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SyPRID sampler: A large-volume, high-resolution, autonomous, deep-ocean precision plankton sampling system

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ABSTRACT

The current standard for large-volume (thousands of cubic meters) zooplankton sampling in the deep sea is the MOCNESS, a system of multiple opening–closing nets, typically lowered to within 50 m of the seabed and towed obliquely to the surface to obtain low-spatial-resolution samples that integrate across 10 s of meters of water depth. The SyPRID (*Sentry* Precision Robotic Impeller Driven) sampler is an innovative, deep-rated (6000 m) plankton sampler that partners with the *Sentry* Autonomous Underwater Vehicle (AUV) to obtain paired, large-volume plankton samples at specified depths and survey lines to within 1.5 m of the seabed and with simultaneous collection of sensor data. SyPRID uses a perforated Ultra-High-Molecular-Weight (UHMW) plastic tube to support a fine mesh net within an outer carbon composite tube (tube-within-a-tube design), with an axial flow pump located aft of the capture filter. The pump facilitates flow through the system and reduces or possibly eliminates the bow wave at the mouth opening. The cod end, a hollow truncated cone, is also made of UHMW plastic and includes a collection volume designed to provide an area where zooplankton can collect, out of the high flow region. SyPRID attaches as a saddle-pack to the *Sentry* vehicle. *Sentry* itself is configured with a flight control system that enables autonomous survey paths to low altitudes. In its verification deployment at the Blake Ridge Seep (2160 m) on the US Atlantic Margin, SyPRID was operated for 6 h at an altitude of 5 m. It recovered plankton samples, including delicate living larvae, from the near-bottom stratum that is seldom sampled by a typical MOCNESS tow. The prototype SyPRID and its next generations will enable studies of plankton or other particulate distributions associated with localized physico-chemical strata in the water column or above patchy habitats on the seafloor.

1. Introduction

Distributions of zooplankton in deep water and processes controlling these distributions and their dynamics remain poorly known, in part due to the challenges of sampling these systems with adequate spatial precision and sample size. Towed plankton-net sampling systems have been in use in oceanography since the 1970s to map the depth-stratified distribution of pelagic organisms with simultaneous collection of information about the physico-chemical environment (Wiebe et al., 1976, 1985). The MOCNESS (Multiple Opening–Closing Net Environmental Sensor System) has been a gold standard for decades and was recently used extensively by the international Census of Marine Zooplankton program (Bucklin et al., 2010). MOCNESS has also been used for distributional studies of deep-sea larvae at hydrothermal vents (Mullineaux et al., 1995) and methane seeps (Arellano et al., 2014). For a typical full-water-column sampling program, the MOCNESS is towed obliquely from the lowest safe limit of bottom approach (typically 50–100 m above bottom) to 100 or 200 m below the surface. A large (0.25–10 m^2, depending on the system) net opening ensures that a large volume of water (up to thousands of cubic meters, depending on towing speed and time a net is left open) is processed through each net, but each sample is necessarily integrated over large vertical and horizontal distances (Fig. 1A), obscuring information on finer-scale spatial distributions and processes. Because nets produce bow waves, some strongly swimming animals, including crustacean larvae, may be able to detect and avoid the net opening. Furthermore, the towed character

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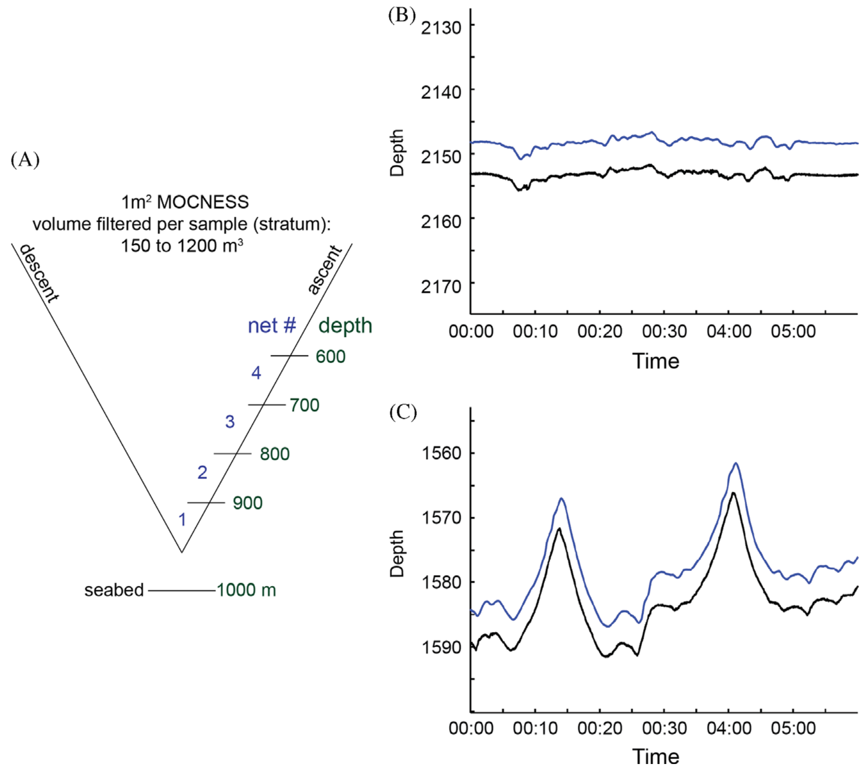

