Biodiversity Baselines and Biopharmaceutical Potential for the Borderland

By Lisa Levin, Paul Jensen, Greg Rouse, Kira Mizell, Gabriel Castro-Falcón, Kaila Pearson, Kaitlin Creamer, Devin Vlach, Steven Auscavitch, and Dwight F. Coleman

A team of ecologists, microbiologists, zoologists, and a geologist explored the Southern California Borderland from October 27 to November 6, 2020. The cruise goals were to collect samples and data needed to generate baseline information describing faunal and microbial community structure and biopharmaceutical potential across two poorly explored, mineral-rich ecosystems associated with phosphorites and ferromanganese crusts. We sought to characterize fish and benthic megafauna and macrofauna associated with these hardground habitats as well as the microbial communities associated with sediments, mineral surfaces, and select invertebrates.

Operations consisted of sonar-based mapping by E/V *Nautilus*, an ROV *Argus* test dive, and ROV *Hercules*/*Argus* dives during which we conducted biodiversity surveys and collected samples in conjunction with hydrographic data collection via an ROV-mounted CTD. ROV work was conducted from 1,800 m to 100 m depth at eight sites located 65 km to 240 km offshore of southern California, including Patton Ridge, San Juan Seamount, Northeast Bank, Cortes Bank, 40-Mile Bank, San Clemente Escarpment, and Osborn Bank (Figure 1). During each eight-hour dive, four to six 100 m-long video transects were conducted for habitat and biodiversity analysis, and samples of megafauna, sediments, rocks, and water were collected for microbiological, animal, and mineral substrate analyses. On San Juan Seamount and 40-Mile Bank, a set of colonization substrates (wood, bone, carbonate, ferromanganese crust on basalt, and phosphorite rock) were deployed at about 1,050–1,100 m and at 693 m depths within the oxygen minimum zone core; they will be recovered in one year. Mapping conducted between ROV dives filled in bathymetric and backscatter data gaps on transits between study sites.

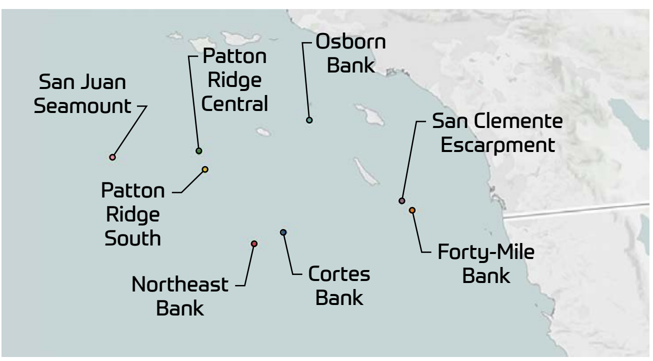

FIGURE 1. Map of ROV *Hercules* dive sites.

Complicated tectonics create the rough topography of the Southern California Borderland, which includes a series of banks, ridges, knolls, seamounts, and escarpments. Each of these geologic edifices provide exposed rock hardgrounds that can act as biological substrates and habitats. The presence of an oxygen minimum zone (<20 µMol O_2) combined with stronger currents that reduce sedimentation, especially at depths below 850 m, has resulted in the precipitation of ferromanganese crusts on the exposed surfaces of basalts and sedimentary rocks. Phosphorites are also prevalent in the region. Both phosphorites (Figure 2a) and ferromanganese crusts (Figure 2b) can contain metals at high enough concentrations to be considered as potential mineral resources. Although no

