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Deep Mapping of Teuthivorous Whales and Their Prey Fields

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14. ABSTRACT
Acoustic echosounders designed to map and discriminate organisms in the water column have primarily been deployed on ships. Because of acoustic attenuation of higher frequencies used to detect and discriminate micronekton and nekton, this has effectively restricted the range of this information to the upper water column. In an effort to overcome these range limitations by reducing the distance between the transducer and the targets of interest, dual-frequency (38 and 120 kHz) split-beam echosounders were integrated into a REMUS 600 autonomous underwater vehicle (AUV), more than doubling the range of quantitative acoustic data into the mesopelagic zone (600-1200 m). Data from the first set of deployments aimed at describing the predator-prey interactions between deep-diving cetaceans and their prey provide important information for mitigating human interactions with these sensitive species. We show that careful integration of a suite of traditional and novel tools is providing insight into the ecology and dynamics of predator and prey in the bathy- and meso-pelagic.

15. SUBJECT TERMS
Echosounder, Bathypelagic, Pelagic, Ocean, Cetacean, Beaked Whale, Risso's Dolphin, Predator-Prey, Anthropogenic Noise, Sonar, Foraging, Ecosystem, Deep-Sea, Heterogeneity, Patchiness

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List of Acronyms

AUTEC	Atlantic Undersea Test and Evaluation Center
AUV	Autonomous Underwater Vehicle
CTD	Conductivity Temperature Depth profiler
DoD	Department of Defense
GPT	General Purpose Transceiver
RECON	REmote CONtrol
SERDP	Strategic Environmental Research and Development Program
SOAR	Southern California Anti-submarine Warfare Range
TOTO	Tongue of the Ocean
UDP	User Datagram Protocol

Keywords

Echosounder, Bathypelagic, Pelagic, Ocean, Marine Mammal, Beaked Whale, Risso's Dolphin, Predator-Prey, Anthropogenic Noise, Sonar, Foraging, Ecosystem, Deep-Sea, Heterogeneity, Patchiness

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1. Abstract

1.1 Objective

In the last decade, great progress has been made in understanding the behavior and biology of many deep diving marine mammals. Much of this progress has resulted from the development of new technologies such as tagging that were supported, in large part, by various components of the Department of Defense (DoD), including SERDP. Although studies of the physical habitat of these deep-diving predators have adequately kept pace with these advancements, understanding of the available prey, a key component in the biological habitat of these animals, has not. This lag has been driven by the difficulties in studying squid (cephalopods of the order Teuthida), the primary prey of particularly deep-diving toothed whales such as sperm (*Physeter macrocephalus*) and beaked whales (Family: Ziphiidae), due to the rapid speed, relatively large size, and depth of these prey animals. Recent advances in active acoustic measurements now allow us to use this powerful remote sensing tool to assess squid behavior and distribution in water depths up to 600 meters. Teuthivores or cephalopod feeding whales, however, including sperm and beaked whales typically feed at depths of 1000 meters. The objective of this work was to develop a new platform to carry the acoustic instruments needed to assess squid and utilize this new tool to understand the foraging ecology of deep-diving teuthivores.

1.2 Technical Approach

Dual-frequency (38 and 120 kHz) split-beam echosounders were integrated into a REMUS 600 autonomous underwater vehicle (AUV), effectively doubling the range of quantitative, multi-frequency acoustic data into the mesopelagic zone (600 to 1200 meters). Data from the first set of missions in a range of conditions revealed that the AUV provided a stable platform for the echosounders and improved vertical and horizontal positional accuracy over echosounders towed by ships. In comparison to hull-mounted echosounders, elimination of ship noise and surface bubbles provided a 17 and 19 dBW decrease in the noise floor for the 38 and 120 kHz echosounders, respectively, increasing the sampling range by 30 to 40%. The extended depth range increased the horizontal resolution from 37 to 40 meters to 0.6 to 3.7 meters, enabling discrimination of individual targets at depth. The project also developed novel, onboard echosounder data processing and autonomy to enable sampling not feasible in a surface ship or towed configuration. The project demonstrated the effectiveness of a new tool for examining the biology of animals in the mesopelagic zone (600 to 1200 meters) in ways previously only possible in the upper ocean by making measurements of prey abundance, size, and distribution within known foraging habitat areas for two deep-diving marine mammal species, Cuvier's beaked whales (*Ziphius cavirostris*) and Risso's dolphins (*Grampus griseus).*

1.3 Results

Study efforts targeted habitat used differentially by deep-diving, air-breathing predators to empirically sample their prey's distributions on and off a United States (U.S.) Navy testing range, the Southern California Anti-Submarine Warfare Range (SOAR), west of San Clemente Island. Fine-scale measurements of the spatial variability of potential prey animals were made from echosounders aboard both the ship and AUV. Significant spatial variability in the size, composition, total biomass, and spatial organization of biota was evident over all spatial scales examined and was consistent with the general distribution patterns of foraging Cuvier's beaked whales observed in separate studies. Striking differences found in prey characteristics between regions at depth, however, did not reflect differences observed in surface layers. These differences in deep pelagic structure horizontally and relative to surface structure, absent clear

physical differences, change long-held views of this habitat as uniform. The revelation that animals deep in the water column are so spatially heterogeneous at scales from 10 meters to 50 km critically affects our understanding of the processes driving predator-prey interactions, energy transfer, biogeochemical cycling, and other ecological processes in the deep sea and the connections between the productive surface mixed layer and the deep water column.

The significant differences in deepwater squid that were observed in neighboring pelagic areas were consistent with the general distribution of foraging beaked whales in these areas. The study combined measurements with published information to estimate the consequences of the environment on beaked whale foraging and found that beaked whales would have a difficult time meeting their energetic needs in areas outside of SOAR, providing information for mitigation efforts. The heterogenous nature of squid in the preferred habitat of beaked whales is a key feature that appears to lead to the success of these predators, likely because of the steep costs they face to access food and limited foraging time. This highlights the relevant prey metrics that must be considered to understand the ecology of deep-diving predators and the scales at which researchers must approach these important questions.

The project explored the behavior of Risso's dolphins foraging in somewhat shallower scattering layers off Santa Catalina, California using a similar approach. Three distinct prey layers were identified: a persistent layer around 425 meters, a vertically migrating layer around 300 meters, and a layer intermittently present near 50 meters, all of which were used by individual animals tagged as part of a companion project funded by the U.S. Navy. Active acoustic measurements demonstrated that Risso's dolphins dove to discrete prey layers throughout the day and night with only slightly higher detection rates at night. Dolphins were detected in all three layers during the day with over half of detections in the middle layer, 20% of detections in the deepest layer, and 10% falling outside the main layers. Dolphins were found less frequently in areas where the shallow, intermittent layer was absent, suggesting that this layer, though containing the smallest prey and the lowest densities of squid, was an important component of their foraging strategy. The deepest layer was targeted equally both during the day and at night. Using acoustic data collected from the AUV, layers were found to be made up of distinct, small patches of animals of similar size and taxonomy adjacent to contrasting patches. Squid made up over 70% of the patches in which dolphins were found and more than 95% of those in deep water. Squid targeted by dolphins in deep water also were relatively large, indicating significant benefit from these relatively rare, physically demanding dives. Within these patches, prey formed tighter aggregations when Risso's dolphins were present.

1.4 Benefits

Application of the new echosounder AUV developed by this project resulted in great progress in understanding cetacean behavior and habitat use and improvements in predictive capabilities of these factors. Careful integration of a suite of traditional and novel tools is providing insight into the ecology and dynamics of predator and prey in the meso- and bathy-pelagic, including in geographical areas in southern California commonly used for sonar by the US Navy where significant DoD resources have been expended on biological and behavioral studies of potential effect. This advancement in providing direct measurements of the ecological context and drivers of foraging behavior and distribution is essential for effective estimation and mitigation of noise effects, including those from military sonar systems, on these deep-diving marine mammals.

2. Objective

Two important needs have been identified in the management of cetaceans sensitive to anthropogenic sound sources: 1) an understanding of the behavioral ecology of these species and 2) the development of tools and technologies to collect this data both in natural conditions and for the quantification of potential responses to anthropogenic sound. Behavioral ecology is defined the study of the evolutionary basis for animal behavior due to ecological pressures. One of the most immediate and important ecological pressures for all animals is the acquisition of food. However, we know very little about the distribution, abundance, and behavior of the prey that deep diving marine mammals rely on or how these factors affect these predators or their interactions with humans. Our first goal was to develop an effective, easily deployed, adaptable tool capable of whales and their prey field simultaneously to depths of 1200 m. Our second objective was to utilize this novel tool to increase our understanding of whale behavior and habitat use in areas of relatively well-studied important habitat that overlap with relatively intense US Navy activities, including mid-frequency active sonar, off Southern California.

3. Background

Sperm whales (*Physeter macrocephalus*) and beaked whales (spp. from the family Ziphiidae) are deep divers that feed on cephalopods (squid and octopus) (Best 1979, Santos et al. 2001) and regularly attain depths of over 1200 m with average foraging dive depths near 1000 m (Kawakami 1980, Tyack et al. 2006). While little is directly known, there is indication that these species (and particularly beaked whales) may be particularly sensitive to operational military sonars and other anthropogenic sound sources (Richardson et al. 1995, Frantzis 1998, Balcomb III & Claridge 2001). This may be because these animals use active acoustic signals that overlap in frequency spectra with some of the most powerful Navy operational signals in order to locate their prey (Johnson et al. 2004, Watwood et al. 2006), or because they may perceive these sonar signals as predatory in nature (Tyack 2009). It also appears that the deep-diving behavior of these species may make them more susceptible to man-made noise.

