Title
Antibiotic Drug Discovery from the New Marine Actinomycete Genus Marinomyces

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Antibiotic Drug Discovery from the New Marine Actinomycete Genus Marinomyces

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Cultures of different marine bacteria extracted from ocean sediments.

BACKGROUND

Actinomycetes are a group of bacteria that live in soil and decompose organic matter such as cellulose. In the realm of drug discovery, these microorganisms are widely recognized for their ability to produce secondary metabolites (chemicals) with commercially viable antibiotic activity. Streptomycin, the first treatment for tuberculosis, was derived from the largest genus of these bacteria, Streptomyces. Erythromycin and tetracycline are two other examples of common medicines derived originally from these microorganisms' metabolites.

PROJECT OBJECTIVE

Within the last decade, researchers have discovered that actinomycetes also inhabit seafloor sediments. These bacteria include an exceptionally chemically prolific group, never observed in terrestrial soils, dubbed MAR2 bacteria or Marinospora.

The goal of this project was to gain a better understanding of the diversity of MAR2 bacteria in marine sediments and their ability to produce new antibiotics. Another closely related objective was to elucidate links between the taxonomy of these bacteria and their chemistry (i.e., the antibiotics they produce).

MILESTONES

During the course of this project, researchers cultured 25 strains of MAR2 bacteria and sequenced the 16S rRNA gene of 22 of them. The nucleotide signatures for the different strains showed that the MAR2 clade is comprised of at least eight species.

The gene sequences were also used to develop MAR2-specific PCR primers – probes that make it possible to rapidly identify MAR2 bacteria from other, more numerous marine actinomycetes.

With an array of enrichment cultures, researchers amended media with six antibiotics (penicillin, rifampicin, vancomycin, streptomycin, kanamycin and gentimycin) at concentrations ranging from 10 µg/ml to 200 µg/ml to select for strains with antibiotic resistance. Marine actinomycetes were isolated using four low-nutrient media: seawater agar, kelp powder, chitin and sediment. After 14 days, researchers extracted DNA from 90 of these cultures and probed the genetic material. Through these experiments, researchers identified conditions needed to culture MAR2 strains.

They also discovered two new interesting compounds—a pentaene spiropyran and polyene glycoside. Both are polyketide-derived, polyene macrolides with considerable variation in the degree of unsaturation, hydroxylation and aromatization in their respective macrocyclic ring systems. The researchers reported a consistent pattern of polyene production, especially the production of polyene macrolides, from MAR2 strains. The ability of MAR2 strains to produce such an important class of pharmaceutical agents (polyene macrolides) is a major reason for continued interest in these bacteria.

Erin Gontang, UCSD

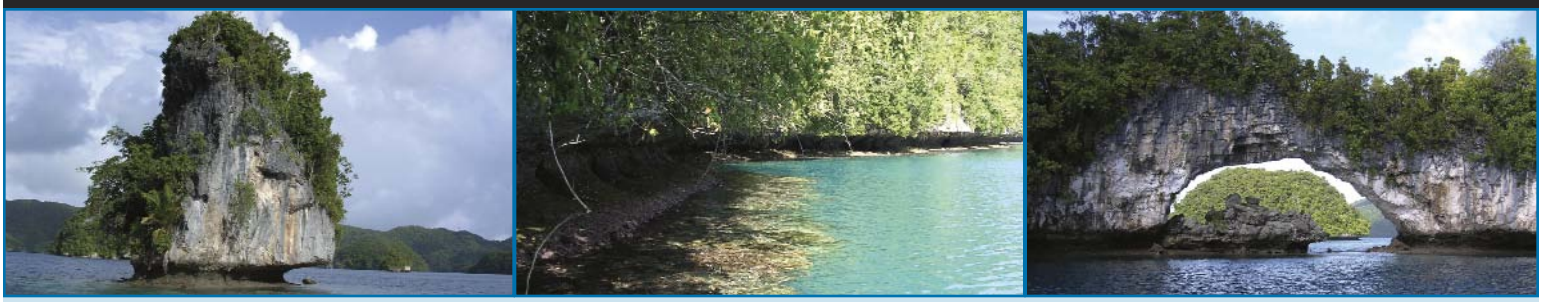
Marine bacteria in culture.

The MAR2 compounds isolated during this project displayed a range of biological activity. Marinomycin A had potent antibacterial activity (0.13 µM) against methycillin-resistant *S. aureus*. Marinomycins B-C displayed considerably less cytotoxicity (2.6–3.1 µM) with approximately equal antibacterial activity (0.25 µM). These compounds have commercial potential and a provisional patent on them has been filed.

In addition to finding new compounds, researchers isolated five new analogues of previously described metabolites. Especially noteworthy were two new derivatives in the aburatolactam series from the MAR2 strain CNQ233. One of these compounds, desmethylaburatolactam A, displayed antifungal activity at less than 1.5 µM against amphoterocin-resistant *Candida albicans*. Desmethylaburatolactam B possessed highly potent cytotoxicity with an IC50 value of 30 µM versus a human colon tumor cell line. These compounds are being investigated as new antifungal and anticancer leads, respectively.

CONCLUSIONS

The MAR2 group has considerable phylogenetic diversity. Indeed, the scientists believe there should be a taxonomic revision of the genus Streptomyces, as during this study it was shown to include multiple genera.



Researchers traveled to Palau in 2004 to collect marine sediments. Erin Gontang, UCSD

Michael Aw

A stunning aerial view of Palau, where many of the bacteria studied in this project were collected.

In terms of finding new compounds for drug discovery, phylogenetic novelty is golden – more important than other factors long associated with biochemical novelty, such as geographic origin. Phylogenetically similar strains of marine actinomycetes were observed to produce the same chemistry regardless of where they were collected. For example, bacterial strains obtained from sediments off San Diego and Palau all produced marinomycin A. The scientists wrote in their report to Sea Grant that "more effort should be focused on developing new cultivation methods or sampling new niches as opposed to traveling to remote sites to collect uniform sample types."

APPLICATIONS

This research has led to the development of a phylogenetic approach to assessing the biosynthetic richness and novelty of individual strains. This approach was accomplished by performing phylogenetic analyses on PCR-amplified keto-synthase (KS) domains from modular polyketide synthase complexes. By looking at the KS domains, researchers could predict that a strain cultured from a Palau sample would produce the macrolide tetronomycin, because the KS domains from this strain clustered closely with those previously reported for the compound. Such an approach could be used as a prescreening process to speed the rate at which new compounds are isolated.

STUDENTS

Sandy Myskowski, Ph.D.
Erin Gontang, Ph.D.

PUBLICATIONS

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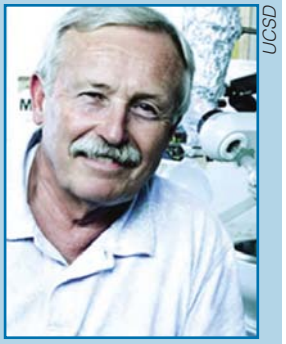

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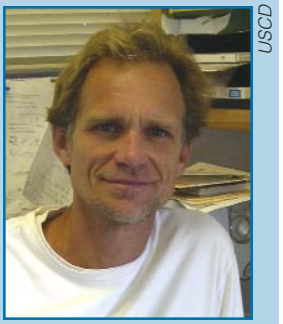

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