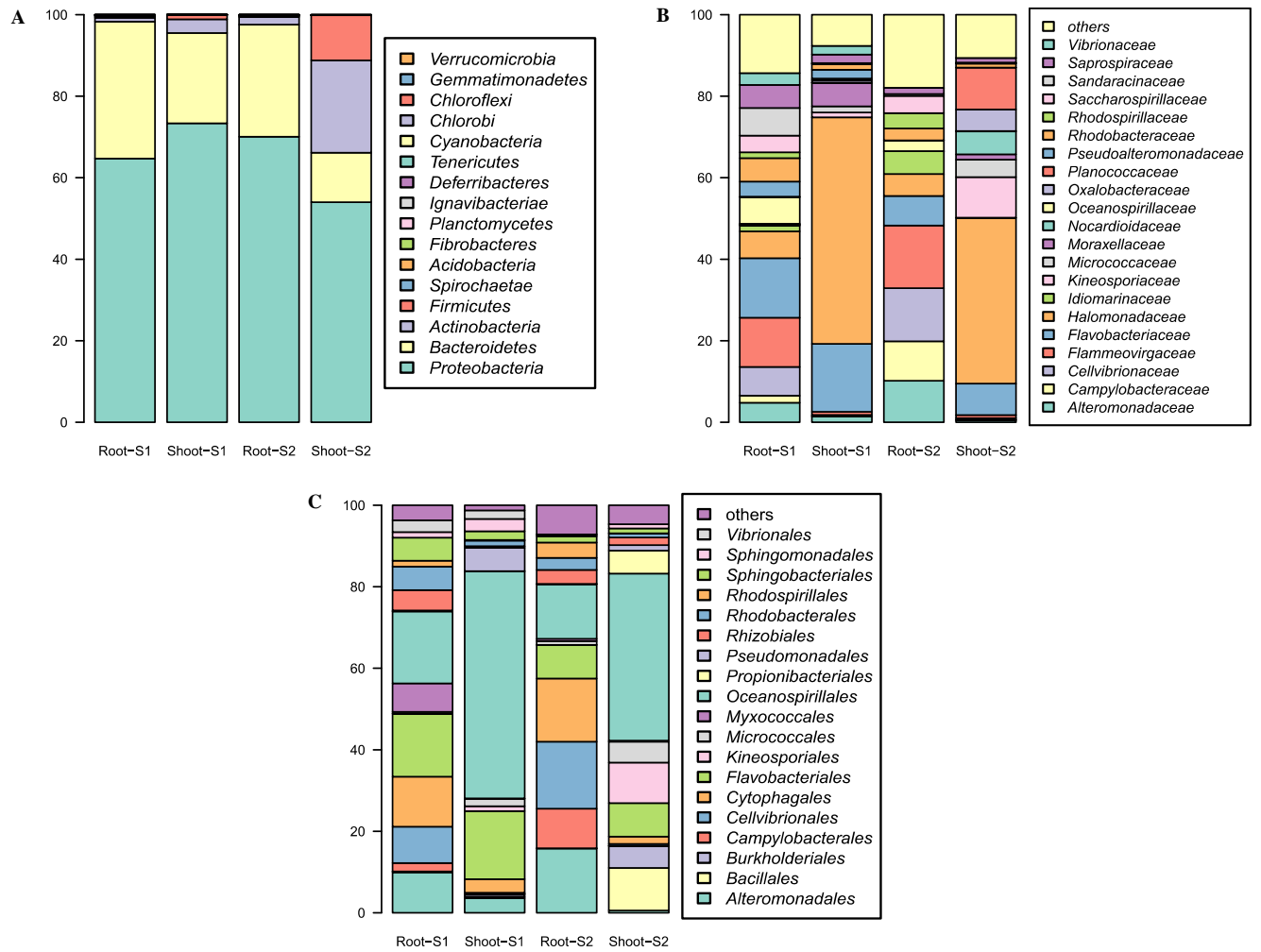


Fig. S3 Endophytic bacterial community structure at the (A) phylum (B) family and (C) order level between test sites and plant organ.

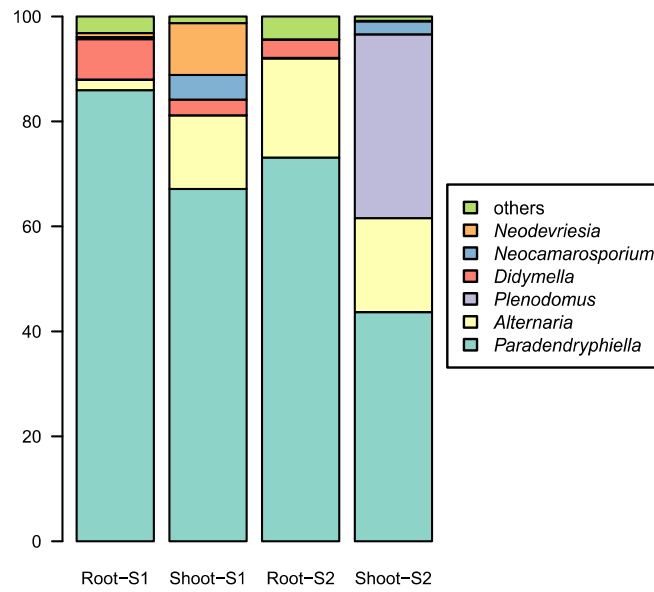


Fig. S4 Endophytic fungal community structure at the genus level between test sites and plant organ.

