

Metagenomic Sequencing of Prokaryotic Microbiota from Tropical Surface Seawater

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Tropical seawater harbors a rich diversity of microorganisms as a result of its nutrient-rich environment, constant supply of sufficient sunlight, and warm climate. In this report, we present the complexity of the microbial diversity of the surface seawater of the Georgetown coast as determined using next-generation sequencing technology.

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Seawater harbors a great diversity of living organisms, ranging from microscopic bacteria, *Archaea*, fungi, and *Protista* to macroscopic crustacean and fishes, each playing a major role in the biogeochemical cycle (1, 2). South Channel, located at the tropical coast at the northern part of the Malacca Straits, Malaysia, is believed to harbor great oceanic biodiversity due to the warm seawater and consistent exposure to sunlight. According to several studies, the microbial diversity increases as one gets closer to the coastal area (3). Therefore, this study aims to investigate the marine microbial diversity of the surface seawater of the Georgetown coastal area.

A sterile amber bottle was used to collect 1 liter of water from the sea surface of the Georgetown coast (05°25.587'N, 100°19.591'E), Penang, Malaysia, and the water was filtered through a filter membrane (pore size of 0.22 μm) (Sartorius, Germany). Metagenomic DNA was extracted and purified with a modified cetyltrimethylammonium bromide (CTAB) DNA extraction method (4, 5). The purified DNA was processed for sequencing-library preparation with the TruSeq DNA sample preparation kit (Illumina) prior to shotgun sequencing with MiSeq (Illumina). A total of 6,701,060 raw reads were channeled to the CLC Genomic Workbench 5.1 (CLC bio, Denmark) for trimming and *de novo* assembly (6). Assembled sequences were compared against the Cyberinfrastructure for Advanced Microbial Ecology Research and Analysis (CAMERA) for microbial diversity, and its outputs were imported into MEGAN4 software (version 4.64.2) for classifications (7, 8). Microorganism abundance was assessed by counting the number of reads.

In terms of the kinds of organisms present, the majority were members of the *Eubacteria* and *Archaea*, making up 75% of the total organism presence. A total of 31 known bacterial phyla, together with a significant number of unclassified bacteria, were reported from this sample, suggesting rich bacterial diversity at the seawater surface of the Georgetown coast. Among the *Eubacteria*, the *Proteobacteria* (72%) make up the most abundance phylum, followed by the *Bacteroidetes* (9.4%), *Firmicutes* (5.1%), *Actinobacteria* (3.0%), *Verrucomicrobia* (1.8%), and *Cyanobacteria* (1.8%). On the other hand, the major phylum of the *Archaea*

found in this sample was *Euryarchaeota*, contributing 65.2% of the *Archaea* diversity. Members of the *Thaumarchaeota* make up 17.4% and members of the *Crenarchaeota* make up 9.6% of the total *Archaea* presence in this sample, making these 2 of the most abundant phyla of *Archaea* found.

In this study, we report the rich microbial diversity found at the sea surface of the tropical Georgetown coast. The benefits of this great microbial diversity of the tropical marine waters, along with the diverse microbial gene activities, might be the key to enhancing the bioremediation dynamics of the polluted seaside.

Nucleotide sequence accession number. The DNA sequences from this metagenomic project were deposited at DDBJ/EMBL/GenBank under the accession no. [SRP014764](https://www.ncbi.nlm.nih.gov/nuclseq/SRP014764).

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