Fig. 1. Comparison of MOCNESS and *Sentry* abilities to sample depth strata. (A) Oblique water-column sampling mode of MOCNESS. Illustrates the absence of sampling capability close to the seabed (< 50 m) and the broad depth strata over which plankton abundance and distribution is integrated in any given net; from Wiebe et al. (2006). (B) *Sentry* 322 bottom tracking with SyPRID, flat terrain (< 5-m relief). (C) *Sentry* 326 bottom tracking with SyPRID, more rugged terrain (up to 25-m relief). (B) and (C): black line, seabed depth; blue line: *Sentry* depth.

of the net system precludes the possibility of sampling effectively the near-bottom (< 50 m) strata. To complement MOCNESS sampling, net-based sampling methods for deep-submergence vehicles (e.g., *Deep Tow* and *Alvin*) have been developed and deployed in multiple settings (Wishner, 1980; Wishner and Gowing, 1987; Kim and Mullineaux, 1998; Metaxas, 2011, among others).

In addition to MOCNESS and similar opening-closing net systems, a variety of other plankton sampling systems have been used to build our understanding of plankton distributions, including imaging systems such as the Video Plankton Recorder (Davis et al., 1996), the Underwater Vision Profiler (Stemmann et al., 2012), and the Lightframe On-Sight Keyspecies Identification (LOKI) system (Hirche et al., 2014). For near seabed systems, *in situ* large-volume (e.g., ~41,000 L) plankton pumps and sediment/larval traps are a proven method of sampling zooplankton, including larvae, and have the valuable capability of time-series sampling (Beaulieu et al., 2009). Prior results with larval traps at chemosynthetic ecosystems suggest that abundances of larvae are significantly higher within 5 m of the seafloor than 50 m above the seafloor (Mullineaux et al., 2005, 2013, CM Young, pers. obs.).

Autonomus Underwater Vehicles (AUVs) have the potential to sample plankton with precision relative to 3D space, but to date, sample volumes have been low. The SUPR pump sampler (Breier et al., 2009) with the AUV *Remus* were successful in sampling abundant larvae from surface waters in coastal areas (Govindarajan et al., 2015). The low pump rate ($\sim 2\,l\,min^{-1}$) did not sample a volume large enough to capture any larvae on extended *Sentry* surveys in deeper waters, where larvae are more scarce (at altitudes of 20 m, 5 m, 3.5 m, 2 m, and 1.2 m above bottom; Kaiser, Young, Van Dover, pers. obs. from R/V *Atlantis* AT26-15).

There remain critical gaps in our plankton sampling and environmental sensing capabilities, including precision sampling at variable, narrow, and spatially constrained depth strata in conjunction with a suite of remote sensing and sensor packages. There is also a need for near-bottom sampling in regions of chemosynthetic seeps and vents, cold-water corals, sponge gardens, and other patchy benthic habitats, where larval densities are reported to be maximal (Baker et al., 2010). For these and other benthic ecosystems, including cold seeps and coral and sponge gardens, our understanding of how populations are maintained will benefit from knowledge of details of larval distributions in space and time and with regard to geochemical and geophysical characters of water strata (Young et al., 2012).

Our goal was to design a large-volume plankton sampler compatible with the autonomous underwater vehicle *Sentry* and other deep-submergence assets. *Sentry* routinely conducts mission-specific surveys with precision horizontal and vertical navigation to within 5 m of the seabed, and with co-registered geochemical, geophysical, photographic, and acoustic sensor data collection. *Sentry* missions 248, 249, and 251 in the Gulf of Mexico (R/V *Atlantis* AT26-15, CL Van Dover, Chief Scientist; May 2014) demonstrated a new survey capability for precision, three-dimensional flight at 120 cm above the seabed. This capability pre-adapted *Sentry* to attempt plankton-sampling missions with a new type of plankton sampler in the critical, high-larval-density regions near the seafloor in chemosynthetic and other benthic habitats, and subsequent missions demonstrated the low-altitude terrain-following capability of the Sentry/SyPRID system in flat (< 3 m relief) and in more rugged (> 25 m relief) terrain (Fig. 1B,C). In this paper, we summarize the design of a novel deep-ocean plankton sampler,