In the last decade, great progress has been made in understanding the behavior and biology of deep diving whales. Much of this progress has resulted from the development of new technologies such as tagging which were supported, in part, by various branches of the Department of Defense. Studies of the physical habitat of these whales have been able to keep pace with these advancements. However, an understanding of the available prey has not. Prey resources are likely to be a critical factor driving the distribution and behavior of whales and thus must be incorporated in any mitigation measures for the species. Estimating the distribution of highly mobile prey over the 1000-meter depth range of regular sperm and beaked whale dives, however, has proven a difficult task. As a result, virtually nothing was known about the behavior, vertical and horizontal migrations, response to environmental forcing, and feeding ecology of squid or the responses of deep-diving teuthivores , including Cuvier's beaked whales and Risso's dolphins included in this study, to these prey features. The acoustic characteristics of squid have recently been measured, creating a new, reliable technique for locating squid and quantifying their abundance in the field (Benoit-Bird et al. 2008). These same acoustic tools have also proven to be effective in detecting diving marine mammals, elucidating their foraging behavior and habitat use (Jochens et al. 2008, Benoit-Bird et al. 2009). However, these techniques are limited to approximately the upper 600 m of the water column when utilized from the surface using a standard research vessel. Our goal was to develop an effective, easily deployed, adaptable tool capable of simultaneously detecting whales and their prey field to depths of 1200 m and to use this tool to increase our understanding of whale behavior and habitat use in areas of important habitat that overlaps with US Navy operational areas off Southern California.

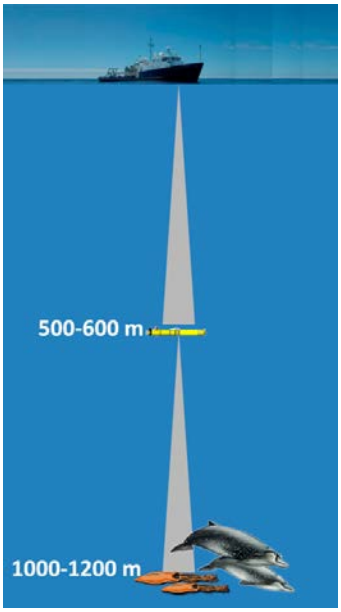

Figure 1. Ship-based hydro-acoustic sampling for squid has a range limitation of 500-600 m from the sampling platform. We proposed to develop a new tool that could autonomously sample from a depth of 500-600 m to provide acoustic data from the depth range over which beaked whales and other deep-diving teuthivores are known to forage, 1000-1200 m.

4. Materials and Methods

4.1 AUV Integration

Acoustic echosounders designed to map and discriminate organisms in the water column have primarily been deployed on ships. Because of acoustic attenuation of higher frequencies used to detect and discriminate micronekton and nekton, this has effectively restricted the range of this information to the upper water column. In an effort to overcome these range limitations by reducing the distance between the transducer and the targets of interest, dual-frequency (38 and 120 kHz) split-beam echosounders were integrated into a REMUS 600 autonomous underwater vehicle (AUV), effectively doubling the range of quantitative acoustic data into the mesopelagic and bathypelagic zone (600-1200 m). Also developed here is novel on-board echosounder data processing and autonomy to allow sampling not feasible in a surface ship or towed configuration. Details of the AUV-echosounder integration can be found in Moline et al., in press but the key details are summarized here.

In selecting an AUV platform ideal for the echosounder application, it was essential to optimize the following capabilities: 1) operational depths of the vehicle needed to be at or beyond the effective multi-frequency surface range of ship mounted echosounders (500-600m); 2) a vehicle required sufficient power to both sustain a significant range/duration and power the transducers and associated computers necessary for onboard data acquisition and processing; and 3) the vehicle needed to both accommodate the relatively large diameter of a narrow beam 38kHz transducer (48 cm) and be as small as possible for logistical ease in deployment and recovery. We selected the REMUS 600. Introduced in 2005 weighs only 250 kg with dimensions of 3.25 m by 0.3 m diameter. It has an operational depth of 600m, with a range (400km) and duration (70 hrs with a standard payload) comparable to the much larger Autosub and THESEUS (Stokey et al. 2005). Other mid-sized AUVs such as the Bluefin-12 and HUGIN possess the required depth capabilities, but have about half the range/duration capacity (Fernandes et al. 2003).
Two, off-the-shelf Simrad EK-60 (Andersen 2001) general purpose transceivers (38 and 120 kHz) were modified to fit inside the dry payload bay of the REMUS. The electronic cards on each transceiver are typically connected through a backplane that arranges the cards linearly. However, the longest dimension of the standard backplane exceeds the interior diameter of the vehicle's payload bay. We used custom backplane boards that arranged the electronics cards in an X-shape to decrease their diameter. Each modified GPT is surrounded by a custom built enclosure that allows it to mount to rails that slide into the standard REMUS guide system. These transceivers are connected through a water tight bulkhead to 1500 m depth rated 7° beam echosounder transducers (Simrad 120-7CD with a diameter of 18 cm; 38-DD with a diameter of 48 cm) mounted in a wet payload bay, forward of the electronics payload. The housings and mounts for the EK-60 general purpose transceivers were designed to allow the GPT electronics to be grounded while being isolated from the REMUS hull to allow for isolation from electrical noise and the electrically driven REMUS hull that serves as part of the REMUS leak detection system. Additionally, we undertook extensive measurements in the laboratory and preliminary deployments to identify and mitigate sources of noise in the AUV system prior to field experiments, for example, adjusting the switching frequency of the AUV's drive motor, shielding cables, installing filters on power lines, modifying internal grounding paths, isolating electronics, and altering cable positions.

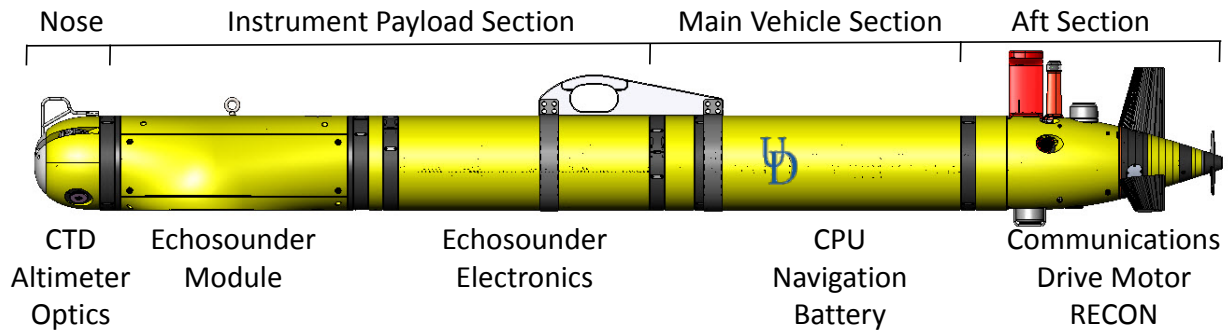

Figure 2. The REMUS 600 vehicle with a specialized Simrad EK60 echosounder payload section. The integrated vehicle includes an aft section, main vehicle section, the 2 echosounder modules and an instrumented nose section. The large red mast in the aft section is for Iridium, wireless and GPS communications. The cylinders below and behind the mast are acoustic transponders for ranging and modem communications. Vehicle length in this configuration is 4.25 m.

The echosounder transceivers were connected via Ethernet to two, PC-104 form factor computer stacks attached to the same rails as the transceivers using custom mounts and adapter plates. Each computer stack is a VersaLogic Leopard based, 2.26 GHz commercial temperature Intel Core 2 Duo processor with 4 GB RAM, dual gigabit Ethernet, and two solid state hard drives running the Windows 7 operating system. Both computers are coupled to the vehicle's network via Ethernet, allowing the computers to be viewed and controlled remotely through the vehicle's wired and wireless connections. The computers are also connected to a separate, gigabit Ethernet system with a dedicated wireless antenna and a wired port to facilitate the rapid transfer of the extensive data sets that can be acquired by the echsounders. One computer stack runs Simrad's ER60 data acquisition software along with the operating system on one hard drive and acquires data directly to its second hard drive. This computer stack also has a serial connection to the vehicle computer which provides time, vehicle depth, and pseudo GPS position which is automatically merged into the acoustic data stream utilizing existing navigation input options within the ER60 software.

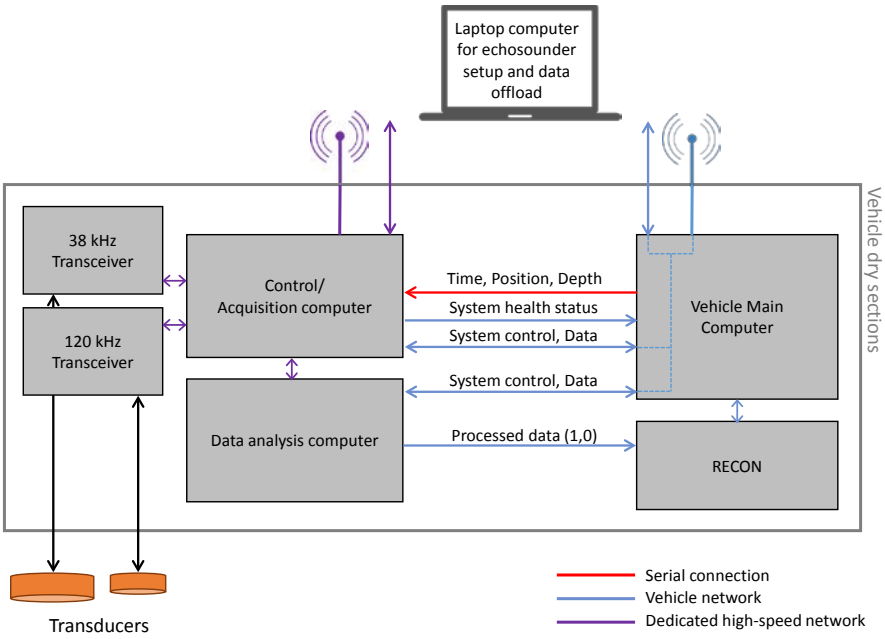

Figure 3. Block diagram of vehicle connections amongst the four computers and 2 echosounder systems inside the AUV as well as an external laptop for setting up the system and offloading data. Connections are indicated by lines with arrows showing the direction of data flow. Dotted lines indicate pass through of information. Connections include a serial link (red) that provides vehicle data to the echosounder data acquisition computer for synching with the data stream, the vehicle's Ethernet network (blue), and a secondary, high-speed Ethernet dedicated to the echosounder system (purple).

