Foresight Workshop on Advances in Ocean Biological Observations: a sustained system for deep-ocean meroplankton

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Abstract

Recent advances in technology have enabled an unprecedented development of underwater research, extending from near shore to the deepest regions of the globe. However, monitoring of biodiversity is not fully implemented in political agendas and biological observations in the deep ocean have been even more limited in space and time.
The Foresight Workshop on Advances in Ocean Biological Observations: a sustained system for deep-ocean meroplankton was convened to foster advances in the knowledge on deep-ocean invertebrate larval distributions and improve our understanding of fundamental deep-ocean ecological processes such as connectivity and resilience of benthic communities to natural and human-induced disturbance. This Meroplankton Observations Workshop had two specific goals: 1) review the state-of-the-art instrumentation available for meroplankton observations; 2) develop a strategy to implement technological innovations for in-situ meroplankton observation. Presentations and discussions are summarised in this report covering: i) key challenges and priorities for advancing the knowledge of deep-sea larval diversity and distribution; ii) recent developments in technology and future needs for plankton observation; iii) data integration and oceanographic modelling; iv) synergies and added value of a sustained observation system for meroplankton; v) steps for developing a sustained observation system for deep-ocean meroplankton and plans to maximise collaborative opportunities.

Keywords

deep-ocean observations, meroplankton, connectivity, underwater technology

Date and place

27-29 May 2019, Universidade de Aveiro, Aveiro, Portugal

List of participants

Same as author list

Introduction

Increasing exploitation of marine resources, pollution and climate change are affecting ocean’s health and the ecosystem services they provide. Fundamental knowledge of marine biodiversity and ecosystem functioning is critical for understanding the magnitude of natural and human-induced impacts on the marine environment, informing marine spatial planning and supporting sustainable and ethical Blue Economy (Benedetti-Cecchi et al. 2018, Henson 2014). Advances in technology over the past four decades have enabled an unprecedented development of underwater research, extending from near shore to the deepest regions of the globe. However, monitoring of biodiversity is not fully implemented in political agendas (Benedetti-Cecchi et al. 2018) and biological observations in the deep ocean have been even more limited in space and time, with a few exceptions at chemosynthetic habitats, and a relatively small number of time-series stations reaching abyssal depths (Glover et al. 2010, Smith et al. 2017).
Cumulative evidence over the years has revealed temporal changes in structure and abundance of deep-sea benthic communities, derived by a combination of habitat disturbance and varying food input (Campanyà-Llovet et al. 2017). Understanding the connections between water column and benthic processes (benthic-pelagic coupling) became more relevant as shifts in benthic assemblages were correlated with climate-driven variations in plankton communities (Smith et al. 2013, Sweetman et al. 2017). Aside from the effects on nutrient and energy fluxes, benthic-pelagic coupling has a critical role in regulating community dynamics. Nevertheless, how individual species respond to environmental variability, both as adults and as larvae or other dispersing stages in the water column, is far from being completely understood.

Species reproductive and dispersal traits can indicate some of the mechanisms that regulate broadscale patterns of community dynamics in the deep-sea benthos (Tyler and Young 1992). The degree to which populations self-recruit or receive subsidy from other populations (e.g. Hardy et al. 2015) has consequences for a number of fundamental ecological and evolutionary processes that affect population regulation, persistence and resilience to natural and anthropogenic disturbance. Specific dispersal strategies (e.g. feeding and non-feeding pelagic larvae, brooding) and hydrodynamic processes (e.g. currents, density interfaces, benthic storms, and tidal oscillations at the seafloor) may significantly influence geographical distribution patterns as well as historical and contemporary rates of connectivity.