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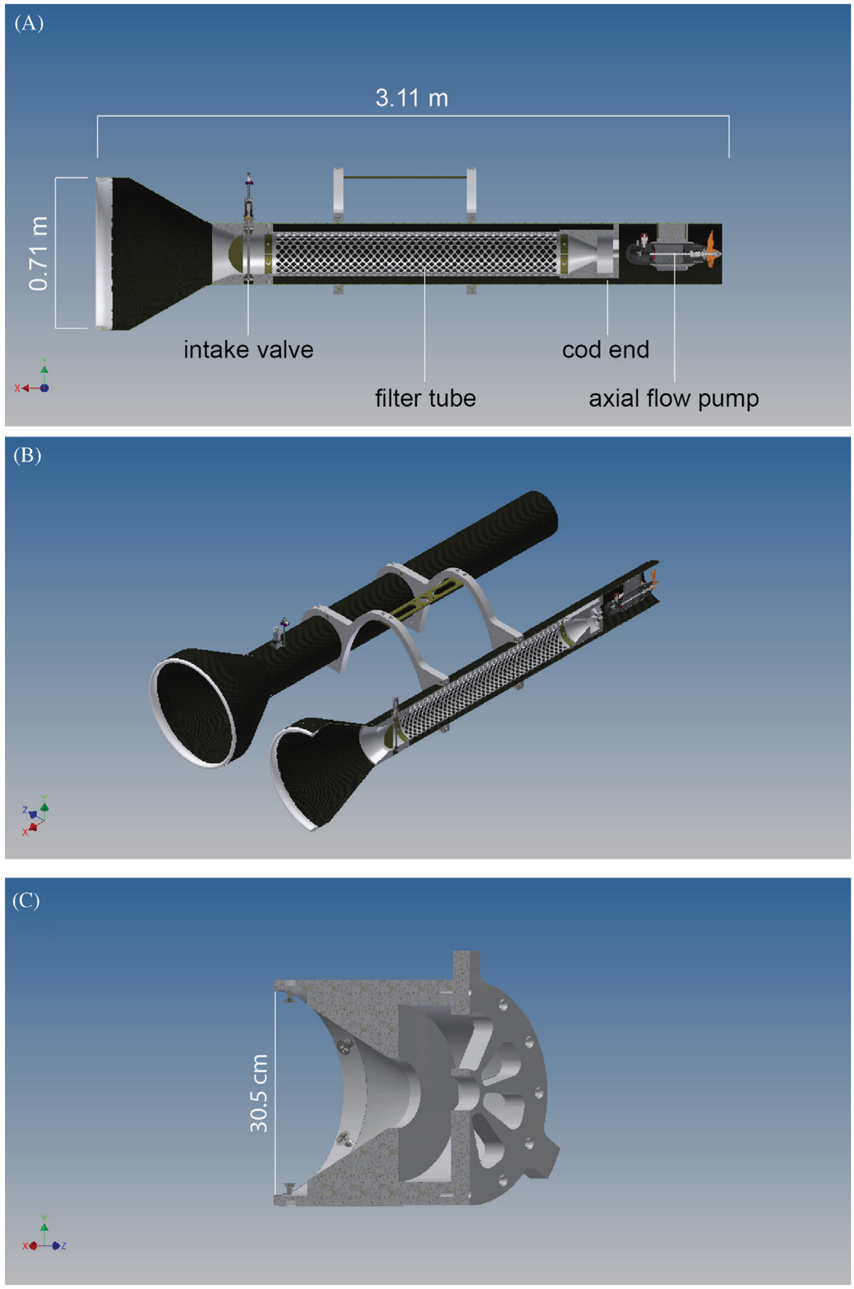

Fig. 2. SyPRID 3D models. (A) Side view of a single unit with cut away to show interior view of intake valve, filter tube, cod end, and axial flow pump. (B) Double unit and saddle mount bracket. (C) Section view of cod end and sample filter plate.

present outcomes from a proof-of-concept deployment at 2160 m above a chemosynthetic seep site off the US Atlantic margin, and identify modest improvements that could be made to the prototype.

2. Design

2.1. Design strategy

We initially aimed for a net system because adaptation of a net to *Sentry* was conceptually simple: net systems require minimal electrical power and can be designed to be nearly neutrally buoyant. Nets would also provide similarity of method for comparisons to the historical record of MOCNESS samples. Further, net sampling is sufficiently gentle to collect intact meroplanktonic larvae and holoplankton for morphological and genetic studies. A pump system was considered less desirable because the power demand and weight requirement of a pump that would generate adequate sample volume would reduce the survey footprint of a *Sentry* mission and limit payload for simultaneous characterization of environmental parameters, habitat, and macrofaunal distribution. There was also concern that the force generated by pumping would damage delicate plankton as they impinged on a collecting surface.

Key specifications for a *Sentry* net system included a sample volume of >700 m^3/h per net, 150-μm mesh with cod-end collectors for larvae (changeable nets for other types of plankton), mission-programmed and acoustic-command-control of opening and closing of nets at depth, and a flight control suite capable of bringing *Sentry* to within 1.5 m of the seafloor.

During the design research phase, which relied heavily on Keen (2013), it became evident that the dimensions of a net required to meet the sample volume specification (6-m long and nearly 1 m in diameter) were impractical for *Sentry* in a near-bottom regime where entanglement was likely. The design strategy shifted to concepts used in drainage ditches and other applications to achieve high flow rates using an axial flow pump to move water through a tube.

To avoid a bow wave that would shed organisms, flow into the opening of the tube system has to be greater than the flow external to the system, so that small organisms will be drawn inward. Likewise, it was desirable to have the inlet well forward of any thruster wash, so the sampling inlets were positioned forward of the leading edge of *Sentry*, ahead of the worst of the vehicle boundary layer effects when the impeller pumps are functioning, and well ahead of the area believed to be significantly disturbed by