In order to validate the performance of newly integrated AUV system, we developed a series of open ocean tests. Each sequential test was designed to build on the previous effort in evaluating the performance of both the vehicle and echosounders. In all tests, the echosounders used a 1.024 ms long pulse (input power: 1000 W at 38 kHz and 500 W at 120 kHz). Testing for this system occurred on the US West Coast and began with a shallow water test in San Luis Obispo Bay, CA in July 2012. This mission followed a 4 km offshore transect, returning to shore and repeating. The mission was in depth mode operating at 4 m, designed to evaluate the basic function of the echosounders and the stability of the AUV platform. The second test in April 2013 was a deep mission off of Scripps Pier in La Jolla, CA and was designed to fly over Soquel Canyon to evaluate the acoustic range of the echosounders and operation in deeper waters. The mission profile was to navigate offshore 10 km at 10 m and then dive to 300m over the canyon.

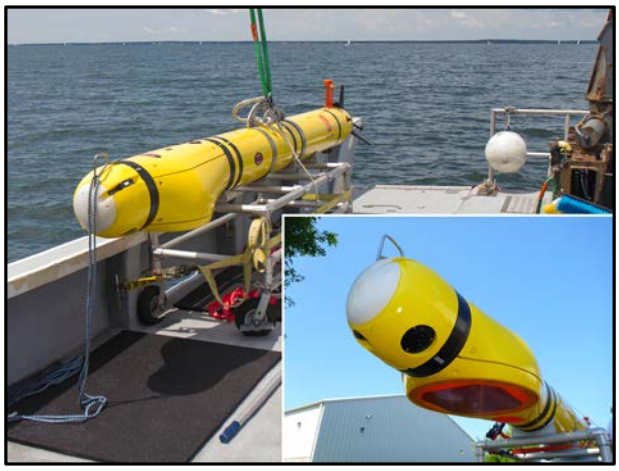

Figure 4. The REMUS 600 AUV on deck ready for deployment. The bulge in the forward wet section was to accommodate for the dual echosounder transducers (inset). The section aft houses the electronics for data acquisition and processing.

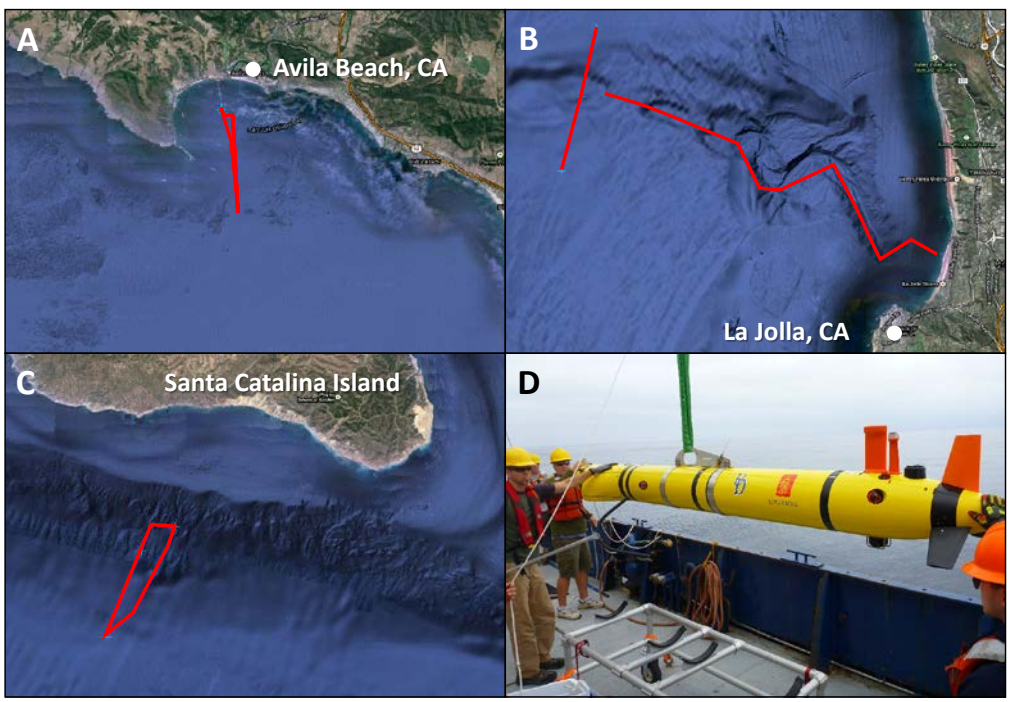

Figure 5. REMUS 600 missions demonstrating the technology. A) Test 1 shallow transect in San Luis Obispo Bay, CA. Mission length 16 km. B) Missions off La Jolla, CA. Mission on east side was Test 2, a km transect out to Soquel Canyon. The western 5 km transect was mission A, part of the primary study examining abundance and distribution of deep prey fields. C) Mission D off the shelf break south of Santa Catalina Island. Mission length was 10 km. D) Deployment of integrated REMUS 600 for mission D. After the vehicle was released it was driven away manually from the vessel via wireless and then sent the command to start the mission. On return, the vehicle would be driven manually back to the side of the ship for pick up.

These tests lead to the first intended application of the vehicle along the southern California coast and in the deep canyons in the Channel Islands between Santa Catalina Island and San Clemente Islands. These primary missions occurred in September/October, 2013 with the goal to measure the distribution and abundance deep-water prey fields (i.e. krill, fish, and squid) in this area to understand how prey affects the behavior of deep-diving whales. This is the first time mesopelagic depths have been evaluated for these sound-scattering prey using an AUV.

4.2 Vehicle Autonomy

Biological systems are dynamic in time and space, ship time is costly, and the organisms being studied often have a high disturbance potential from the sampling platforms. Sensors that measure of biological processes have also generally not kept pace with the development of physical sensors. On this last point, biological information is not obtained from a single, simple proxy, rather often the synthesis of information from a suite of sensors and some degree of sensor fusion is required. In the Echosounder AUV, a second, identical VersaLogic computer stack is connected to the vehicle's network via Ethernet. This second computer stack is responsible for processing data and providing synthesized results to the vehicle to modify navigation. As processing the echosounder data is computationally demanding, separation of data acquisition and data processing ensures the robustness of the system. The processing computer runs Echoview software (Echoview Software Pty Ltd, Hobart Tasmania, Australia) as well as a custom, stand-alone Windows-based application written in C++ that manages the software and passes processed information to the vehicle's computer via Ethernet at the frequency determined within the data processing program implemented within Echoview. Echoview provides robust near real-time analysis that can incorporate basic data processing along with tools for combination of the two frequencies of acoustic data, analysis of solitary targets, volume scattering integration, and more. These analyses are incorporated into a visually programmed "data flow" that is saved as a distinct file that can easily be replaced as analysis needs change.

The data processing and synthesis example first used in this effort targeted squid and will be describe here. Data from the two frequencies of the entire water column are processed (removal of the seafloor, correcting data depth as the vehicle dives, removal of noise, etc.) to identify individual targets. The target fields of the two frequencies are then combined and a target filter applied, here a 'squid' target filter using the known differences in the volume scattering across frequencies from a variety of shallower-dwelling squid species. The data products generated from this work flow include distributions of squid length, the target density at any given depth within range, and squid biomass. Importantly, this analysis can easily be altered to target fish, krill, or marine mammals either by the user between missions or predetermined internally. While not attempted, these multiple filters could be applied simultaneously to provide combined target scenarios if desired.

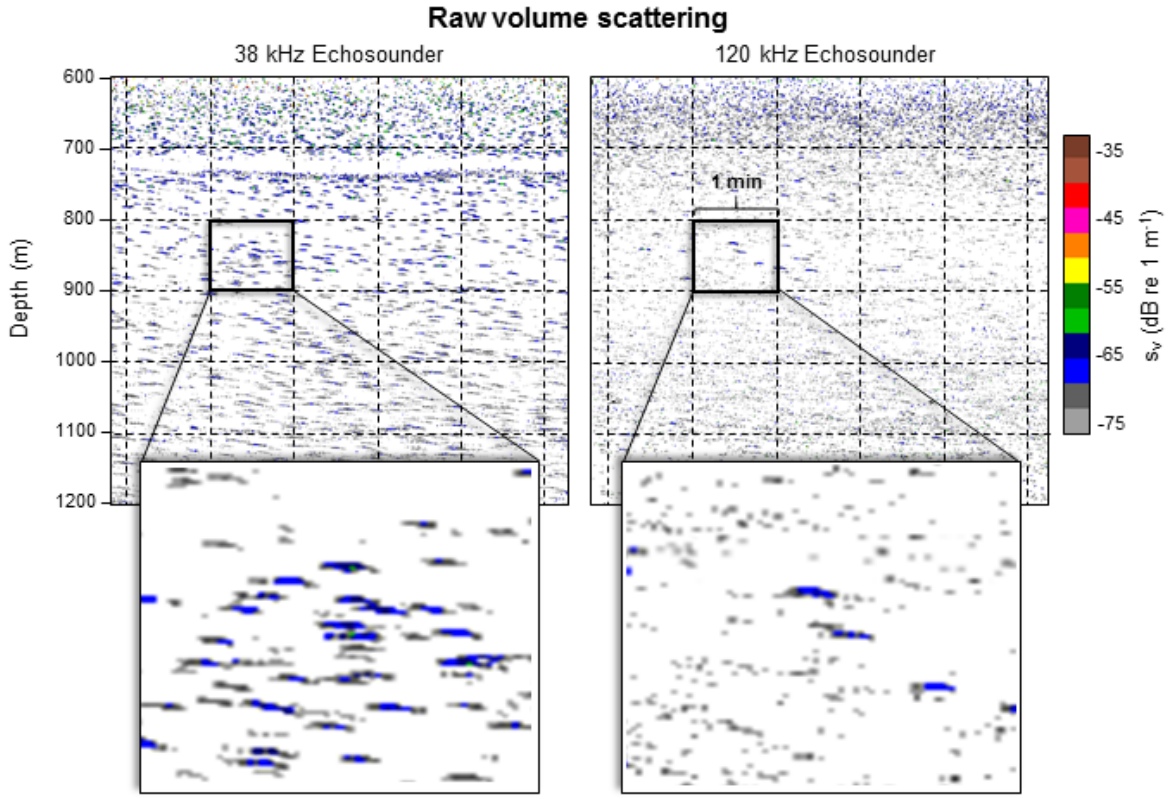

Figure 6. Example of output from the dual-frequency echosounders integrated into the AUV. Both panels show the same slice of water within an acoustic scattering layer off California. Evident is the ability of this system to discriminate individual targets and the clear differences in the raw volume scattering between the two frequencies. The difference in raw scattering allows for individuals to be classified into taxonomic categories.