In recent years, integrated multidisciplinary approaches, incorporating high-resolution biophysical modelling and genetic markers, have been applied increasingly to assess spatiotemporal scales of dispersal and connectivity in the deep sea (Vic et al. 2018, Breusing et al. 2016, Hilário et al. 2015). However, these approaches suffer from fundamental limitations, preventing realistic estimates. Empirical information on larval lifespans, movements and depth distributions is required to validate emerging connectivity models. Vertical ontogenetic migrations have been inferred by occurrences of several deep-sea larvae in the upper water column (Arellano et al. 2014). Demersal dispersal also appears to be a viable strategy in the deep ocean, although research has been hampered by the difficulty of collecting plankton near the sea floor (Young et al. 2018). Development of dedicated gear and technological assets that deliver direct observation of deep-ocean larval distributions is crucial for the collection of process-oriented data and local short-term applications. This effort must be coordinated and integrated into sustained long-term observatories, that ensure the necessary replication and resolution to cover the multiple spatial and temporal scales at which marine connectivity operates.

**Aims of the workshop**

The aim of this workshop was to foster advances in the knowledge on deep-ocean invertebrate larval distributions and improve our understanding of fundamental deep-ocean ecological processes such as connectivity and resilience of benthic communities to natural and human-induced disturbance. This knowledge is also key for understanding fundamental biological processes, improving model predictions, and providing essential
data for advanced marine spatial planning, particularly for the design of Marine Protected Area networks, increasingly being implemented for deep-sea ecosystems in national and international jurisdictions.

Specific goals of the workshop were:

1. **to review state-of-the-art instrumentation available for plankton observations in shallow and deep waters** – summarise the current state-of-the-art in sampling the distribution of larvae in the deep sea, articulate pressing research questions, and identify the current methodological and technological challenges to overcome.

2. **to develop a strategy to implement technological innovations for in-situ meroplankton observations** that can be combined with biophysical oceanographic modelling, to provide accurate, reliable and cost-efficient data for deep-ocean realms - define a strategy for going forward, particularly with implementation of sampling concepts, ideally creating a collaborative project for the integration of data collected (e.g. available time-series samples) and standardisation of methodologies using current ocean observing platforms.

The workshop was initially organized around three major themes:

1. Knowledge advances in deep-sea larval diversity and distribution: key challenges and priorities - synthesis of the latest progress on larval ecology studies in deep-ocean habitats and discussion of major challenges and priorities for larval collections in deep waters.

2. Recent developments in plankton observation technology and approaches - inventory of available instrumentation used in plankton research, identification of future technical development, and brainstorming for innovative ideas in the field of in situ observation of deep-ocean larvae.

3. Data integration and oceanographic modelling - discuss the data requirements to improve model predictions and the necessary framework for data management and access to end users, including the science community and other stakeholders (e.g. governmental agencies, environmental managers, policy makers).

During pre-workshop interactions, it also became apparent that it was important to discuss possible synergies with current initiatives and infrastructures involved in ocean observation and the added value of deep-ocean meroplankton sustained observations to socio-economic sectors, specifically to marine conservation and management.

**Methodology and Agenda**

During a pre-workshop meeting at the 15th Deep-sea Biology Symposium in Monterey, USA (9-14 September 2018), relevant information regarding the three topics was collected, based on which, a questionnaire was prepared and distributed to the registered participants one month before the workshop. The replies, as well as relevant literature for each theme, were compiled and digested for a more structured discussion during the workshop. Participation of Luciana Génio in the European Ocean Observing
The Agenda of the workshop can be found below. The workshop was co-convened by Marina R. Cunha, Luciana Génio, Florence Pradillon, and Morane Clavel Henry. Each thematic session of the workshop started with one or more invited talks (abstracts available at Suppl. material 1) followed by a period of discussion, the presentation of the results of the questionnaire, and a second round of discussion among the participants. Interactive videoconference facilities enabled the remote but vivid contributions of several participants who could not travel to Aveiro.

Workshop Agenda:

**Day 1 (Monday 27th May)**

09.00 Opening remarks (Florence Pradillon & Marina R. Cunha)

Workshop introduction

Funding support and acknowledgments

Theme 1. Knowledge advances in deep-sea larval diversity and distribution: key challenges and priorities

09.30 Keynote talk. Deep-sea larval diversity, dynamics and distribution. What would we like to know? What do we know and how did we learn it? What are we missing and why? (Craig Young, University of Oregon, USA)

10.15 Discussion

11.00 Coffee Break

11.30 Summary of pre-workshop inputs from participants - Theme I (Florence Pradillon)

11.45 Discussion

12.30 Lunch

Theme 2. Recent developments in plankton observation technology and approaches

14.00 Keynote talk. Ocean observing: new technology and approaches (James Birch, Monterey Bay Aquarium Research Institute, USA)