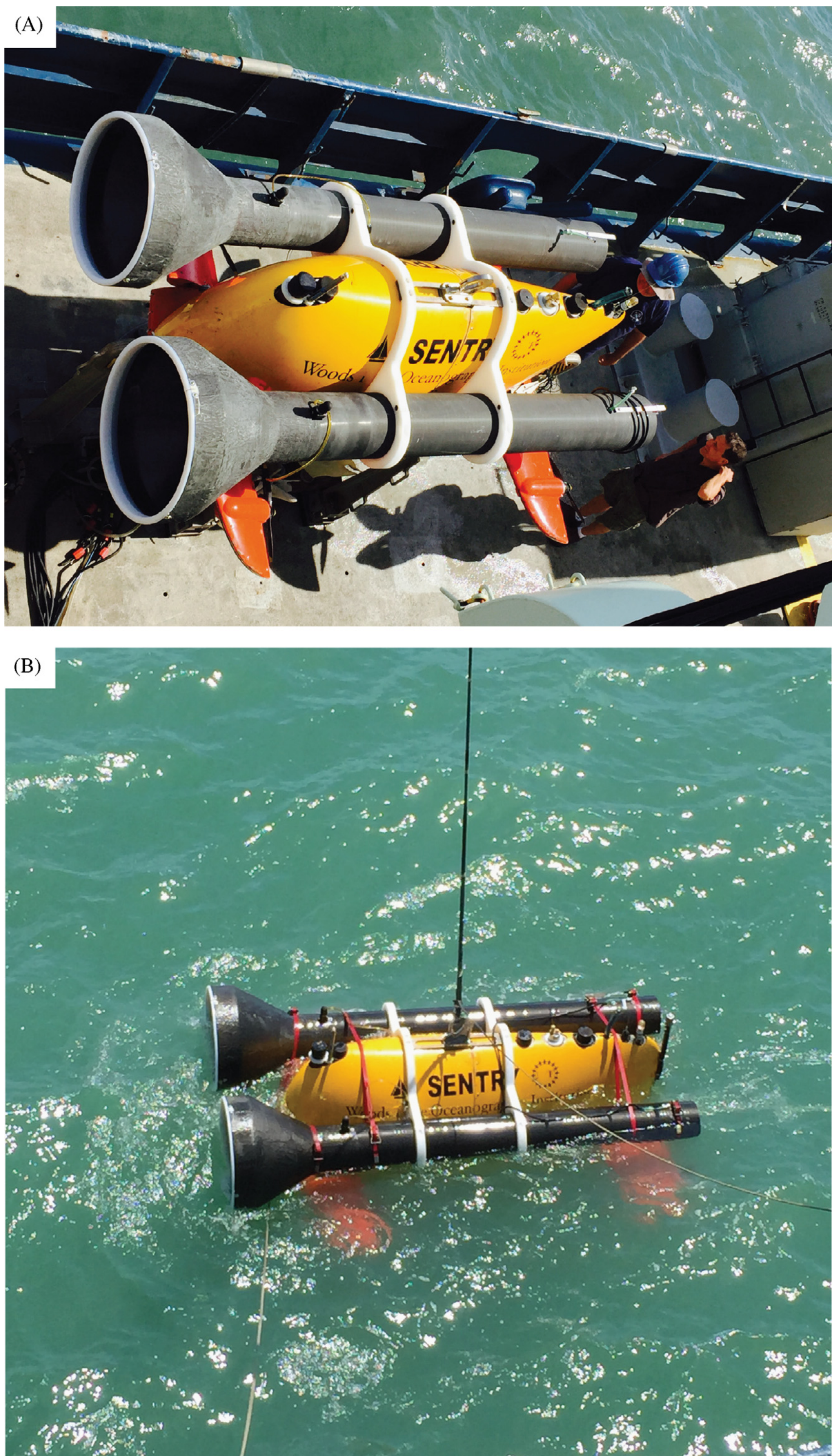

Fig. 3. SyPRID installed on *Sentry* with saddle brackets. (A) On deck in cradle. (B) Test dunk prior to sailing.

the thrusters. Although *Sentry* is capable of moving at near zero velocity in low current environments, the specified sample sites required the ability to deal with currents of up to 0.20 m s^{-1}. A challenge was to determine the optimal flow velocity within the tube given a nominal forward vehicle velocity of 0.25 m s^{-1} (0.5 kts).

An analytical study indicated that for a filtration surface using Nitex Seafarer 150 μm mesh net, the filter area should be 1 × to 1.5 × the area of the mouth opening, with < 1 psi across the filter surface. In an empirical study, the time it took to empty a water-filled PVC tube across a 150 μm filter mesh at 1 psi was used to determine an optimal flow velocity of 0.5 m s^{-1} in the tube. This modeled velocity is greater than the vehicle velocity of 0.25 m s^{-1}, as required to prevent a bow wave, and would deliver a 1500 m^3 h^{-1} flow rate. This assumes a uniform pressure differential across the entire mesh, which is likely close to the case at the low velocities in question, but cannot be proven short of substantial modeling or empirical evidence. In practice, the velocity in the large diameter inlet to the tube is believed to be > 0.25 m s^{-1}, but < 0.5 ms^{-1}, with a flow rate on the order of 1000 m^3 h^{-1}, though this number represents estimates separately derived from hydrodynamic principles and a relatively *ad hoc* measurement at the surface. Neither finite element modeling, nor empirical measurement techniques have been applied to confirm sufficient flow to eliminate bow wave.