References

1. Aly AH, Edrada-Ebel R, Indriani ID, Wray V, Müller WEG, Totzke F, et al. Cytotoxic Metabolites from the Fungal Endophyte *Alternaria* sp. and Their Subsequent Detection in Its Host Plant *Polygonum senegalense*. *J Nat Prod.* 2008; 71:972–80. doi:10.1021/np070447m.
2. Beiranvand M, Amin M, Hashemi-Shahraki A, Romani B, Yaghoubi S, Sadeghi P. Antimicrobial activity of endophytic bacterial populations isolated from medical plants of Iran. *Iranian Journal of Microbiology.* 2017;9(1):11-18.
3. Celador-Lera L., Menéndez E., Flores-Félix J.D., Mateos P.F., Rivas R. Analysis of the PGPB Potential of Bacterial Endophytes Associated with Maize. In: González-Andrés F., James E. (eds) *Biological Nitrogen Fixation and Beneficial Plant-Microbe Interaction.* Springer, Cham 2016 doi:https://doi.org/10.1007/978-3-319-32528-6_3
4. Cho E-S, Cha I-T, Park J-M, Choi H-J, Lee JH, Roh SW, et al. *Flavimarina flava* sp. nov., isolated from *Salicornia herbacea*. *Int J Syst Evol Microbiol.* 2017;67:4240–5. doi:10.1099/ijsem.0.002292.
5. Conn V M; Walker A R; Franco C M M. Endophytic actinobacteria induce defense pathways in *Arabidopsis thaliana*. *Molecular Plant-Microbe Interactions* 2008, 21, 208-218.
6. Crous PW, Shivas RG, Quaedvlieg W, et al. Fungal Planet description sheets: 214–280. *Persoonia : Molecular Phylogeny and Evolution of Fungi.* 2014; 32:184-306. doi:10.3767/003158514X682395.
7. de la Haba RR, Sanchez-Porro C, Leon MJ, Papke RT, Ventosa A. Draft genome sequence of the moderately halophilic bacterium *Pseudoalteromonas ruthenica* Strain CP76. *Genome Announc.* 2013;1. doi:10.1128/genomeA.00268-13.
8. Díaz-Cárdenas, Carolina, Cantillo, Angela, Rojas, Laura Yinneth Sandoval, Tito, Fiorentino, Susana, Robles, Jorge, Ramos, Freddy A., Zambrano, María Mercedes, Baena, Sandra. Microbial diversity of saline environments: searching for cytotoxic activities. *AMB Express* 2017; 7:223 <https://doi.org/10.1186/s13568-017-0527-6>
9. Fidalgo C, Rocha J, Proença DN, Morais PV, Alves A, Henriques I. *Saccharospirillum correia* sp. nov., an endophytic bacterium isolated from the halophyte *Halimione*