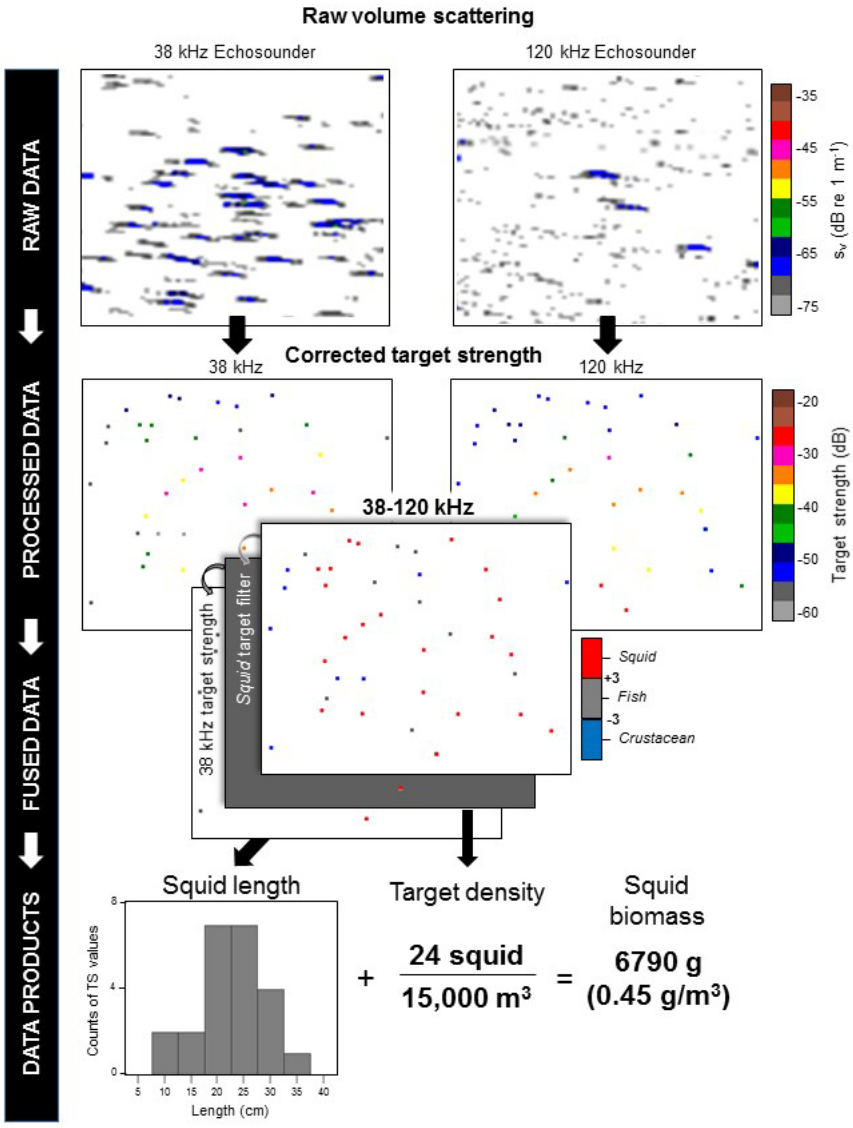

Figure 7. Diagrammatic flow of data processing, synthesis and data product generation with squid as the target organisms. From the raw data taken off California in 2013, targets are determined for each frequency and combined with a target filter to achieve a set of data products to be used in vehicle decision-making and autonomy.

Autonomy implemented thus far has been based on a simple binary signal. The key point however outlined in the introduction is that the signal is based on a processed data product. Here, after the three data products are generated, we determine a threshold for providing a positive signal. Figure 8 illustrates the decision-making threshold. Integrated over a particular time, here 1 minute, and over the full range (600m) the system identifies the target number and the size of the target. If the criteria set in the data flow for size and number (here 20 squid > 20

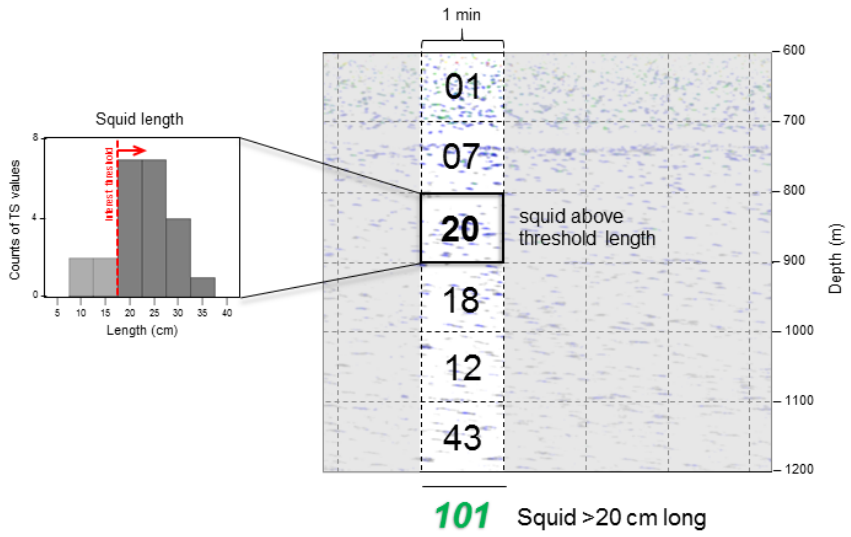

Figure 8. Example of threshold determination for autonomous decision-making based on real-time data products from data taken off California in 2013. Here, squid number and length are used to determine changes in vehicle navigation.

cm long) is met, the custom application running on the stack sends a "1" to the vehicle's RECON (Remote Control) computer, otherwise it sends a zero. A 1 or 0 flag is encapsulated and transmitted as a UDP data packet through the vehicle network. These outputs occurred within 30 seconds of the acquisition of the data, providing close to real time feedback to the vehicle. The RECON computer was programmed to respond when the UDP data packet sends a "1", taking control over from the vehicle's primary navigation computer, pausing the primary mission, and executing a secondary mission for either a set amount of time, until completing the secondary mission, or until additional sensor input met some prescribed condition.

4.3 SOAR Navy Range

We used the echosounder AUV to remotely sense and quantify the distribution of potential marine mammal prey items in deep-water areas off southern California where several species of deep-diving marine mammals are known to occur and feed. We specifically focused on areas known to be important foraging habitat for the deepest diving marine mammals, the beaked whales. These include offshore areas around San Clemente Island where considerable research on the extreme deep-diving and geographical movement of Cuvier's beaked whales (*Ziphius cavirostris*) has been conducted due to the overlap between these animals and the U.S. Navy's Southern California Anti-submarine Warfare Range "SOAR". Local information on the behaviour, distribution, and individual behaviour of beaked whales was available from visual surveys and photo-identification (Falcone et al. 2009), long-term satellite tag monitoring (Schorr et al. 2014), as well as a broadly-distributed array of monitoring hydrophones on the range tuned to detect foraging beaked whales in collaboration with the visual observation and tagging efforts and to provide long-term (monthly patterns over multiple years) acoustical monitoring of their distribution across a deep-water area of the SOAR range covering hundreds of square kilometres (D. Moretti, personal communication). This information was used to stratify our sampling within

two defined sections of this area, allowing *a priori* predator distribution patterns indicating differential habitat use to identify regions of potentially contrasting prey characteristics. We combined acoustic and direct sampling measures of biotic resources at various depth in order to quantify the variability in deepwater resources over 10 m-50 km scales and to examine the connection between surface layers and these features.

A key consideration for the active acoustic sampling design to measure the distribution and density of deep-water biota was the incorporation of what was known about the sub-mesoscale habitat use of deep foraging predators, particularly Cuvier's beaked whales, within the survey region. Recent progress has been made in understanding various life history characteristics, including habitat utilization, through applications of medium-term tags (days to months) for tracking surface locations and some aspects of diving behaviour of Cuvier's beaked whales off California (Falcone et al. 2009, Schorr et al. 2014) and passive acoustic monitoring of their species-typical echolocation clicks (Dave Moretti, pers. comm); these researches have deployed over 20 satellite-linked tracking tags on this species on the SOAR range. This underwater acoustic monitoring facility contains 172 bottom-mounted hydrophones covering nearly 1800 km^2 that are designed to track undersea vehicles but that have been used to monitor Cuvier's beaked whales and other species. Combined recent data from visual observations and encounters, tagging/tracking of individuals, and passive acoustic monitoring strongly suggests that this is an important beaked whale feeding area and that there may be preferential habitat use within it. Specifically, Cuvier's beaked whales detected on the SOAR range using these methods have historically more commonly distributed in the western portions of the range relative to eastern areas. Based on these *a priori* observations of the distribution of deep-foraging predators, we constructed a blocked sampling design to investigate prey distribution in lower use ("eastern") and higher use ("western") zones of the SOAR range, as well as a bathymetrically similar "off-range" zone immediately to the north.