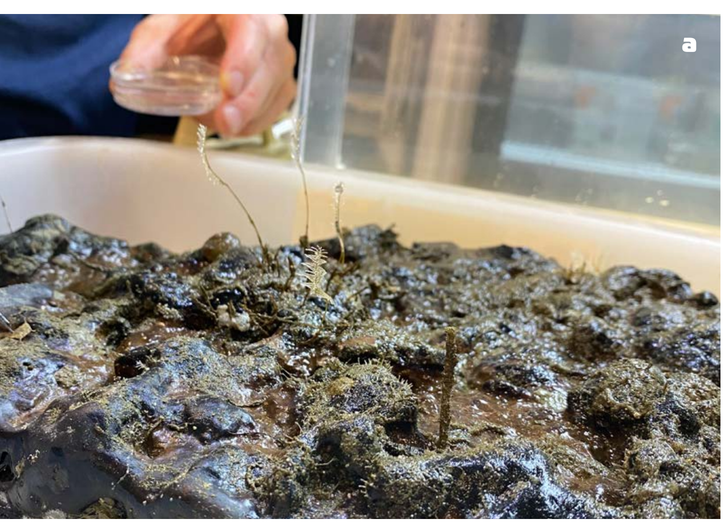

FIGURE 2. (a) Phosphorite rock with fauna collected on Cortes Bank. (b) Ferromanganese crust collected on San Juan Seamount.

FIGURE 3 (left). Brisingid starfish and sponges imaged on San Juan Seamount.

FIGURE 4 (below). Sedimentary oblate rock showing fossilized marine mammal with exposed rib cage.

exploitation of either marine mineral type has occurred anywhere in the deep sea, and there is currently no interest in resource exploration in the Southern California Borderland, a multidisciplinary research effort such as ours, examining marine mineral and concomitant ecosystem resources, can inform decisions about their potential exploitation.

The hardgrounds appear to host a moderately high diversity of megafauna, macrofauna, and fish, and in some places, high densities of animals. Different taxa dominate the megafaunal assemblages in each place visited. Both offshore/inshore and depth/oxygen differences were evident. The base of escarpments, banks, and ridges were typically sediment-covered, with high densities of sea cucumbers (*Scotoplanes*), sea pens (*Halipteris or Anthoptilum*), other cnidarians (*Umbellula*), brittle stars, and other taxa. Oxygen availability appears to exert a strong influence on the hardground biota; zonation of assemblages on the slopes and flanks of the explored features was evident between 1,100 m and 500 m depth. Dense bands of brittle stars or zones of abundant brisingid starfish (Figure 3), sponges (*Hyalonema, Mycale, Farrea, Abestopluma, Heterochone*), sea cucumbers (*Pannychia, Psolus,* and others), sea urchins (*Araesoma, Asthenosoma*), or bivalves (*Acesta*) were observed, possibly reflecting oxygen minimum zone edge effects (oxygen thresholds intersecting with high food availability). Corals such as bubblegum coral (*Paragorgia*) and *Swiftia* were common at shallower depths (<600 m). Crabs (*Paralomis*) were common, and kelp falls (mixed with *Phyllospadix* seagrass), observed at Patten Ridge South and 40-Mile Bank, were characteristically colonized by pink urchins (*Strongylocentrotus fragilis)* and small consumers (amphipods or dorvilleid polychaetes). Large erosional carbonate discs were observed on Cortes Bank (which previously was subaerially exposed), with one containing the fossilized skeleton of a marine mammal, likely a juvenile whale (Figure 4). Fishing gear and debris (soda cans, lost instrumentation, cable connectors) were observed on the seafloor on many of the dives.

Rocks, sediments, invertebrates, and water collected by the ROV manipulator, push cores, vacuum slurp, and Niskin bottles will be studied further to determine the biopharmacological properties of associated bacteria. We will use a culture-dependent approach to grow and isolate individual colonies of Actinobacteria, which are prolific producers of molecules that have been used clinically to treat numerous ailments. Culture collections of these isolated bacteria will be screened to find whether they inhibit bacterial and cancerous cell growth. Additionally, we will assess the microbiome of the samples using a culture-independent approach. Metagenomics and amplicon sequencing will be used to find and compare genes putatively involved in the generation of medicinal metabolites and to assess the overall microbial community. This will allow us to evaluate the biopharmacological potential of a broader range of bacteria and compare microbial diversity across the samples.

It is currently unclear whether the animal and microbial communities occupying phosphorite or ferromanganese crust differ. To investigate this further, we will consider the location, water depth, seawater oxygen, mineral texture, and geochemical composition of each sample and their correlations with the associated biological communities. This analysis will elucidate the dominant factors controlling their diversity and abundance.