- portulacoides. *Int J Syst Evol Microbiol.* 2017; 67:2026–30. doi:10.1099/ijsem.0.001914.
10. Haldar, S. and Sengupta, S. Impact of plant development on the rhizobacterial population of *Arachis hypogaea*: a multifactorial analysis. *J. Basic Microbiol.*, 2015; 55: 922-928. doi:10.1002/jobm.201400683
 11. Hollants, J., Leroux, O., Leliaert, F., Decleyre, H., De-Clerck, O., and Willems, A. Who is in there? exploration of endophytic bacteria within the siphonous green seaweed *Bryopsis* (Bryopsidales, Chlorophyta). *PLoS ONE* 2011; 6:e26458. doi: 10.1371/journal.pone.0026458
 12. Hong, J.-H.; Jang, S.; Heo, Y.M.; Min, M.; Lee, H.; Lee, Y.M.; Lee, H.; Kim, J.-J. Investigation of marine-derived fungal diversity and their exploitable biological activities. *Mar. Drugs* 2015, 13, 4137–4155.
 13. Kjer J, Wray V, Edrada-Ebel R, Ebel R, Pretsch A, Lin W, et al. Xanalteric Acids I and II and related phenolic compounds from an endophytic *Alternaria* sp. Isolated from the Mangrove Plant *Sonneratia alba*. *J Nat Prod.* 2009; 72:2053–7. Doi: 10.1021/np900417g.
 14. Lafi FF, Ramirez-Prado JS, Alam I, Bajic VB, Hirt H, Saad MM. Draft genome sequence of *Halomonas elongata* strain K4, an endophytic growth-promoting bacterium enhancing salinity tolerance in planta. *Genome Announc* 2016 4(6):e01214-16. doi:10.1128/genomeA.01214-16.
 15. Li, AX., Guo, LZ. & Lu, WD. Alkaline inulinase production by a newly isolated bacterium *Marinimicrobium* sp. LS–A18 and inulin hydrolysis by the enzyme World J Microbiol Biotechnol 2012; 28: 81. <https://doi.org/10.1007/s11274-011-0794-3>
 16. Marin-Felix Y, Groenewald JZ, Cai L, et al. Genera of phytopathogenic fungi: GOPHY 1. *Studies in Mycology* 2017; 86: 99–216
 17. Mesa J, Mateos-Naranjo E, Caviedes MA, Redondo-Gómez S, Pajuelo E and Rodríguez-Llorente ID Endophytic cultivable bacteria of the metal bioaccumulator *Spartina maritima* improve plant growth but not metal uptake in polluted marshes soils. *Front. Microbiol.* 2015; 6:1450. doi: 10.3389/fmicb.2015.01450
 18. Mora-Ruiz, M. D. R., Font-Verdera, F., Orfila, A., Rita, J., and Rossel-Mora, R. Endophytic microbial diversity of the halophyte *Arthrocnemum macrostachyum* across

- plant compartments. FEMS Microbiol. Ecol. 2016; 92, 1–10. doi:10.1093/femsec/fiw145
19. Mora-Ruiz, M. R., Font-Verdera, F., Díaz-Gil, C., Urdiain, M., Rodríguez-Valdecantos, G., González, B., et al. Moderate halophilic bacteria colonizing the phylloplane of halophytes of the subfamily Salicornioideae (Amaranthaceae). Syst. Appl. Microbiol. 2015; 38, 406–416. doi: 10.1016/j.syapm.2015. 05.004
 20. Naysim Lo, Kyung Hyun Kim, KyunghwaBaek, Baolei Jia and Che Ok Jeon *Aestuariicella hydrocarbonica* gen. nov., sp. nov., an aliphatic hydrocarbon-degrading bacterium isolated from a sea tidal flat. International Journal of Systematic and Evolutionary Microbiology 2015, 65, 1935–1940 DOI 10.1099/ijs.0.000199
 21. Okane, I., and Nakagiri, A. Assemblages of endophytic fungi on *Salicornia europaea* disjunctively distributed in Japan: towards clarification of the ubiquity of fungal endophytes on halophytes and their ecological roles. Curr. Sci. 2015; 109, 62–71
 22. Papizadeh, M., Wijayawardene, N.N., Amoozegar, M.A. et al. *Neocamarosporium jorjanensis*, *N. persepolis*, and *N. solicola* spp. nov. (Neocamarosporiaceae, Pleosporales) isolated from saline lakes of Iran indicate the possible halotolerant nature for the genus *Mycol* Progress 2018 17: 661. <https://doi.org/10.1007/s11557-017-1341-x>
 23. Pinhassi J, Bowman J, Nedashkovskaya O, Lekunberri I, Gomez-Consarnau L, Pedrós-Alió C. *Leeuwenhoekella blandensis* sp. nov., a genome-sequenced marine member of the family Flavobacteriaceae. Int J Syst Evol Microbiol 2006; 56(7):1489-1493 doi:10.1099/ijs.0.64232-0
 24. Stabili Loredana, Rizzo Lucia, Pizzolante Graziano, Alifano Pietro, Frascetti Simonetta. Spatial distribution of the culturable bacterial community associated with the invasive alga *Caulerpa cylindracea* in the Mediterranean Sea, Marine Environmental Research, 2017; Vol. 125, Pg. 90-98, ISSN 0141-1136, <https://doi.org/10.1016/j.marenvres.2017.02.001>.
 25. Yaish MW, Al-Lawati A, Jana GA, Vishwas Patankar H, Glick BR. Impact of soil salinity on the structure of the bacterial endophytic community identified from the roots of *Caliph medic* (*Medicago truncatula*). PLoS One. 2016; 11:e0159007. doi:10.1371/journal.pone.0159007.

26. You Y-H, Jin YJ, Kang S-M, Oh S, Lee M-C, Kim J-G. Plant growth-promoting activity and identification of endophytic fungi isolated from native plant in East coast. *Korean J Microbiol.* 2015; 51:14–20. doi:10.7845/kjm.2015.5005.
27. Yuan, Z. et al. Specialized microbiome of a halophyte and its role in helping non-host plants to withstand salinity. *Sci. Rep.* 2016; 6, 32467; doi: 10.1038/srep3.