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Microbe Profile: *Salinispora tropica*: natural products and the evolution of a unique marine bacterium

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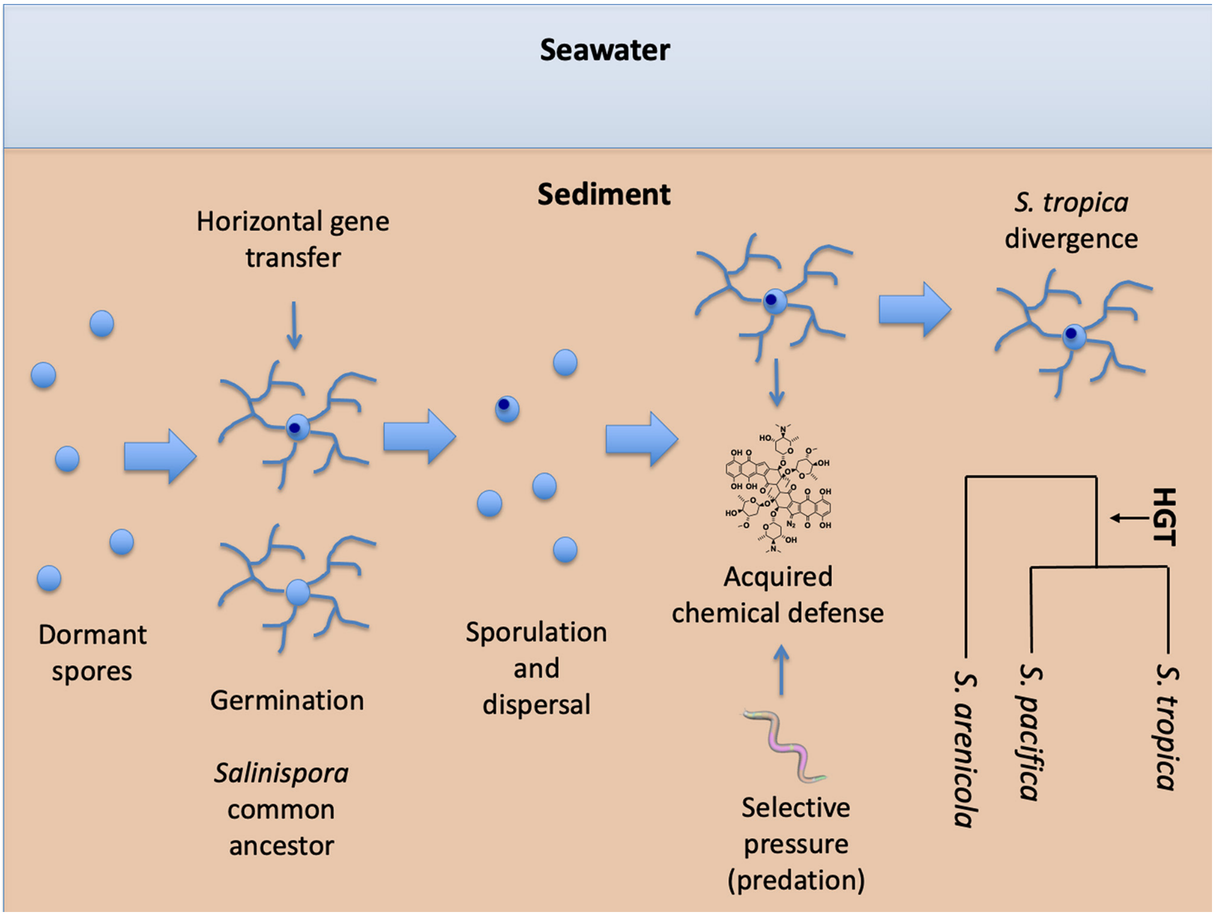

Graphical abstract

Theoretical model of *Salinispora tropica* ecology and evolution. Dormant spores are distributed over unknown distances, germinating when environmental conditions are suitable for growth. Vegetative growth provides opportunities for the acquisition of new traits via horizontal gene transfer (HGT event represented by dark blue circle) followed by subsequent rounds of sporulation, dispersal and germination. Acquired traits that provide a selective advantage, such as a chemical defence against predation, are maintained through vertical inheritance, facilitating niche expansion, and ultimately contributing to *Salinispora* diversification.

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Abstract

Salinispora tropica was originally cultured from tropical marine sediments and described as the first obligate marine actinomycete genus. Soon after its discovery, it yielded the potent proteasome inhibitor salinosporamide A, a structurally novel natural product that is currently in phase III clinical trials for the treatment of cancer. If approved, it will be the first natural product derived from a cultured marine microbe to achieve clinical relevance. *S. tropica* produces many other biologically active natural products, including some linked to chemical defence, thus providing ecological context for their production. However, genomic analyses reveal that most natural product biosynthetic gene clusters remain orphan, suggesting that more compounds await discovery. The abundance of biosynthetic gene clusters in *S. tropica* supports the concept that the small molecules they encode serve important ecological functions, while their evolutionary histories suggest a potential role in promoting diversification. Better insights into the ecological functions of microbial natural products will help inform future discovery efforts.