2.2. Design features

The *Sentry* Precision Robotic Impeller Driven (SyPRID) sampler is a paired system with each sampler a tube-within-a tube design (Fig. 2), secured on the upper portion of the AUV *Sentry* using a saddle-pack harness of Ultra-High Molecular Weight (UHMW) plastic (Fig. 3). Overall length of the sampler is 3.11 m. The outer carbon-fiber composite tube is flared (0.71 m diameter) at the inlet and tapers to a cylinder (0.31 m diameter) sized to contain the axial flow pump and to be accommodated by *Sentry*. There is an in-line valve in front of the filter tube (Fig. 4A) to prevent contamination of samples at the surface and during ascent and descent when it is in the closed position. The filter tube (UHMW plastic) has a cross-sectional area equal to the area of the inlet and is lined along the entire inner wall by Nitex Seafarer 150 μm mesh, which is the standard mesh size available on MOCNESS systems (Fig. 4B). The cod end is a hollow truncated UHMW cone (Fig. 4C), with the narrow end opening into a sample collecting chamber with a flat filter surface (Fig. 4D). A custom designed, direct-drive thruster of the same type used for *Sentry* propulsion with a commercially available propeller serves as the axial flow pump at the

Fig. 4. SyPRID components. (A) In-line valve, (B) filter, (C) filter detail, (D) cod end, (E) sample mesh, and (F) axial flow pump.

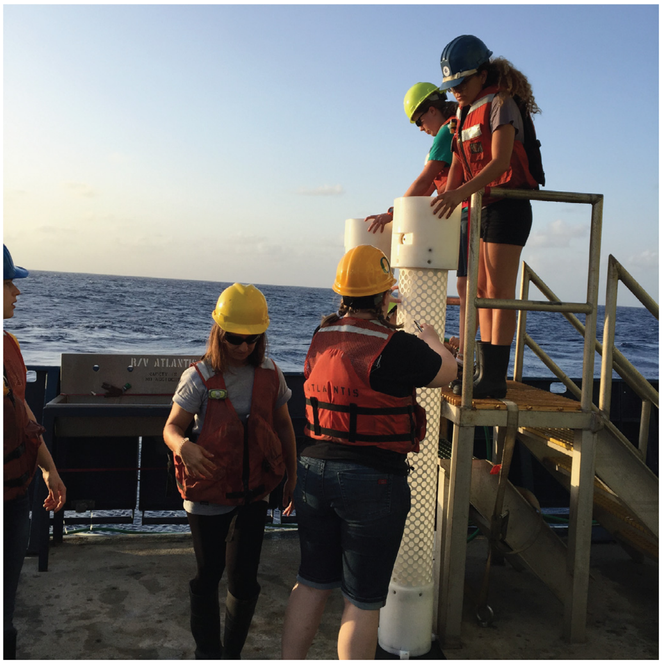

Fig. 5. SyPRID sample processing. Filter and cod end are washed down, then filter is removed, cod end is removed, and the sample filter is washed into a sample container.

aft end of the system and distal to the sample mesh (Fig. 4E). The internal cylinder and cod end are removable for sample processing (Fig. 5). The SyPRID sampling tubes are independently controllable, allowing for two replicate or two independent samples per *Sentry* mission.

Sentry flies over the seafloor using four thrusters and four wings that are actuated in pairs (fore and aft). This allows for exceptional maneuverability. When properly ballasted, *Sentry* is nearly neutrally buoyant, giving it a thrust-to- weight ratio sufficient to run at a very high angle of attack in a highly stalled regime. Missions very close to the seafloor are possible by flying *Sentry* in an unstalled regime except when a bottom impact is imminent—then the vehicle moves to the stalled regime. In effect, this is the underwater equivalent of transitioning rapidly and frequently between a fixed wing aircraft and a helicopter. The vehicle takes advantage of each regime as the situation requires. Stalled-unstalled regime-switching enables very low-level flight in flat and low-relief terrain despite minimal forward velocity, and it preserves mission duration. Previous photographic work very close to the seafloor has shown little to no evidence of sediment resuspension unless physical contact is made with the bottom.

Sentry's bottom-following capability uses a proportional/derivative (PD) controller, with altitude provided by the same Doppler Velocity Log (DVL) sonar system that assists in vehicle navigation. Altitude is used to set a goal depth and the PD controller is applied to the depth. Depth is a very precise, robust, and high-update-rate measurement, enabling effective altitude control even at very low altitude. Obstacle avoidance is provided by forward-looking beams of the DVL sonar system and, if required, is supplemented by forward-looking multibeam sonar.

Modem communications can be used to confirm the functionality of the SyPRID sampler impellers. While *Sentry* is in principle an autonomous vehicle, it does incorporate two low bandwidth (128b/30 s or 2 kb/30 s) acoustic modems. During initial testing, the modem was used to receive telemetry from both *Sentry* and the SyPRID sampler, allowing for incremental steps toward more aggressive (lower altitude) flight characteristics within a single dive.

3. Verification deployment

SyPRID was deployed during R/V *Atlantis* AT29-04 (CL Van Dover, Chief Scientist, July 2015) to the Blake Ridge North seep area (32.505142N, −76.196501W) on *Sentry* Dive 322 (9–10 July 2015). Plankton was sampled at ~2150m along ten 100-m long, E-W track lines and two N–S track lines spaced at 10 m intervals (1.2 km total, Fig. 6). *Sentry* began sampling at an altitude of 60 m and, via the acoustic modem, flight controls were gradually enabled to bring flight to 5 m above the seafloor. *Sentry* then flew at an altitude of 5 m (SyPRID intake altitude of 6 m) during an ~6-h period. Impeller RPMs and current were logged throughout the dive and confirmed the operation of the impellers during sampling. The position of the sample isolation valves (open/closed) was not sensed. Comparison of current (directly proportional to torque) with RPMs is sufficient to confirm the open or closed state of the valve while the impeller is running, but yields no information about the valve state when the impeller is off.