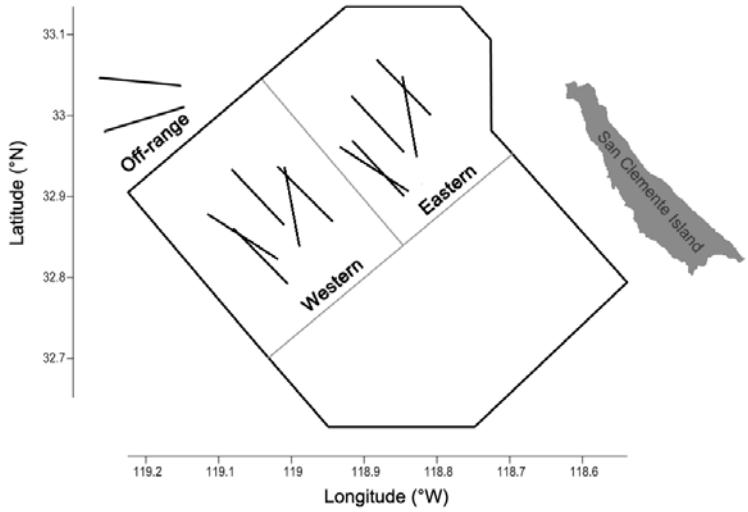

Figure 9. The locations of our sampling transects, each 10 km in length, are shown relative to the US Navy's SOAR range, outlined in black, off Southern California. A priori *Ziphius* habitat use zones are delineated by grey lines to show a 'high use' zone in the northwest quadrant of the range and 'low use' in the northeast quadrant. Zones to the north of the range are the closest similar habitats that might be used by animals if range activities displace them.

In each sampling zone, 10 km long transects were surveyed during daylight hours over 4 days in September, 2013 using the *R/V New Horizon* and the specialized REMUS AUV. Transect locations within each sampling zone were chosen to sample discrete areas that represent the general bathymetry of the region and to effectively utilize limited available time with suitable weather conditions and access to the SOAR range. In the each of the sampling zones on SOAR, five transects were conducted. The off-range zone was only sampled with two transects, averaging 1310 and 1380 in measured bottom depth on 29 September. Each transect consisted of a single CTD (Conductivity Temperature Depth) profile to a depth of 1000 m near the beginning of the survey, ship-based acoustic measurements taken continuously at a vessel speed between 1.8 and 2.6 m/s offset slightly (~100 m) from the AUV traveling along the same course at a speed of 1.8 m/s, and a depth targeted oblique trawl conducted at 1.8 m/s.

Active acoustic sampling was conducted from both a deepwater AUV and ship-based acoustic sampling to provide measures of animals throughout the water column. Ship-based echosounders included Simrad EK60s a 38 kHz (12 degree split-beam), and 70, 120, and 200 kHz (7 degree split-beams) split-beam system with transducers deployed downward looking 2 meters beneath the surface of the vessel. Each echosounder used a 512 microsecond long pulse at a rate of 1 Hz and a source level <180 dB re 1 μPa (rms). The AUV carried two, downward-looking, split-beam echosounders (Simrad EK60s) at 38 and 120 kHz (7 degree beams) and a PC104 format computer that control data acquisition. Each echosounder used a 512 μs long pulse at a rate of 1 Hz and a source level <180 dB re 1 μPa (rms). As the AUV was flown at a consistent depth of 550 m for all surveys, this corresponds to data covering 600 and 1200 m of water depth.

Acoustic scattering data from both platforms was processed using Echoview software as described in Benoit-Bird et al. (In review). From the ship based data, scattering was integrated from 5 m-600 m. Integration was conducted from 600-1200 m in the AUV data. Single targets, e.g. only one target per acoustic reverberation volume for each pulse (Sawada et al. 1993), were extracted from both the 38 kHz and 120 kHz data from both the ship (upper 600 m) and AUV (600-1200 m). For all targets identified at both frequencies (>85% of all single targets detected), the intensity of the echo at 120 kHz was subtracted from the 38 kHz intensity to provide information on target identity. These are interpreted here as specific taxonomic classes based on measurements of known targets from shallower waters as acoustic measurements from the species likely present are not available. While this has the potential to introduce, the differences between sites observed remains, despite potential errors in interpreting those differences. For example, the absolute measures of target strength were different in solitary targets consistent with squid scattering between the northeastern and northwestern range. Converting these target strengths to length relies on curves developed for other species and thus the absolute measures of length presented as a helpful too for biological thinking may be inaccurate. However, the consistent slope of relationships between length and target strength across all taxa (McClatchie et al. 2003) means that the relative difference in length between locations is unlikely to be affected by the lack of in situ measures of target strength for these species. The relative composition, target density, and integrated scattering strength were compared across sampling zone for the full water column, upper water column, lower water column, and 900-1200 m. In addition, the heterogeneity of the distribution of targets consistent with squid or fish at the deepest range of our sampling (900-1200 m) was analysed as a function of spatial scale ranging from 10 km down

to 10 m. and the effects of scale and sampling zone on the measures of distributional heterogeneity were analysed.

4.4 Santa Catalina Basin

Sampling in the Santa Catalina basin focused on habitat identified to be important for Risso's dolphins (*Grampus griseus*) by other ongoing efforts in the area. Sampling integration was similar that that described for the SOAR range, incorporating ship and AUV-based acoustics, net tows, oceanographic profiles, and visual surveys. Sampling was supplemented by animal-bourn, acoustic and behavioral recording tags (e.g., Madsen et al. 2002) placed on three individuals that overlapped with prey sampling. Given the known diving characteristics of Risso's dolphins and the depth of the seafloor, the AUV was deployed differently than at SOAR. Rather than flying well above the potential prey of interest, the AUV was flown within scattering layers identified from the ship, covering a variety of potential depths within these features throughout the study period. This allowed us to resolve characteristics of potential prey even in densely aggregated features, examining the internal structure of scattering layers in ways that have not been previously attempted.

Another unique feature of our sampling in the Santa Catalina basin was the high rate of active acoustic detections of marine mammal targets. These targets were ground-truthed visually and identified as Risso's dolphins, allowing us to examine the vertical habitat use of Risso's dolphins and the subsurface overlap between predator and prey for a large number of individuals and integrate this with the detailed observations of behavior of a few tagged individuals.

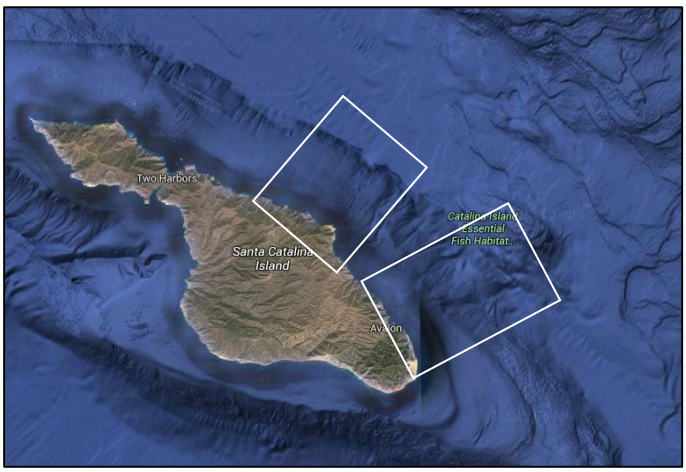

Figure 10. Sampling was conducted off the east side of Santa Catalina Island in two areas of the Santa Catalina Basin where Risso's dolphins were observed visually before and during the study period.

5. Results and Discussion

5.1 AUV Integration

Application of the complete integrated system first took place off the coast of California in 2013 with 28 mission traveling over 650 km at depths to 550m with the longest mission of ~17 hours. Flight metrics showed robustness in maintaining depth target (±0.05 m), heading (±0.1^0), pitch (±0.3^0) and roll (±0.3^0). Level flight is especially important for these acoustic sensors making measurements 600 m away from the vehicle. In these same initial tests we determined the noise floor of the integrated system and robustness in delineating single targets. Data from the first set of missions in a range of conditions revealed that the AUV provided a stable platform for the echosounders and improved vertical and horizontal positional accuracy over echosounders towed by ships. The echosounders maintained a constant calibration, regardless of deployment depth and, in comparison to hull-mounted echosounders, elimination of ship noise and surface bubbles provided a 17 and 19 dBW decrease in the noise floor for the 38 and 120 kHz, respectively, effectively increasing the sampling range by 30-40%. The extended depth range also increased the resolution of the acoustic horizontal footprint from 37-40 m to 0.6-3.7 m, enabling discrimination of individual targets at depth. Taken together, these data demonstrate an effective new tool for examining the biology of animals in the mesopelagic zone (600-1200 m) in ways previously only possible in the upper ocean.

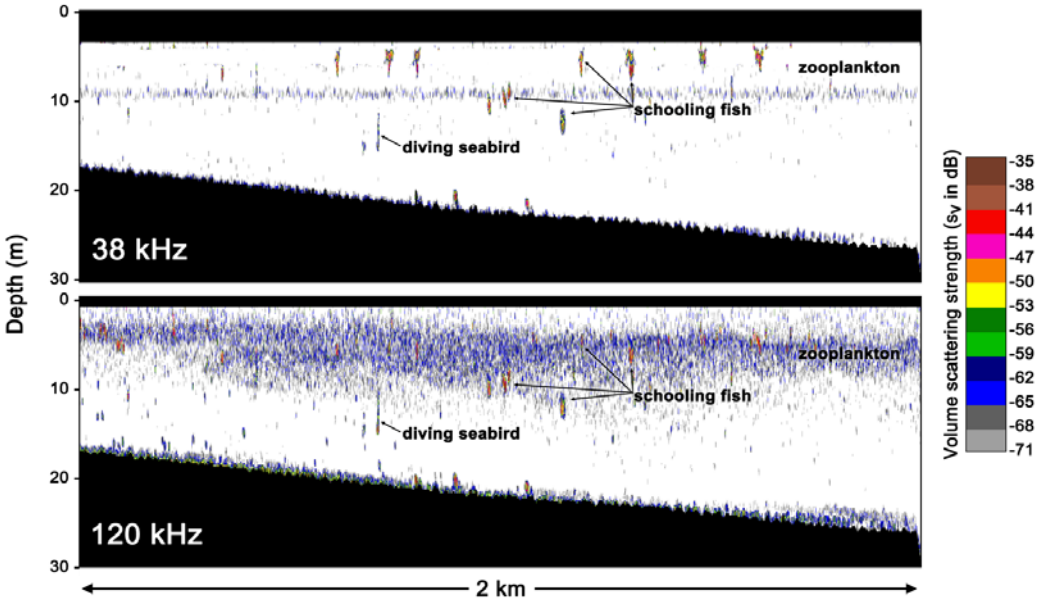

Figure 11. . Echograms from the A) 38 kHz and B) 120 kHz echosounders for a 2 km portion of Test 1. This initial deployment indicates the importance of using more than one frequency to aid in the classification of biological targets. During this mission, diving seabirds were visually observed at the surface. As in previous work (Benoit-Bird et al. 2011), echoes from a stream of bubbles leaving the plumage of a diving bird are clearly visible at both frequencies utilized. Similarly, schools of fish are detected at both frequencies while zooplankton are visible only at 120 kHz.

