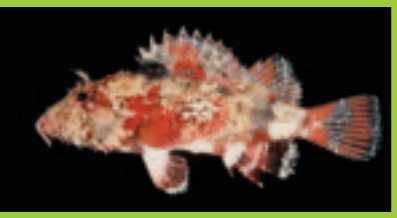
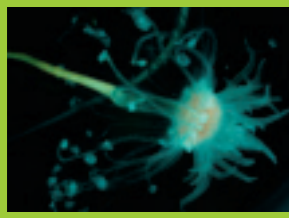
Genetic Analysis on the High Seas

The Census of Marine Life Project
Funded by the National Oceanic and Atmospheric Administration

Begun in 2000, the Census of Marine Life is an unprecedented 10-year, global scientific collaboration that has, as its goal, the inventorying of biodiversity throughout the seas to understand marine populations and their effect on global warming. In addition to discovering species, the census will also try to determine the patterns of species distribution, their abundance, and the factors that maintain those patterns. Oceanographic researchers from more than 14 countries are engaged in the project. The laboratory, which included an Applied Biosystems 3130 Genetic Analyzer, was set up on the research vessel and experiments were carried out at sea.







Main image (top): Sunflower sea stars *(Pycnopodia helianthoides),* Photo: Casey Debenham. Directly above: New species of scorpionfish *(Scorpaenopsis Vittapinna),* Photo: Bill Eschmeyer and John E. Randall; Lobate ctenophore *(Bathycyroe fosteri),* Photo: Marsh Youngbluth; Jelly fish of the genus Crossota, Photo: Kevin Raskoff, NOAA; New species of comb jelly, a Cydipppid ctenophore, Photo: Kevin Raskoff, NOAA; A yet unidentified cnidarian, Photo: Bluhm and Iken, NOAA.

"By 2010 the research conducted by this project will provide a baseline against which future generations can measure changes to the zooplankton and their provinces caused by pollution, overfishing, climate change, and other shifting environmental conditions."

Ann Bucklin, Ph.D., Chief Scientist, CMarZ and Department Head of University of Connecticut Marine Sciences

Surveying the Climate of the Planet

Excess carbon in the atmosphere is frequently cited as a cause of global warming. The oceans absorb carbon through interaction of air and sea currents. Many zooplankton migrate daily through the water column, eating their own weight in carbon-absorbing phyto-plankton, which drift with the currents. As they migrate, large volumes of carbon are transported to the ocean depths. The biological redistribution of carbon away from the surface affects carbon dioxide accumulation and pH change, both of which threaten ocean ecosystems. Thus, knowing the current biodiversity of the oceans as a baseline is crucial for evaluating the effects of climate change. Such a baseline allows researchers to assess the changes, man-made and natural, that are taking place in the largest habitat on earth.

The Importance of Zooplankton

Zooplankton play an important role in the food chain, providing food for both fish and cetaceans. They also provide valuable information about global ecosystem processes and help moderate climate. Scientists study them to learn about the ocean's function as Earth's largest carbon sink and the impact that ocean acidification may have on life in the seas. For this project, net systems were specially designed to sample zooplankton from the deep sea. They were equipped with sensors to gauge water pressure, temperature and salinity that allowed scientists to determine the depth at which the organisms originated.

Scuba divers also collected specimens during open ocean dives. Among the species captured were copepods and ostracods, swimming worms, pteropods (flying snails) and pulsing jellyfish. The cruise captured and identified an astonishing fraction of the species diversity known in the Atlantic Ocean.

Analyzing DNA on a Rolling Ship

Because many of the organisms live deep, in water that is cold, dark and salty, as they are brought to the surface, they die quickly and their DNA degrades. In the interests of time and DNA quality, a decision was made to perform DNA sequencing on board. The team was able to get much better results by analyzing the DNA immediately, with no ethanol preservation required and less chance of dirty sequences. As a result, the expedition sequenced nearly 100 species onboard and 120 more on land. On-board taxonomic experts estimate that at least eight new species were identified and 16 - 20 more are awaiting identification.

Applied Biosystems' Field Service Engineer Bob Newman was dispatched to set up the instrument and instruct the team on its use. According to Robert Jennings, Ph.D., University of Connecticut, "We had already taken a thermal cycler to sea before, and since we have a 3130 Analyzer in our lab, it seemed like a natural progression. We have always been happy with its performance and portability. It gives clean sequences and longer reads. It's also easy to set up and run. The instrument adapted quite well to life on a rolling ship. We thought the buffer chambers might spill, but the Septa sealed O-rings were tight enough. Ten-foot waves were the max, and they were rare. The 3130 instrument was the only thing onboard that wasn't sea sick."

Sequencing the C01 Gene

The team sequenced the mitochondrial cytochrome c oxidase, subunit 1 (C01) gene, which has been found to provide excellent species differentiation. A C01 sequence library was established that will be used for marine species recognition. Without DNA sequence data, some species can be identified only as far as family or genus. Because the ship was equipped with satellite Internet, the staff were able to BLAST GenBank from their location off the Carolina coast.

DNA Barcoding

By using the Applied Biosystems 3130 Genetic Analyzer, scientists were able to perform DNA barcoding (a short DNA sequence used for species recognition and discovery) to distinguish new species quickly and accurately. DNA barcodes are revealing unanticipated diversity in species, in many cases by uncovering hidden variation within an existing species. DNA technology also enabled the researchers to examine the gut contents of various species to determine the food chain. Upon project completion, a baseline will exist that future generations can use to compare changes to the zooplankton and to their coastal waters.

CENSUS OF MARINE LIFE

For more information about the study and on the zooplankton identified in the expedition, visit **www.CoML.org**

The Applied Biosystems 3130 Genetic Analyzer

The 3130 Genetic Analyzer was used to perform DNA barcoding and advanced sequencing on board the vessel. It is scheduled to accompany the ship when it returns to sea in 2007.