With a conservatively estimated flow rate of 500 $m^3\,h^{-1}$ through each sampling chamber (based on design parameters), this corresponds to a total sample volume on the order of 6000 m^3 (3000 m^3 per chamber) at 6-m altitude. This is nearly 2.5 × the volume of an Olympic swimming pool and roughly 5 to 6 × the maximum volume reported for a 1-m^2 MOCNESS sample across a 100-m-deep stratum in a typical oblique tow (Wiebe et al., 2006).

Sentry collected simultaneous temperature, optical backscatter, oxygen, oxidation–reduction potential, and magnetometer data during the mission (Fig. 7). Sensor data indicated that the temperature was uniformly ~3 °C with an average 296 μmolar O_2 (~75% saturation) at the collection depth. The water temperature was 3 °C at 5 m altitude, 3.5 °C at 80 m altitude, and there was elevated optical backscatter (turbidity) at the 5 m depth. Redox anomalies (dORP/dt < 5.0*std dORP/dt) were evident in the SE quadrant of the survey path, consistent with an otherwise undocumented site of active seepage.

Once on deck, starboard and port filters and cod ends were removed from the SyPRID sampler and washed with filtered seawater (10 μm) to collect the plankton sample onto the cod-end filter (Fig. 5). Samples were rinsed into collecting bins using filtered seawater and stored at 4 °C. The sample recovery process was reasonably swift, on the order of 30 min.

SyPRID samples were sorted under dissecting microscopes and larvae were photographed under a Laborlux K (Leitz) compound microscope fitted with an Optronics CCD camera, using Picture-Frame imaging software (Optronics). The port sample contained 39 larvae of 16 morphotypes plus juvenile echinoderms that had undergone metamorphosis either in the collector or water column. Quality of the specimens, including unarmored, soft-bodied larvae and larvae with shells, was excellent (Fig. 8), with most larvae appearing undamaged and alive. A mechanical failure (slipping of a shaft coupling on the inlet valve) could have caused surface contamination in the starboard collector. The samples from the starboard sampler were not enumerated or identified for this reason. On subsequent deployments, once a repair was made, both samplers only opened at the sample depth, with no surface contamination.

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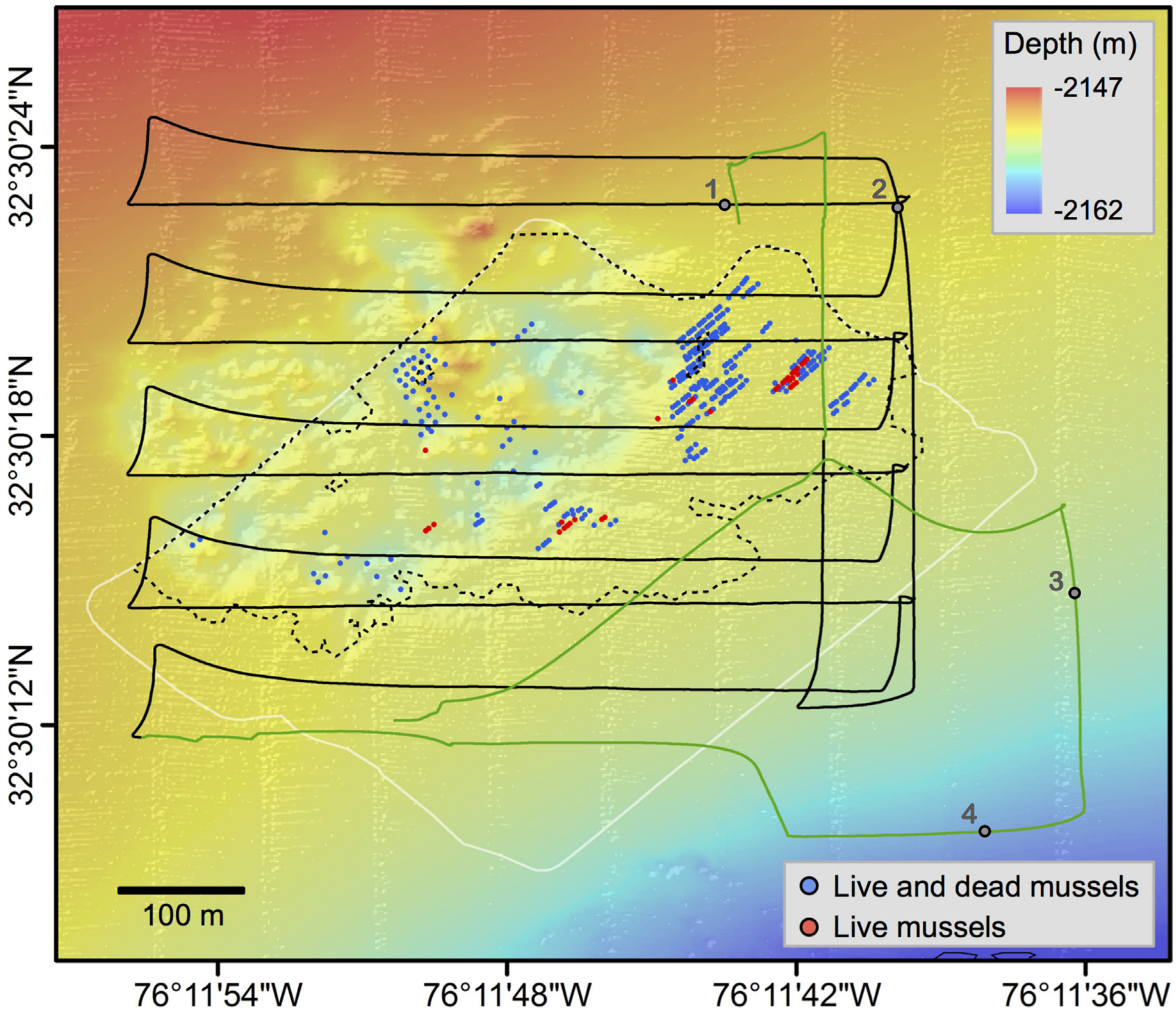