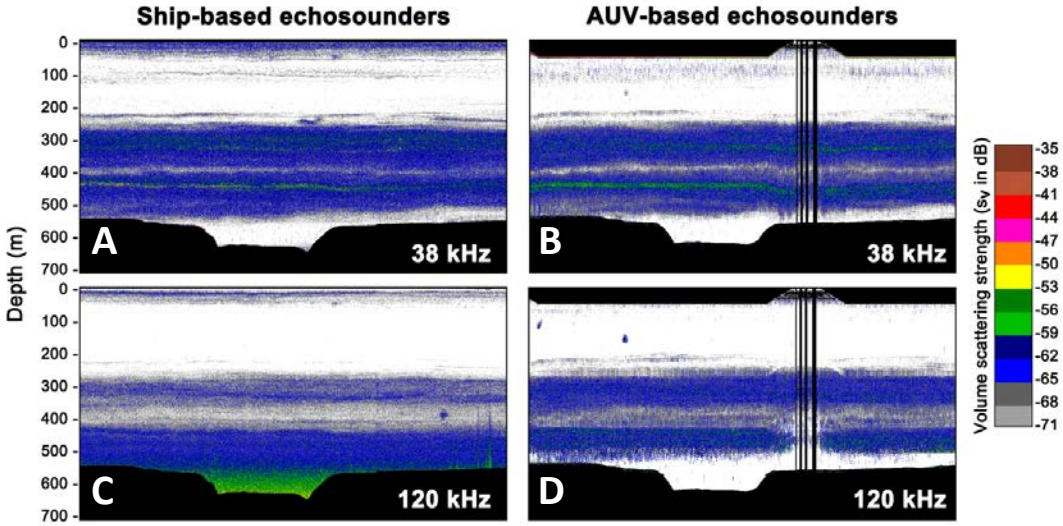

Figure 12. A 10 km long echogram from mission A surveyed by both A&C) the ship and B&D) the AUV at 38 kHz and 120 kHz off La Jolla, California. The AUV sampled from a depth of 50 m with a surfacing near the middle of the transect. Both data sets show true depth to allow easy comparison. Data from below the seafloor, areas of acoustic ring-down near the transducers, and some regions of bubble-induced data wash outs when the AUV was at the surface are shown in black. Background noise, however, is not removed. The volume scattering strengths measured are not significantly different between platforms throughout the overlapping effective ranges of each echosounder. However, one important thing to note is the area of yellow and green scattering near the depression in the seafloor in the 120 kHz data from the ship that is missing from the AUV-based data. This monotonically increasing scattering is caused by amplification of noise by the time-varying gain of the echosounder. The lower noise floor in the AUV-based echosounder shows effective range of the AUV based ech0sounder is larger than the ship-based system under typical conditions.

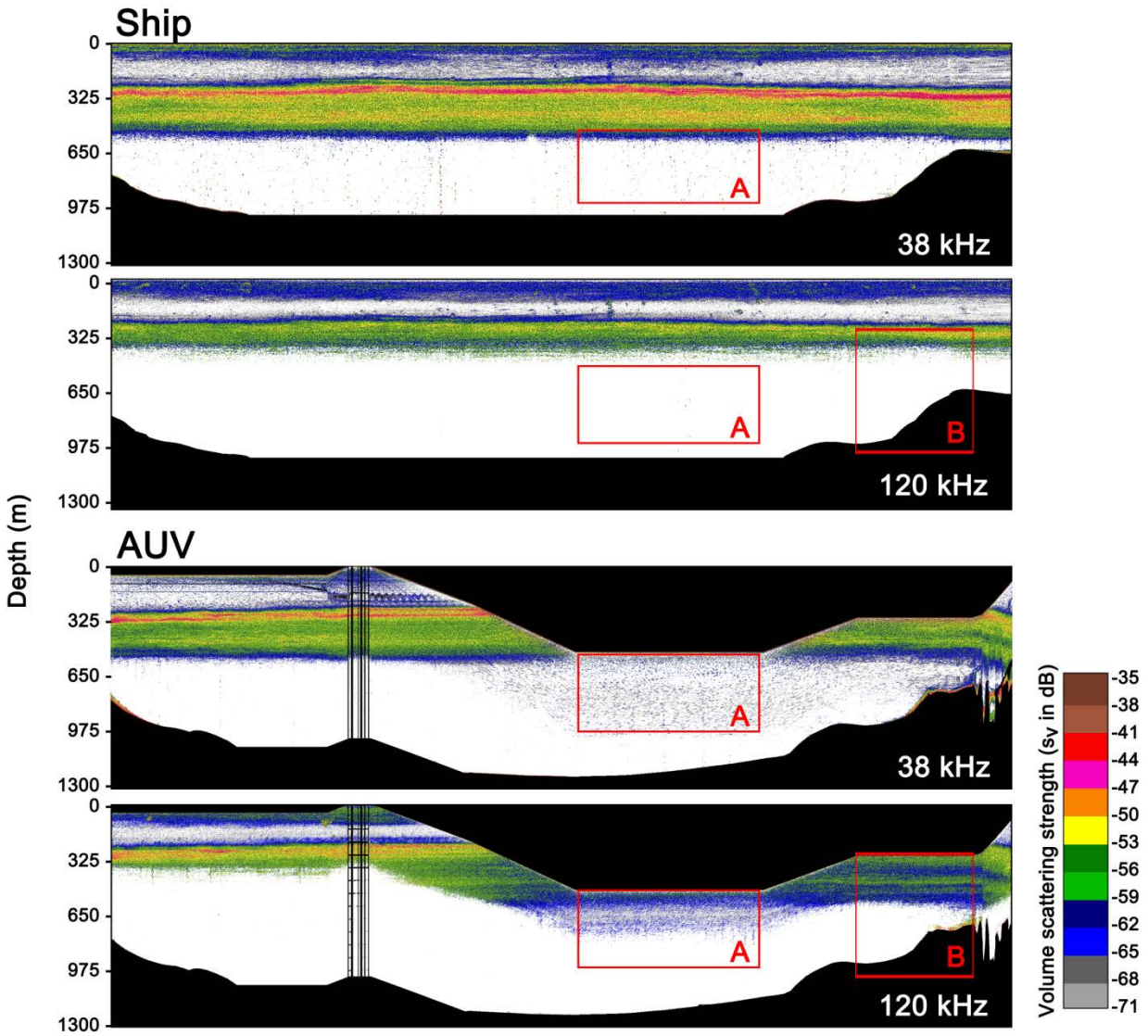

Figure 13. Echograms from mission D, a 10 km transects run in parallel by A&B) the ship and C&D) the AUV at 38 kHz and 120 kHz in California's Catalina Basin. The AUV sampled from a depth of 50 m, followed by a surfacing, then at depths of 500 m and 300 m before surfacing again. Data from below the seafloor, areas of acoustic ring-down near the transducers, and some regions of bubble-induced data wash outs when the AUV was at the surface are shown in black. Data are processed here as is typical for acoustic surveys, removing background noise resulting in the apparent loss of all but the strongest targets at great depths, particularly from the 120 kHz data. Details of these effects are shown for boxes A and B in Figure 15.