14.45 Discussion

15.30 Coffee Break
16.00 Summary of pre-workshop inputs from participants - Theme II (Marina R. Cunha)

16.15 Discussion

17.30 End of first day

Day 2 (Tuesday 28th May)

Theme 3. Data integration and oceanographic modelling

09.00 Short talk and discussion. Modelling deep-sea currents and impacts for dispersal of larvae (Jonathan Gula, University of Brest, France)

Overarching issues: Synergies and added value of a sustained observation system for meroplankton

09.30 Short talks.

Deep-sea meroplankton and conservation in the deep-sea: the design and implementation of marine protected areas (Anna Metaxas, Dalhousie University, Canada)

EMSO Azores Deep-sea Observatory - 9 years of operations (Jozée Sarrazin, Ifremer, France)

How can Ocean Networks Canada’s NEPTUNE observatory support future monitoring of meroplankton communities in the NE Pacific? (Fabio De Leo, Ocean Networks Canada & University of Victoria, Canada)

10.15 Discussion

10.30 Coffee Break

11.00 Summary of pre-workshop inputs from participants - Theme III (Morane Clavel Henry)

11.15 Discussion

12.30 Lunch

14.00 Synthesis presentation (Marina R Cunha)

14.15 Discussion Overarching questions and focus of review paper

15.00 Coffee Break

15.30 Discussion Way forward

17:30 End of second day

19.30 Group dinner
Day 3 (Wednesday 29th May)

09.00 Hands-on session Assign lead authors and initiate draft of review paper

10.00 Coffee Break

10.30 Continue hands-on session Workshop report

12.00 Lunch

13.30 Wrap up (Marina R. Cunha)

15.00 End of Workshop

Key outcomes and discussions

Fifteen researchers were gathered at the University of Aveiro for the workshop, and seven more joined via videoconference. The introduction session was initiated by brief roundtable presentations of the participants’ backgrounds and expertise which included larval biology, benthic ecology, taxonomy, molecular biology, genomics, modelling and engineering (underwater technology development), among other disciplines. Representatives of the Neptune Observatory – Ocean Networks Canada and the EMSO-Azores observatory – European Multidisciplinary Seafloor and water-column Observatory (EMSO distributed Research Infrastructure) were also present (Fig. 1).

Figure 1. [do](#)
Workshop participants “under the microscope” at the campus of Universidade de Aveiro: Back row, left to right: Fábio Matos, Jonathan Gula, Henrique Queiroga, Rob Young, Sven Laming, Kirstin Meyer-Kaiser, Jozée Sarrazin, Craig M. Young, Fabio De Leo. Front row, left to right: Jim Birch, Morane Clavel Henry, Marina R. Cunha, Clara Rodrigues, Florence Pradillon, Anna Metaxas.
The session continued with the presentation of the specific goals, the three main themes, and expected outcomes and impact of the workshop. Funding entities were acknowledged with brief institutional presentations of their mission and goals. Finally, the workshop agenda and procedures were discussed and approved.

**Theme 1. Knowledge advances in deep-sea larval diversity and distribution: key challenges and priorities**

The keynote talk by Craig Young was centered around three main questions:

1. What would we like to know?
2. What do we know and how did we learn it?
3. What are we missing and why?

**What would we like to know?**

Craig Young introduced key concepts in larval biology and highlighted the importance of distinguishing mechanisms of dispersal versus genetic connectivity, which is influenced by many processes including patterns of dispersal. Neither of these parameters are addressed equally well by a single, currently-available method, because they operate on different time scales. Marine populations are connected through individual exchange and they persist, decline or increase because of survival and recruitment. Thus, larval dispersal is often a key parameter when marine metapopulations are examined to understand connectivity patterns and develop conservation management models (Puckett and Eggleston 2016). Metapopulation studies involve either demographic modelling approaches or the analysis of the migratory component linking source and sink populations (Neubert et al. 2006, Mullineaux et al. 2018). The latter is based on the concept of a larval pool, through which populations are connected. Yet, the main question to be answered is how do larval interactions within their dynamic environment contribute to species and ecosystem resilience. In order to answer this question, we need more data and information on the temporal trends of connectivity, recruitment and survival.

