

Biotechnological Prospects of Marine Bacteria Isolated from the Sado Estuary

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Introduction & Aim

Marine ecosystems host diverse bacterial communities that play a crucial role in marine ecology and biogeochemical processes, including organic matter decomposition, nutrient recycling, and bioremediation. The harsh environment in marine conditions (e.g., temperature shifts, light variations, osmotic pressure, limited nutrients, and competition) influences and shapes these microorganisms. This fosters distinctive genetic and phenotypic adaptations to stress, ultimately giving rise to novel biosynthetic pathways for unique bioactive compounds.

Marine microorganisms also evolved several mechanisms to interact with each other, with many bacteria capable of establishing complex relations with other bacteria and eukaryotes such as microalgae. In mutualistic relations bacteria can uptake metabolites from microalgae phycosphere while recycling and providing important nutrients such as vitamins, nitrogen, phosphorous, and sulphur. Moreover, marine bacteria can also produce and secrete info chemicals such as phytohormones (indole 3- acetic acid), quorum-sensing molecules, among others, which actively impact microalgae physiology.

The unique properties of beneficial marine bacteria are key for the development of biopharmaceutical and biotechnological applications, and to boost biotechnological microalgae production and downstream processes.

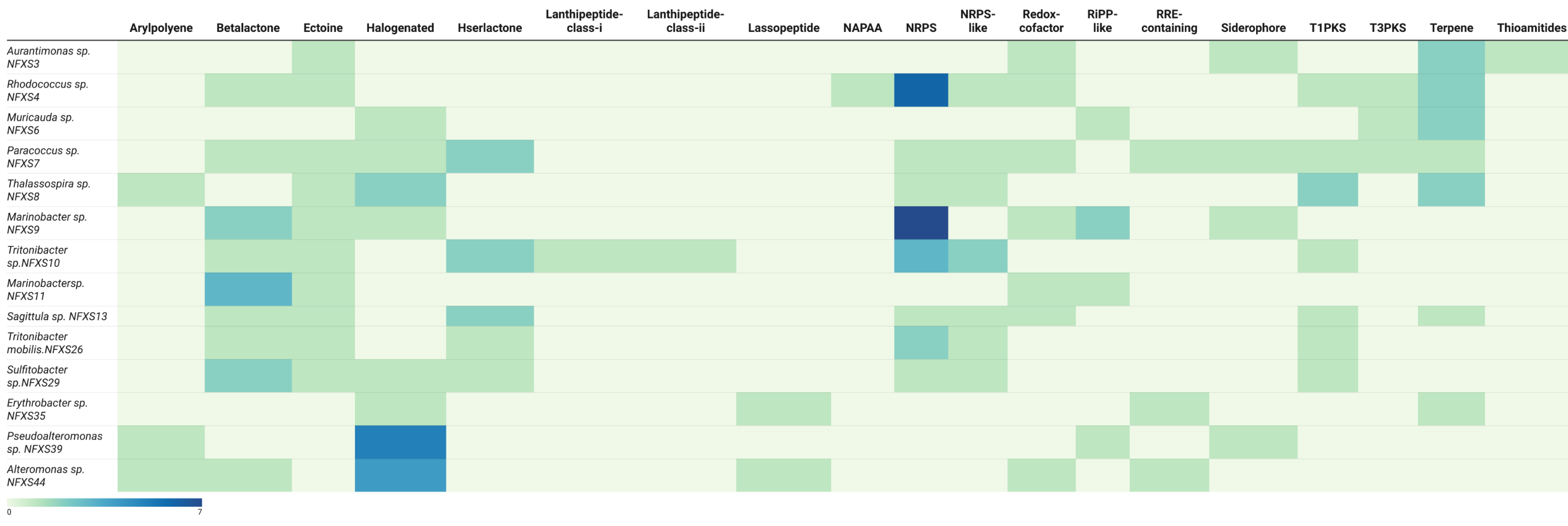
In this work, the phenotypic and genotypic characterization of fourteen marine bacteria isolated from seawater collected in diverse locations across the Sado estuary in Portugal is presented. In addition, the isolated bacteria were tested for their ability to promote the growth and the accumulation of valuable compounds in the microalgae, *Phaeodactylum tricornutum*.