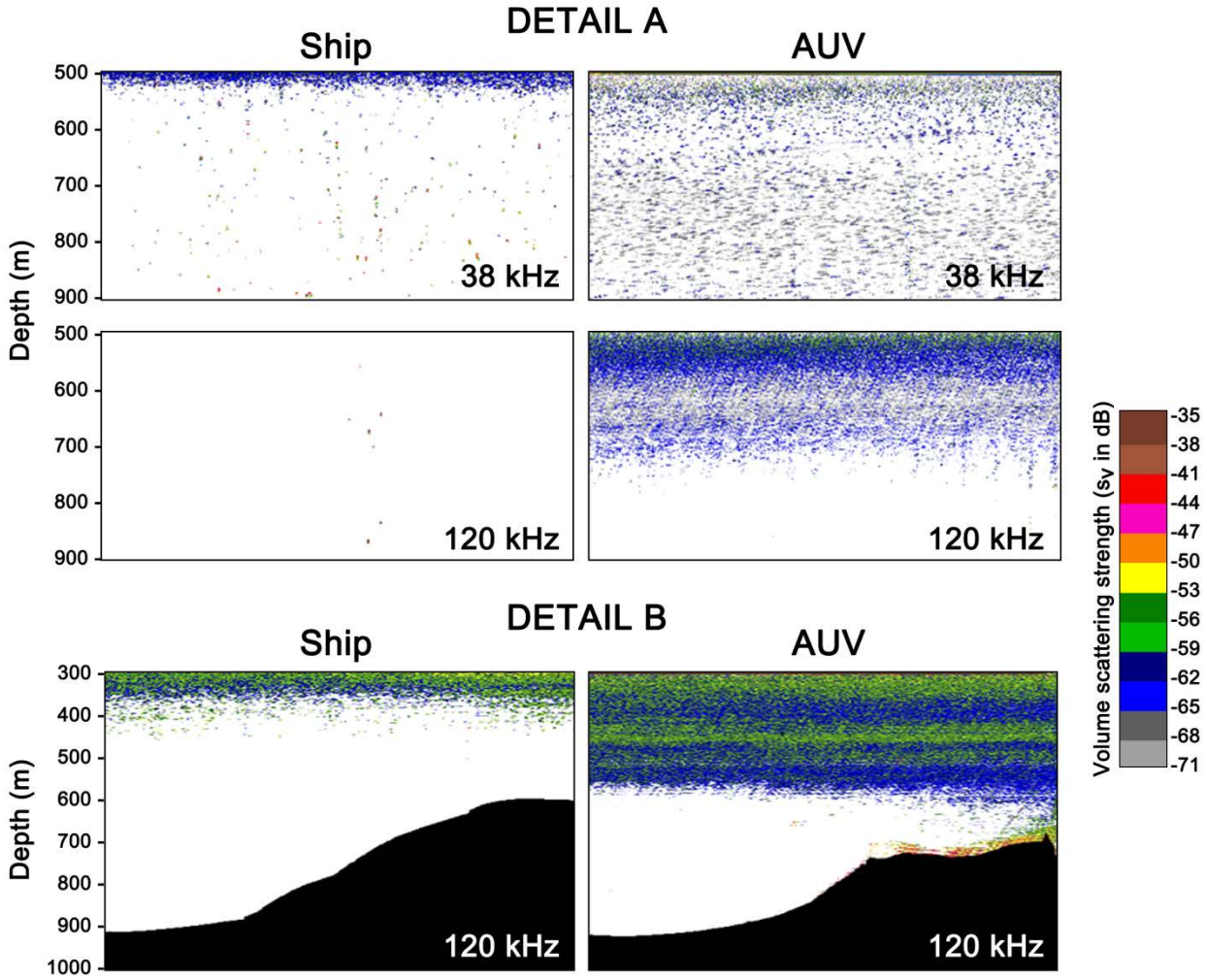

Figure 14. Details of data from A and B boxes in Figure 14 illustrate the effects of lowering the transducers on the detection of targets. In A, a diffuse layer of acoustic scattering is detectable at 120 kHz from the AUV but completely outside the range of the ship-based sensor. While a depth of 600 m is typically considered well within the effective range of the 38 kHz sensor, the AUV reveals numerous relatively weak solitary targets that are undetectable below the strong scattering layer detected by both systems. Panel B shows that even at a depth of 350 m, the ship-based 120 kHz sensor fails to detect a moderately intense scattering layer that is clearly observed by the AUV-based sensor.

5.2 Autonomy

During all missions, we logged the real-time processed data and compared this with the results of more intensive analysis supervised by an experienced operator. Autonomous detections of squid were linearly correlated with those made post-hoc. However, the detection rates were significantly lower for the autonomous approach. This is a result of the stringent requirements we utilized in the autonomous classifier as well as limitations in processing power due to power and computing limitations inside the vehicle.

We conducted a single mission to test the autonomy response from the real-time data processing as described above. The vehicle was programed to start the fine grid mission and complete the entire mission prior to additional commands or continuation of the larger mission. The vehicle approached from the east and began the secondary mission. The grid was designed as an expanding box starting in the center with 100 m separation between boxes, growing to 200 m, and finally one at 500 m separation with dimensions of the largest box of 750m. On completing the mission, the vehicle surfaced, acquired a GPS position and corrected for the offset by ocean currents ending the secondary mission in the northeast quadrant (intended box – pink) and continuing with the original mission in the center and then follow the trackline to the west. As the vehicle does not have inertial navigation and only relies on compass heading, ocean currents pushed the vehicle off course to the south. Even though the mission data was not symmetrical, the mission goal of sampling over smaller scales was achieved and demonstrated in challenging field conditions. Data collected here showed a number of spatial features in the organism distributions that would have otherwise not been possible by other means, both in responsiveness and of a depth beyond the ships echosounder range.

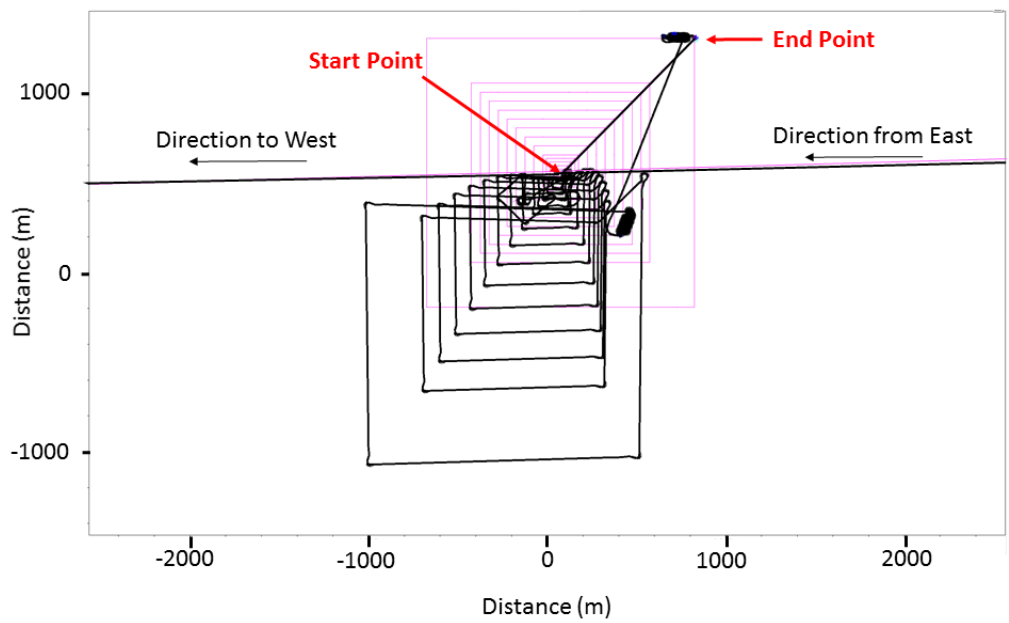

Figure 15. Mission designed to resolve smaller scale distributions of organisms that was run on 10 July, 2015 in the Bahamas. The intended track is in pink with the actual track in black. Offset is due to currents (see text).

5.3 SOAR Navy Range

We targeted habitat used differentially by deep-diving, air-breathing predators to empirically sample their prey's distributions off southern California. Significant spatial variability in the size, composition, total biomass, and spatial organization of biota was evident over all spatial scales examined and was consistent with the general distribution patterns of foraging Cuvier's beaked whales (*Ziphius cavirostris*) observed in separate studies. Striking differences were found in prey characteristics between regions at depth, however, did not reflect differences observed in surface

layers. The incoherence between the surface and the deep reveal that using proxy measures or conventional ship-based sensing systems would provide misleading information about the foraging habitat for beaked whales. In addition, these differences in deep pelagic structure horizontally and relative to surface structure, absent clear physical differences, change our long-held views of this habitat as uniform. The revelation that animals deep in the water column are so spatially heterogeneous at scales from 10 m to 50 km critically affects our understanding of the processes driving predator-prey interactions, energy transfer, biogeochemical cycling and other ecological processes in the deep sea, and the connections between the productive surface mixed layer and the deep water column.

The distribution and density of prey is likely critically important in the habitat usage and foraging behavior of deep-diving marine mammals. We combined our measurements of the distribution, size, and abundance of prey with published information to estimate the consequences of the environment on beaked whale foraging, finding that beaked whales would have a difficult time meeting their energetic needs in areas outside of US Navy testing range off Southern California, providing information to inform both potential mitigation efforts and the potential longer-term or population level consequences of repeated disturbance from relatively high-quality foraging areas. The heterogeneous nature of squid in the preferred habitat of beaked whales is a key feature that appears to lead to the success of these predators, likely because of the steep costs they face to access food and limited foraging time. This highlights the relevant prey metrics that must be considered to understand the ecology of deep-diving predators and the scales at which we must approach these important questions.

5.4 Santa Catalina Basin

In Santa Catalina Basin, we identified three distinct prey layers: a persistent layer around 425 m, a vertically migrating layer around 300 m, and a layer intermittently present near 50 m, all of which were used by individual tagged Risso's dolphins. Active acoustic measurements demonstrated that Risso's dolphins dove to discrete prey layers throughout the day and night with only slightly higher detection rates at night, in contrast with the previous view that Risso's dolphins are primarily nocturnal predators.

Dolphins were detected in all three layers during the day with over half of detections in the middle layer, 20% of detections in the deepest layer, and 10% falling outside the main layers. Dolphins were found less frequently in areas where the shallow, intermittent layer was absent, suggesting that this layer, while containing the smallest prey and the lowest densities of squid, was an important component of their foraging strategy. The deepest layer was targeted equally both during the day and at night.

Using acoustic data collected from the AUV, we found layers were made up of distinct, small patches of animals of similar size and taxonomy adjacent to contrasting patches. Previously, our view of scattering layers was as horizontally extensive features made up of a variety of species mixed together. Variance in acoustic scattering was interpreted as variation in the density of organisms. Our net tow data agreed with this fnding as catches were consistent throughout the entire basin region sampled. However, the acoustic data from the AUV flown within these scattering features show that instead of being made up of mixed species, each layer is organized into smaller aggregations or shoals of one type and size of organism. The relative species

composition of these aggregations matched those made in net tows when assessed at the same, relatively large (multiple kms) spatial scale, resulting in consistent catches throughout the region. Each patch was typically smaller than the horizontal resolution of the ship-based acoustic systems due to the effect of range. Identification these aggregations would not be possible without the use of the novel AUV technology.