**What do we know and how do we know it?**

Currently, knowledge on deep-sea larvae is collected using different approaches. Genetic connectivity is estimated for some species by molecular methods (sequencing), while potential dispersal capabilities or trajectories are estimated by modelling (e.g., Yearsley and Sigwart 2011, Young et al. 2018). Cultures of selected species also provide data on some physiological, developmental and behavioural traits, which are useful for constraining models of larval dispersal. Lastly, discrete data on vertical distribution are provided from *in situ* plankton sampling, as well as from a few time series, which allow both ground-truthing of model predictions and insights into *in situ* distribution patterns of deep-sea larvae.

Ocean current models can help estimate larval trajectories, but they are based on underlying assumptions, especially for biological parameters. Biological information is often missing, and modelling is thus best used as a predictive tool to inform future sampling.
Planktonic larval duration (PLD) is one of the most important biological parameters needed to estimate larval dispersal. PLD is however extremely difficult to measure, due to the difficulties in culturing deep-sea larvae, and the fact that many factors, both biological and environmental, introduce intraspecific variability in PLD (e.g. Larsson et al. 2014, Rouse et al. 2009): poecilogony, delay of metamorphosis, maternal yolk investment, temperature, food availability, larval budding, and pre-settlement metamorphosis. Extrapolation from shallow relatives often fails to accurately estimate deep-sea PLDs, mainly because physiological and embryological (larval) processes are generally slower in deep-sea than in shallow-water species (e.g., Hilário et al. 2015). Indirect estimations from spawning and settlement patterns are sometimes used to infer development times and PLDs, but such estimates require knowledge of reproductive patterns. \textit{In situ} collection of larvae is mainly hindered by the enormous dilution factor in the deep ocean that makes larvae extremely rare in this environment. Large volume plankton pumps deployed near vents may collect several hundreds of larvae (Mullineaux et al. 2005, Mullineaux et al. 2013). However, in most deep-sea ecosystems, the number of collected larvae is often less than 10 specimens, representing only a few morphotypes, with different morphotypes at different depths. Moreover, sorting is time consuming, and identification requires significant expertise. Although various systems have been employed over the years to overcome this issue, all have their caveats (see further discussion below and also Theme 2).

**What important pieces are we missing?**

Larval diversity per se tells little of ecological interest, since it is a poor and unpredictable reflection of adult diversity in the context of metapopulation ecology. Critical knowledge gaps on deep-sea connectivity are mainly in the temporal patterns of dispersal and settlement (Metaxas 2004, Mullineaux et al. 2010), the vertical trajectories of larval dispersal (Arellano et al. 2014, Yahagi et al. 2017) and the comparison of larval and juvenile stages to identify post-settlement process (Arellano and Young 2010). Assessment of spatial-temporal patterns is a huge task that can only be tackled by strategic, long-term sampling through the deployment of multiple devices including automated plankton samplers (Doherty and Butman 1990, Lewis and Heckl 1991, O’Hara 1984), low cost tube traps, and recruitment experiments (Baldrighi et al. 2017, Cunha et al. 2013, Cuvelier et al. 2014). Assessing both larval supply and settlement patterns would allow identifying post-settlement processes that are relevant for recruitment success.

Because metapopulations are connected by dispersal and dispersal potential varies with depth, important unanswered questions are: \textit{At what depth do larvae disperse?} and \textit{How much time do larvae spend drifting at various depths?} Together with PLD, ontogenetic vertical trajectories of larval dispersal are the most essential biological parameters in modeling connectivity (Young et al. 2012). Possible approaches are:

1. target larval sampling at defined depth horizons sampling larvae using precision large-volume plankton samplers (e.g. Sentry/SyPRID, Billings et al. 2017);
2. collect larvae and juveniles from the ocean floor and demersal water column using tube larval traps; and
trace the characteristics of water masses (\textsuperscript{18}O tracer) where larvae develop using isotopic and geochemical markers (elemental fingerprinting, Génio et al. 2015). Such approaches contribute crucial information to overcome the lack of knowledge on ontogenetic vertical trajectories of larval dispersal – a persistent and perhaps the greatest barrier to understanding connectivity.