Results

Genomes highlights

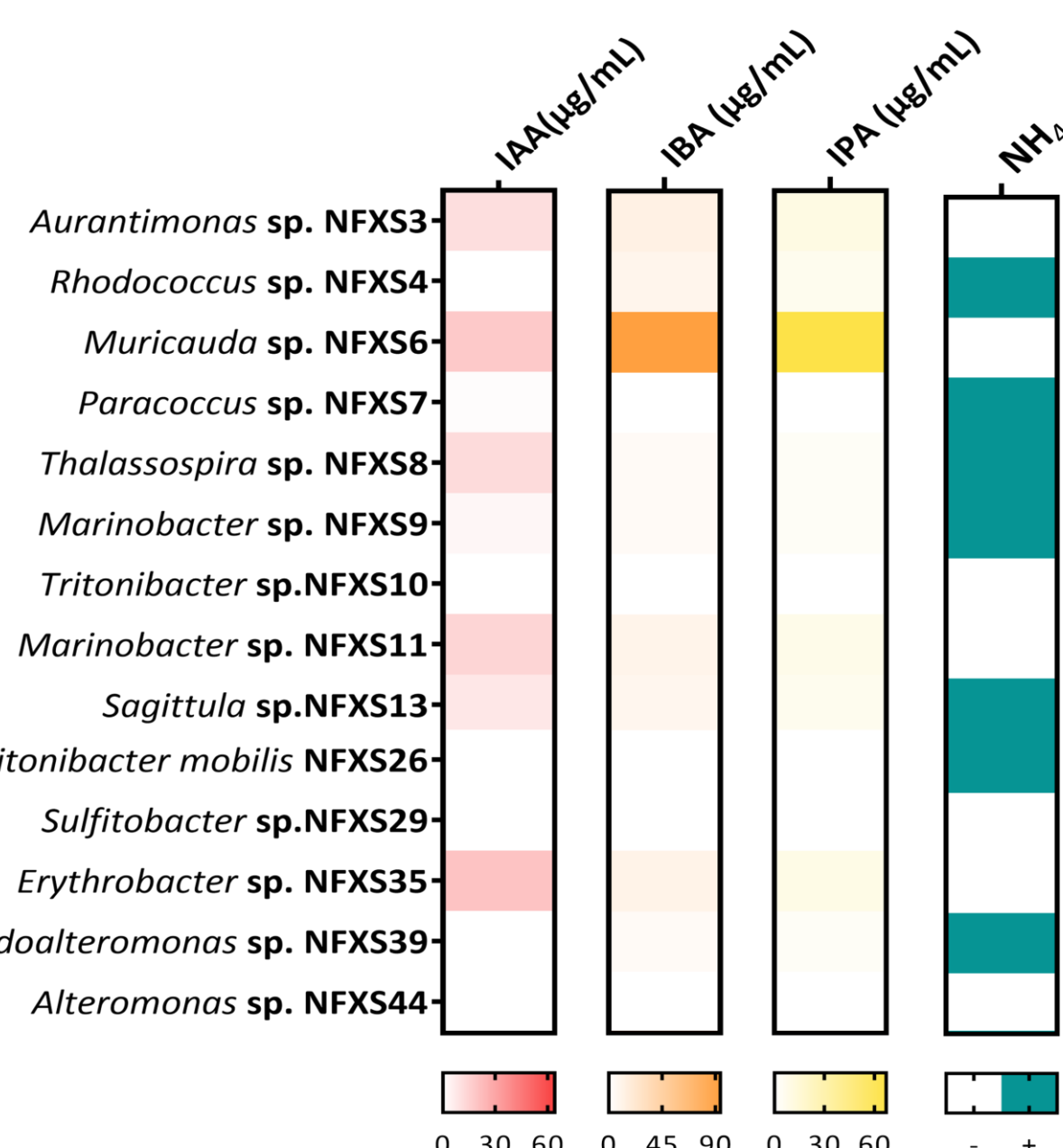
- NFXS35 has a photosynthetic system
- NFXS3, NFXS7, NFXS13 and NFXS35 have pathways for carotenoids production
- All isolates present the ability to metabolize several vitamins and cofactors B1, B2, B5, B6, B7, B12, K2, NAD, CoA, CoQ10, heme, etc...
- NFXS9, NFXS11 have a type VI secretion system, often related to interaction with eukaryotic hosts