Fig. 6. Latitude/longitude plot of *Sentry*/SyPRID 322 survey track. Post-processed navigation. Black track: altitude 5 m; green track: altitude 80–5 m. Numbers indicate locations of redox anomalies (see Fig. 7). The survey track is underlain by *Sentry*-generated, high-resolution bathymetry (1-m grid) together with benthic coverage of mussels (red and blue circles) and the area occupied by clams (black dashed line) generated from 2012 *Sentry* photosurveys. The full extent of the near-saturation 2012 photosurveys, including non-seep habitats, is outlined with a white line. Bathymetry and photosurveys were collected during E/V *Okeanos Explorer* EX1205L1, CL Van Dover, Chief Scientist (Brothers et al., 2013).

4. Discussion

In studies of larval dispersal and connectivity in patchy habitats, the combination of larval data from SyPRID samples and co-registered geochemical and physicochemical data, benthic habitat maps, and planktonic community composition data simultaneously or serially collected by *Sentry* will provide previously unobtainable insights into distributional patterns of larvae of benthic invertebrates and other planktonic form with respect to bottom features and water column characteristics. SyPRID provides alternative approaches for studying pelagic systems, e.g., following isobaths, adaptive sampling near redox anomalies, etc., with correlative environmental data from its partner *Sentry*.

Design, materials, and construction costs of the SyPRID prototype were < $100,000. While SyPRID was successful in sampling plankton in its verification deployment, modest improvements will increase scientific utility substantially. Near-term proposed improvements include:

- Modeling of flow conditions using finite element analysis and installation of flow-rate sensors. The most accurate mechanism may be to install an Acoustic Doppler Velocimeter (ADV) near the inlet. The ADV would provide real-time flow-rate data.
- In its prototype configuration, the position of the SyPRID intake valves (open/closed) during a mission is unknowable. The valves were commanded to move to a specific position, but sensors were not installed to confirm proper positioning. This made it difficult to be certain that the intakes were fully closed during ascent and descent and fully open at sample depth. Several positioning sensor options are readily available and straightforward to install.
- The off-the-shelf propellers used on the impeller are not well matched to the application. They are propellers intended to operate in open water, not as impellers. A custom impeller design would allow the use of a smaller direct-drive thruster, reducing weight and enabling *Sentry* to carry additional sensors to contribute to the synoptic view of the area that is sampled.
- Additional software development in the flight controller would allow *Sentry* to remain stationary over a specific target while running the SyPRID sampler, as in the case of adaptive sampling in response to a chemical signal. This would require incorporating the thrust from the impellers into the onboard vehicle