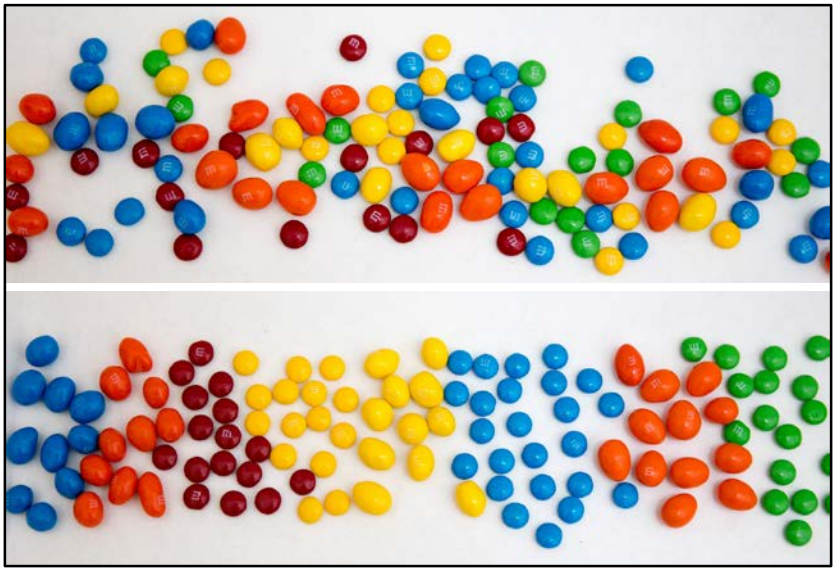

Figure 16. Visualization of the distribution of taxonomic groups (different colors) and sizes of organisms that make up acoustic scattering layers. We previously assumed that the animals in these features were mixed (top). However, data from the echosounder AUV revealed that layers are instead made up of small shoals of one type and size of organism adjacent to each other (bottom).

The distribution of organisms made it possible for us to identify the type of prey that surrounded individual, diving Risso's dolphins detected by the AUV system. Risso's dolphins were highly selective with patch compositions surrounding them quite different than the surrounding region. Squid made up over 70% of the patches in which dolphins were found and more than 95% of those in deep water. Squid targeted by dolphins in deep water were also relatively large, indicating significant benefit from these relatively rare, physically demanding dives. Within these patches, prey formed tighter aggregations when Risso's dolphins were present, supporting the interpretation that these prey aggregations are indeed schools with highly coordinated behavior that may be driven by the need to defend themselves against predation.

6. Conclusions and Implications for Future Research/Implementation

In this effort, we developed a new tool for studying biology beneath the ocean's surface. Test applications of the echosounder AUV we developed showed that the realized effect was to more than double the range of quantitative acoustic data into the meso and bathy-pelagic zones of the ocean, providing the opportunity to describe the biotic environment for important and sensitive marine mammal species that feed at these depths. We also developed novel on-board echosounder data processing and autonomy to allow sampling not feasible in a surface ship or towed configuration. The package is capable of meeting all of the sampling objectives we identified in project planning. providing an effective new tool for examining the biology of animals in the mesopelagic zone (600-1200 m) in ways previously only possible in the upper ocean.

The custom modifications that were made to commercially available software (Echoview) including real-time export of synthesized data and the ability to offset the data by the depth of the vehicle dynamically have been integrated fully into the latest software release and are thus available for other applications. These tools are already being in other projects by projects at Oregon State University and the British Antarctic Survey. Building on the autonomy approach developed here, a team at Oregon State is developing an autonomous vehicle that can use inputs from multiple sensors, including acoustics, to modify sampling – a great step forward in autonomous sampling of complex biological systems that would not be possible without the developments undertaken as part of our SERDP funded effort. Kongsberg, the parent company of both Hydroid and Simrad, is working on plans to market a fully integrated echosounder AUV. Echosounder technology has advanced considerably in the time since the start of this project and thus the payload will be quite a bit smaller and lower power. However, many of the advances we made and the challenges we overcame during the integration, which was a partnership with both companies, will guide their efforts.

The greatest challenges for the work we present is not in the hardware or software, but in the lack of controlled measurements that would aid interpreting the data that result. As in previous work, it is difficult if not impossible to adequately ground-truth the rapidly swimming prey of deep-diving predators at the scales at which we observed significant variation using traditional tools. This was part of the impetus for the development effort we conducted but it does limit the way the data is interpreted. A second limitation is the lack of in situ measurements of target strength of known individuals. This second limitation is one that could be addressed post-hoc, allowing additional interpretation of the data already collected. We are currently working to develop new approaches incorporating other deep platforms and imaging to obtain target strength measurements from individual animals at depth that can be taxonomically identified and measured in situ. If successful, an approach like this could also serve to groundtruth future sampling efforts.

Applications of the new tool we developed with support from SERDP to address the ecology of two marine mammal predators demonstrate the utility of the tool while providing key new information on the basic biology and ecology of these sensitive species in areas of high relevance to DoD. We chose beaked whales and Risso's dolphins as our study subjects. Both are primarily teuthivores (squid eaters) with overlapping habitat that overlaps with areas of regular US Navy operational activity and rhus regularly exposed to mid-frequency active sonar. However, beaked

whales feed considerably deeper than Risso's dolphins, allowing us to attempt different approaches to sampling tailored to the relevant habitats. Beyond specific results relevant to these species, discussed above, this work shows that the careful integration of a suite of traditional and novel tools can provide previously unavailable insight into the ecology and dynamics of predator and prey in the meso- and bathy-pelagic.

These kinds of ecological contextual data to understand the baseline diving and distribution of protected marine mammal species and the nature and potential fitness consequences of disturbance from military sonar operations has current and significant relevance to the Navy's ability to meet its environmental compliance requirements in order to train. Our demonstration of its effective use in southern California provides critically needed data to inform these assessments. Clearly, additional studies are needed to evaluate potential seasonal variability in the prey distributions measured in a single season here. Additional studies should also consider additional areas of the SOAR range that are differentially utilized by beaked whales, based on subsequent satellite tag and passive acoustic monitoring. Finally, relatively nearby off-range areas that have recently been deemed no-sonar transmission areas based on negotiations in litigation over sonar use would be interesting areas in which to conduct similar predator-prey assessments for comparison with SOAR range areas that will continue to have (or may have additional) active sonar operations.

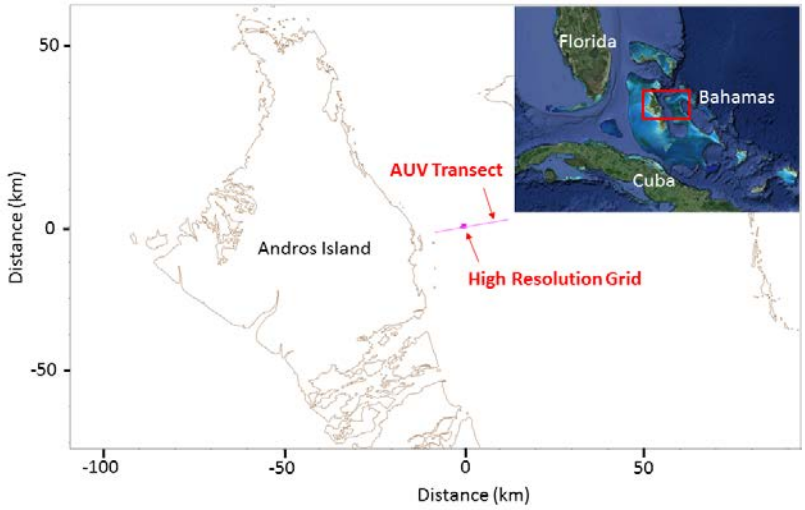

Figure 17. The Tongue Of The Ocean area where the AUV was tested in the Bahamas in July, 2015. Highlighted is a single AUV transect and the location where the high resolution grid was run to demonstrate the system autonomy.

The technology and approaches utilized here are already being transitioned to additional applications to marine mammals. With funding from the U.S. Office of Naval Research, our team recently partnered with the Bahamas Marine Mammal Research Organisation to complete sampling within the Tongue of The Ocean (TOTO) (Figures 17 and 18), Bahamas in July, 2015. Here we completed 22 missions over 11 days with over 700 km underwater time. These

missions like the previous ones off California were designed to quantify distributions of whale prey (fish, squid, and crustaceans) in the mesopelagic (200 - 1,000 m) in conjunction with visual and passive acoustic surveys for the mammals themselves. This effort builds not only on the technology we developed under our SERDP (Strategic Environmental Research and Development) program, but on the successful sampling design at SOAR, using information collected by the Navy's passive acoustic monitoring to let the animals themselves aid in providing a statistically powerful sampling design. Data analysis of the active acoustic information at depth has begun with a focus on potential prey abundance, density, and distribution, prey clustering at scales ranging from 10 m to tens of km, and individual prey identity and size inspired by parallel analyses at SOAR, off California. Our goals are to identify differences in prey resources between the range and a remote, off-range habitat, between areas within the range, and to estimate the potential losses whales suffer when they move off range. Integration of these results with those obtained from this effort will highlight the key features across sites and related species that drive the success of deep diving predators and will provide information critical to managing human impacts on these species.

Figure 18. Our sampling at AUTEC utilized a blocked design to examine prey as a function of historical habitat use by beaked whales. Sampling included four, 30 km total length v-shaped transects in each of 5 zones, a high use on range habitat (1), a low use on range habitat (2), two areas of refuge immediately adjacent to the range (3, 4), and an area with remarkably different beaked whale demographics but undescribed differences in prey (5).

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Appendix A: Scientific and Technical Publication List

Moline, M.A., Benoit-Bird, K.J., Robbins, I.C. & O'Gorman, D. "Integration of scientific echosounders with a dynamically adaptable autonomous platform to extend our understanding of animals from the surface to the bathypelagic". Journal of Oceanic and Atmospheric Technology, 32:2173-2186.

Moline, M.A. & Benoit-Bird, K.J. "Sensor Fusion and Autonomy as a Powerful Combination for Biological Assessment in the Marine Environment" Robotics, In press.

Benoit-Bird, K.J., Southall, B., & Moline, M.A. "Predator guided sampling reveals biotic structure in the bathypelagic" Proceedings B, In press.

Southall, B.L., Benoit-Bird, K.J, Moline, M.A., & Moretti, D. "Deep-water biological heterogeneity affects potential energy gains for foraging beaked whales" In review.

Benoit-Bird, K.J. & Moline, M.A., "Schooling within sounds scattering layers in the ocean" In preparation

Benoit-Bird, K.J., Southall, B.L, Moline, M.A., Arranz, P., Tyack, P. "Predator-prey dynamics in the mesopelagic: odontocete foraging ecology and anti-predator behavior of prey" In preparation