Secondary metabolite biosynthetic gene clusters



When mining the genome, it was possible to detect nineteen clusters regarding the biosynthesis of secondary compounds, namely siderophores that can be associated with microalgae interactions.

Biosynthesis of indolic compounds and ammonia



Eight isolates were capable of producing at least one of the three indolic compounds analysed (indole-3 acetic acid, IAA; indole-3-butyric acid, IBA; and indole-3-butyric acid, IPA). Being NFXS6 the biggest indolic compounds producers;

Nine bacterial isolates were capable of producing ammonia.

Conclusions

This study provides a deep analysis of Portuguese marine bacteria isolates and their biotechnological potential.

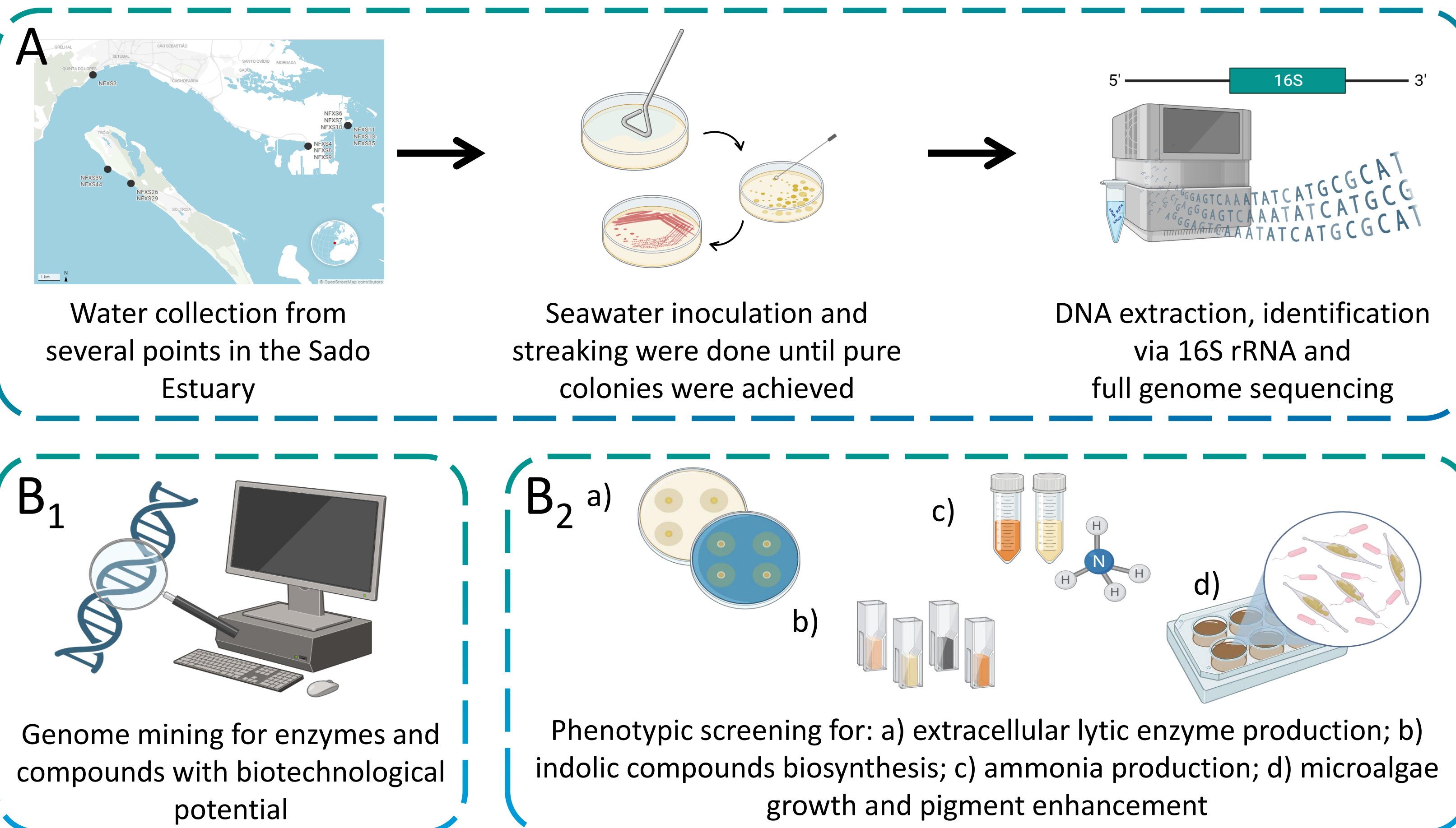
Bacterial genomic analysis revealed the presence of unique genes involved in the production of secondary metabolites of biotechnological potential.