Discussions following the keynote talk focused on the various caveats of existing sampling equipment. Fabio De Leo questioned the reliability of sampling to accurately quantify densities of larvae, knowing that tools can be selective. It was acknowledged that the best tools to achieve really large sampling volumes currently are MOCNESS nets as well as the SyPRID system mounted on the Sentry AUV. However, MOCNESS nets are semi-quantitative, because mesh size may be too large to collect some taxa and estimates of sample location and volume are not always accurate. Stace Beaulieu and Dhugal Lindsay suggested some adaptations (fine mesh sizes and longer nets or double-net systems to separate nekton and plankton), but Craig Young argued that such adaptations may bring other operational issues. Anna Metaxas mentioned that data and results obtained by the SyPRID system need to published in order to understand its effectiveness in different environments and for different purposes. It was recognized that comparisons between sampling tools are difficult, and that results are better compared in terms of diversity patterns than densities.

Strategies to overcome the limitations set by the time-consuming process of sorting and identifying larvae were also debated. Jim Birch asked how humanpower could be replaced or aided by an automated system in this process. The consensual perspective of the participants about this was that automated solutions, although desirable, are not achievable in the near future mainly because current equipment such as the Continuous Plankton Recorder (CPR) and other video plankton recorders do not have sufficient resolution, and reference databases to assist both genetic and morphological identification are lacking. Stace Beaulieu and Dhugal Lindsay reminded participants of some existing platforms (e.g. EcoTaxa, Picheral et al. 2017), which are currently cataloguing predominantly holoplankton and could be expanded for meroplankton. In any case, the need to reinforce taxonomy training of young scientists is undeniable.

**Synthesis and discussion of the questionnaire’s responses**

Florence Pradillon presented the summary of the pre-workshop responses to the questions concerning Theme 1.

**Question 1.1. Why do we need to quantify deep-sea larval diversity and distributions?**

Data on larval diversity and distribution can provide insights about the reproductive traits of some species, such as reproductive timing, seasonality, fecundity, and larval traits such as developmental mode, swimming ability, and buoyancy. These biological traits are important parameters for accurate modelling of larval trajectories, and thus for predictions of connectivity patterns. A better understanding of basic biological parameters will also allow
for assessment of the effects of geographic and seascape settings, and provide more accurate predictions of species’ dispersal ranges. Besides bridging gaps in the fundamental knowledge of the life history of deep-ocean species, this will help increase understanding of metacommunity dynamics and inform spatial planning strategies (e.g. MPA networks).

Deep-sea larval diversity and distribution can also inform patterns of larval supply and settlement potential. These parameters will help better evaluate the role of larvae in regulating diversity and distribution of deep-sea benthic fauna, increase mechanistic understanding of deep-sea benthic community dynamics and resilience, facilitate the development of realistic and spatially explicit population models, and provide informed scientific advice on the status of and threats to marine resources.

Larvae are not only a major determinant of benthic and pelagic faunal distribution through settlement; they may also have a role through trophic networks. Meroplankton may represent a yet-undocumented food source in the deep ocean, but also potentially a competitor for food in the planktonic community. However, the trophic contribution of propagules (gametes and larvae) to the carbon cycle is likely to be significant only on local patchy scales, since dilution effects probably reduce its relevance to large scales.

Lastly, the importance of quantifying and monitoring meroplankton in the deep ocean was recognized with regards to its likely sensitivity to global change. It is critical to understand how larvae in the deep ocean may respond to natural variation and multiple climate change stressors, and how this may influence the functional role of meroplankton. Larval distributions could even be used as biological indicators of climate-driven influence on deeper water layers, although the current lack of knowledge on deep-ocean larvae makes this a distant prospect.

**Question 1.2. What are the key challenges and limitations?**

The most relevant limitations identified by the participants can be grouped into the following categories: ecological and biological knowledge, reference databases and taxonomical expertise, observation design and technological limitations.