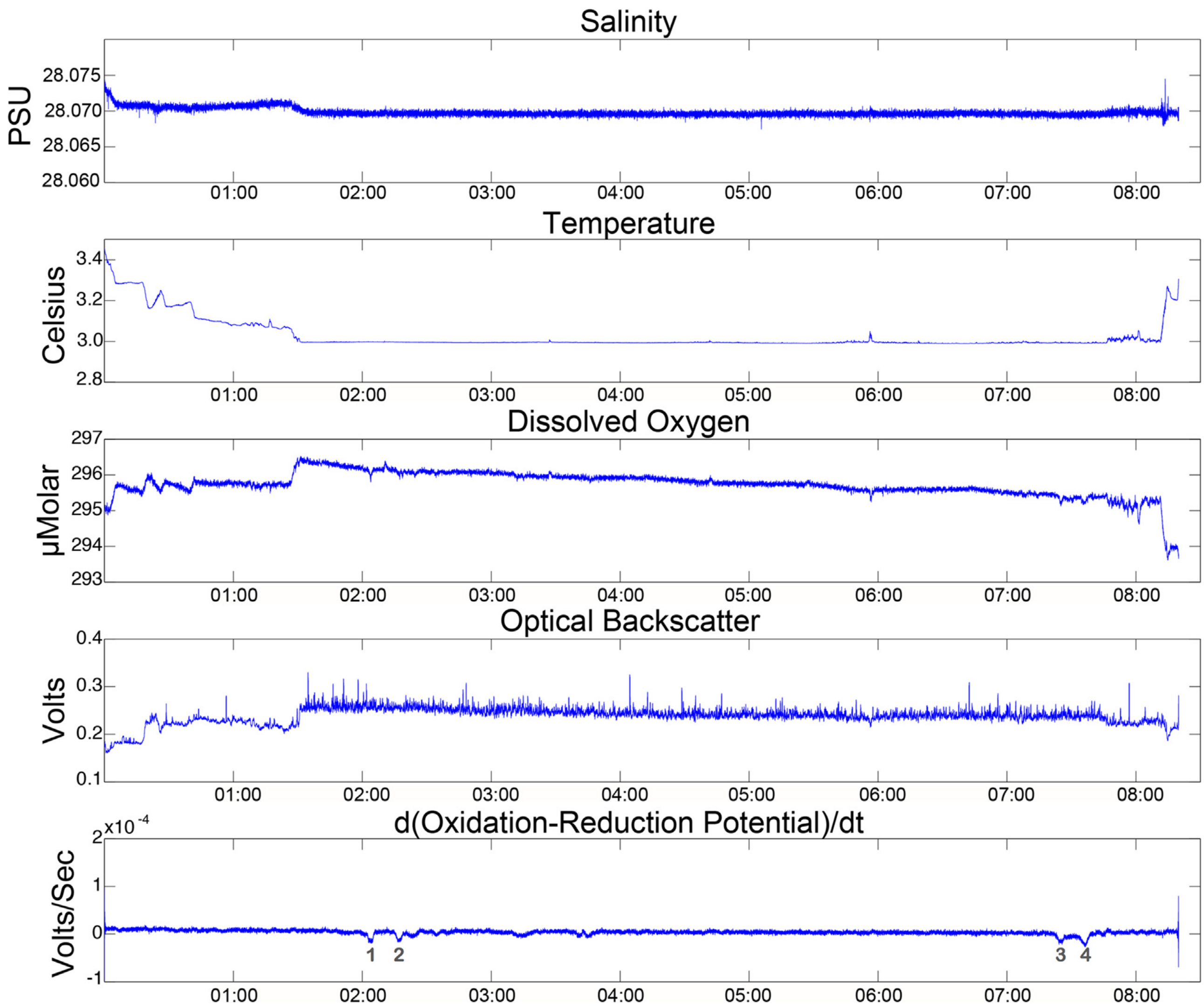

Fig. 7. Time-series plots of *Sentry* sensor data. Temperature (°C), dissolved O_2 (μmolar), optical backscatter (V), redox anomaly 'events' [*d*(Oxidation–Reduction Potential)/*dt* ($V\,s^{-1}$)].

dynamic model used for thruster allocation, and would likely require reverse operation of at least some vehicle thrusters. Enforcing a net positive flow through the impeller is straightforward when vehicle velocity is zero, provided that an accurate upper bound can be placed on current velocity, which is possible in most of the deep ocean. This would enable additional styles of precision studies of the spatial distribution of larvae and zooplankton.

- The SyPRID sampler is likely to make an effective sampling platform for particles other than larvae. Identification of these sampling needs and of appropriate filter design and tuning of pump rates should enable new directions for science in other fields.
- On most non-SyPRID missions, *Sentry* carries a high-resolution digital still camera (DSC). At present, this camera must be removed during SyPRID missions to provide space and payload for the additional sonar needed to operate *Sentry* lower than 4 m altitude. This is not a fundamental limitation, but requires some redesign of the *Sentry* payload bay to overcome. Simultaneous operation of the camera and SyPRID will provide an even more synoptic view of the environment from which the larvae are sampled.

Innovation in sampling and sensing is prevalent in the field of zooplankton research. The SyPRID system described here takes advantage of major strides in autonomous vehicle design, including command and control, and it steps outside the box of towed-net filtration systems. While MOCNESS and similar towed net systems will continue to play an important role in studies of pelagic zooplankton distributions, SyPRID and its follow-on generations will enable tests of long-standing and emergent hypotheses regarding processes that control the distribution of plankton and planktonic productivity. Salient issues in zooplankton research include understanding the formation, persistence and significance of zooplankton hot spots, the role of zooplankton in biogeochemical cycling, processes that control of biodiversity and biocomplexity in zooplankton communities, and predictive capabilities for zooplankton distribution and function in a changing ocean (Marine Zooplankton Colloquium 2, 2001). One important long-standing question that SyPRID might resolve is the degree to

Fig. 8. Representative larval forms collected in the SyPRID sampler over Blake Ridge. All scale bars are 100 um. A, B: Bivalve veliger larva. C. Gastropod veliger larva. D. Setigerous polychaete larva. E. Mitraria larva of oweniid polychaete. F. Pelagosphera larva of a sipunculan. G. Barnacle cyprid larva. H. Echinopluteus larva of a sea urchin. I. Cyphonautes larva of an anascan bryozoan.

which larvae originating in isolated chemosynthetic habitats are retained locally, corralled by mesoscale oceanographic features (Van Dover et al., 2001) or are advected over long distances by currents (Adams and Mullineaux, 2008; Arellano et al., 2014). Both processes likely act in concert to maintain local populations and facilitate gene flow. SyPRID precision sampling may resolve this and other hypotheses related to genetic exchange, phylogeography, community ecology, and conservation.

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