The bacterial isolates produced functional and stable extracellular lytic enzymes under high salt conditions, interesting for application in downstream processes in marine industries.

Several genes related to the biosynthesis of microalgae growth-promoting compounds were found in bacterial genomes

The bacterial isolates promoted *P. tricornutum* growth and may be used to boost *P. tricornutum*-based biotechnological processes such as industrial fucoxanthin production.

Methods



Production of extracellular lytic enzymes in high salinity media

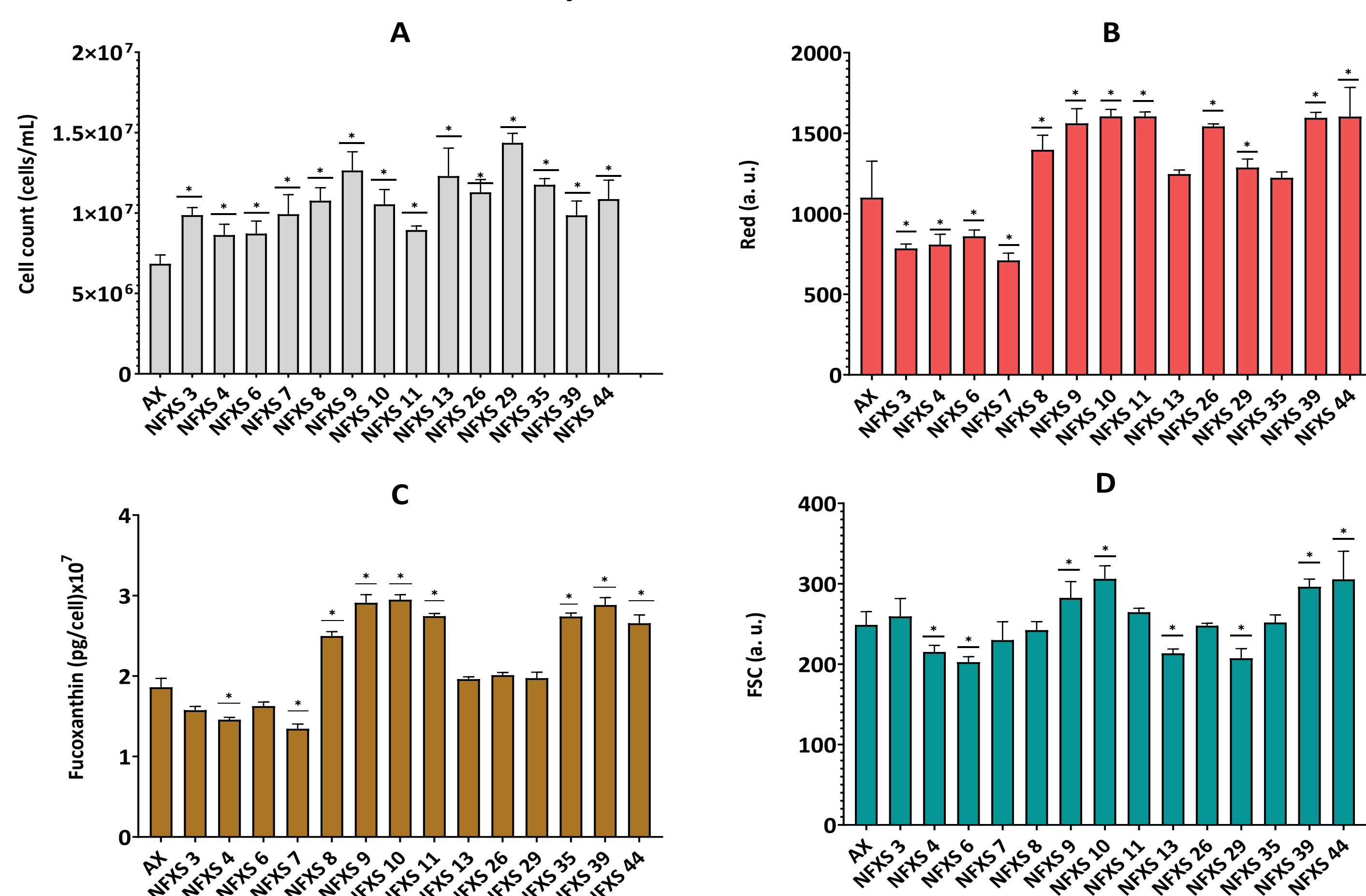
	Skimmed milk	Tributyrin	Sunflower oil	Chitin	Alginate	Cellulose	Starch	Pectin	Agar-agar
<i>Muricauda</i> sp. NFXS6	1.0	1.7	1.0	1.4	1.4	1.4	0.0	0.0	0.0
<i>Paracoccus</i> sp. NFXS7	1.1	0.7	1.6	0.6	0.0	0.5	0.0	0.6	0.0
<i>Marinobacter</i> sp. NFXS9	2.4	1.8	1.2	2.2	1.7	1.7	1.0	1.9	0.0
<i>Tritonibacter</i> sp. NFXS10	0.0	0.0	1.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>Tritonibacter</i> sp. NFXS11	0.0	0.0	1.6	0.0	0.0	0.0	0.0	0.0	0.0
<i>Sagittula</i> sp. NFXS13	0.0	0.0	1.4	0.0	0.0	0.0	0.0	0.0	0.0