TAXONOMY

Phylum: Actinobacteria; class: Actinomycetia; order: Micromonosporales; family: *Micromonosporaceae*; genus: *Salinispora*; species: *Salinispora tropica*.

PROPERTIES

Salinispora tropica is a slow-growing, chemo-organotrophic, Gram-positive bacterium. It is non-motile, spore-forming and an obligate aerobe. Strains produce substrate hyphae that are finely branched, do not fragment and carry smooth surfaced spores. Colonies take two or more weeks to appear on agar media, lack aerial hyphae and form bright to pale orange colonies that darken during sporulation. All strains fail to grow when seawater is replaced with deionized water in the growth medium, an observation that has been linked to the loss of a mechanosensitive channel gene whose product helps overcome osmotic stress.

GENOME

S. tropica strain CNB-440 is the type strain for the species and the first member of the genus to be genome sequenced. The 5.2 Mb circular chromosome encodes 4536 predicted proteins and has a G+C content of 69.5%. Remarkably, this strain devotes approximately 9% of its genome to natural product biosynthesis and, at the time it was sequenced, encoded the most diverse assemblage of polyketide biosynthetic machinery observed in a single organism [1]. Analysis of the genome aided in the structure elucidation of a new polyketide and helped establish the concept of genome mining as a new approach to natural product discovery. This approach enabled the rapid detection of a candidate biosynthetic gene cluster for the potent proteasome inhibitor salinosporamide A, which is currently in phase III clinical trials for the treatment of cancer. Broader genomic studies revealed that a majority of gene clusters remain orphan [2], while transcriptomics revealed that many of these were not expressed during laboratory cultivation [3], suggesting that specific environmental cues are missing when traditional culture media are employed. Comparative genomics revealed that both horizontal gene transfer [4] and vertical inheritance [5] play key roles in both the diversity and distribution of natural product genes and in the evolution of chemical diversity in *Salinispora*.

PHYLOGENY

While *S. tropica* was originally identified based on 16S rRNA sequence analysis, this highly conserved gene subsequently proved insufficient to resolve species-level relationships within the genus. Additional phylogenetic markers, and ultimately phylogenomic analyses, facilitated an increase in the number of described species to nine [6]. As originally observed for specialized metabolite production, and corroborated with additional genome sequencing, natural product biosynthetic gene clusters are conserved in species-specific patterns in *Salinispora*, with the gene cluster that encodes salinisporamide A observed in all *S. tropica* genomes sequenced to date.

KEY FEATURES AND DISCOVERIES

While it was long recognized that spore-forming actinomycetes could be recovered from ocean sediments, it was not clear if they originated from dormant spores or were active members of the marine microbial community. The cultivation of *S. tropica* and its recognition as an obligate marine bacterium provided the first evidence that some filamentous actinomycetes are endemic to marine habitats. To date, *Salinispora* represents the only marine obligate actinomycete genus. It has proven to be a rich source of novel natural products and a model organism for biosynthetic, ecological and evolutionary studies [7]. The discovery of salinosporamide A, and the unique mechanism of its biosynthesis, suggest that adaptations to life in the ocean can include the ability to produce unique natural products. While the ecological functions of most *S. tropica* natural products remain unknown, evidence suggests that they mediate competitive interactions [8] and defend against eukaryotic predators [9]. Furthermore, phytoplankton exudes have been shown to trigger the production of cryptic *Salinispora* metabolites [10], suggesting that simulated ecological interactions represent a path forward to find the products of orphan gene clusters. For reasons that remain unknown, *S. tropica* cultures have only been reported from the Caribbean, while other *Salinispora* species display broader distributions. Possibly owing to this restricted geographic distribution, *S. tropica* has undergone minimal diversification compared to other *Salinispora* species. However, certain biosynthetic pathways, such as that encoding the production of salinisporamide A, are fixed at the species level, despite evidence that they were acquired by horizontal gene transfer [4]. This provides strong support that the products of some gene clusters serve important ecological functions and raises the intriguing possibility that specialized metabolites facilitate ecological differentiation and, ultimately, diversification.

OPEN QUESTIONS

- Why does the geographical distribution of *S. tropica* appear to be restricted to the Caribbean?
- How long do *S. tropica* spores survive and how far can they travel?
- What are the ecological functions of salinosporamide A and other compounds produced by this species?
- What are the small molecule products of the orphan biosynthetic gene clusters in *S. tropica* and what factors are needed to induce their production?
- Are there additional *Salinispora* species that have yet to be identified?

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Conflicts of interest
The authors declare that there are no conflicts of interest.

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