<i>Tritonibacter mobilis</i> NFXS26	0.0	0.0	1.6	0.0	0.0	0.0	0.0	0.0	0.0
<i>Sulfitobacter</i> sp. NFXS29	0.0	0.0	1.3	0.0	0.0	0.0	0.0	0.0	0.0
<i>Erythrobacter</i> sp. NFXS35	0.6	0.0	0.8	0.0	0.7	0.0	0.0	0.0	0.0
<i>Pseudoalteromonas</i> sp. NFXS39	0.4	0.4	2.6	0.2	0.0	0.0	0.0	0.2	0.0
<i>Alteromonas</i> sp. NFXS44	0.0	0.3	2.0	0.0	0.0	0.0	0.0	0.2	0.0

Degradation Halo Diameter (in cm)

Created with Datawrapper

Eleven isolates were capable of degrading at least one substrate, and none was capable of degrading agar-agar; Isolate NFXS9 was able to degrade nine of the ten substrates and presented the highest inhibitions halos.

Phaeodactylum tricornutum co-cultures



All the bacterial isolates showed a notable capacity to increase *P. tricornutum* cell count (A);

Several bacteria were able to modify *P. tricornutum* chlorophyll production, hereby quantified via RED autofluorescence (C);

Regarding and fucoxanthin production (B) and cell size (D) variable effects were observed.

Acknowledgements

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