**Ecological and biological knowledge**

One of the major challenges to gain knowledge of larval diversity and distribution in the deep ocean is in their *in situ* observation and collection. Deep-sea larvae exhibit very low abundance in general, even near ‘biomass hotspots” such as hydrothermal vents (dilution factor - densities in the deep ocean are 100 to 10000 times lower than in coastal systems; Table 1). Tools allowing the screening of very large volumes of waters are required (in the order of tens to thousands of m³). Developing such tools is costly, in terms of energy, operational time…and few of them exist.
<table>
<thead>
<tr>
<th>Abundance (ind/m³)</th>
<th>Setting</th>
<th>Depth (m)</th>
<th>Instrument (mesh µm)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.03-0.3</td>
<td>Abyssal plain, BBL (CCZ)</td>
<td>4050</td>
<td>Pump (63)</td>
<td>Kersten et al. 2017</td>
</tr>
<tr>
<td>0.25-18</td>
<td>Oceanic ridge axis near hydrothermal vents (Pacific)</td>
<td>2500</td>
<td>Pump (63)</td>
<td>Beaulieu et al. 2009, Mullineaux et al. 2005,</td>
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<td>Mullineaux et al. 2013</td>
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<tr>
<td>&gt;1</td>
<td>Oceanic ridge axis &gt;1 km from hydrothermal vents (Pacific)</td>
<td>2500</td>
<td>Pump (63)</td>
<td>Mullineaux et al. 2013</td>
</tr>
<tr>
<td>0.01-9</td>
<td>Seamounts, (western Pacific)</td>
<td>350-1600</td>
<td>Net (63)</td>
<td>Metaxas 2011</td>
</tr>
<tr>
<td>Up to 1000</td>
<td>Fjord (Arctic)</td>
<td>0-300</td>
<td>Net (180)</td>
<td>Michelsen et al. 2017</td>
</tr>
</tbody>
</table>

Describing larval diversity and distribution is not sufficient to assess dispersal trajectories and connectivity patterns. Sources of dispersing larvae, as well as the identification of settlement and recruitment mechanisms, are also needed to understand how benthic communities are established and vary temporally. The use of tracers such as elemental fingerprinting of larval shells may help identifying the source of some larvae, but these tools first require mapping the signature of potential sites of origin.

Another challenge is the lack of biological information on deep-sea species' life cycles (reproductive timing, larval biology and ecology). *Ex situ* experimentation with larval stages could provide information on larval physiology, behaviour and biology (e.g. Larsson et al. 2014, Strömberg and Larsson 2017), but remains an extremely challenging task due to the difficulties in culturing deep-sea animals (Jiang et al. 2016). *In situ*, large, and multi-scale experiments may be conducted but require the development of a coherent, coordinated and comprehensive network of observation stations/platforms.

**Reference databases and taxonomical expertise**

A major limitation to our knowledge of meroplankton is the identification of deep-sea larvae, most of which remain undescribed with no recognition of their adult counterpart. The use of genetic barcoding is promising as an accurate tool to link adult forms with their respective larval forms, but depends heavily on the availability of reference data which are still scarce for the deep sea (some data are available through disparate databases: e.g., BOLD, WORMS, EOL, OBIS). Although relevant work has been carried out on some taxa from specific deep-ocean environments (e.g. Mills et al. 2007), there is currently no global taxonomic database dedicated to deep-sea larval identification, and a large effort in sequencing and imaging remains necessary to develop such a platform. New imaging technologies (3D holographic images, fluorescence; Colin et al. 2017) are being developed.
and would allow automated (potentially in situ) recognition of larvae using supervised machine-learning methods. However, such tools require prior learning with annotated training sets and the development of computer-based classification tools for high-throughput identification.

Observation design and technological limitations

From an operational point of view, developing our knowledge on deep-sea meroplankton diversity and distribution would require global engagement among marine observing systems; integration of the deep ocean with the coastal component of the marine observing systems covering physical, chemical and biological variables; seascape representativeness, and observation of key locations, with criteria based on seascape variability, biodiversity hotspots, MPAs vs non-protected areas, major environmental boundaries, larval gateways, etc.

Technological development required for meroplankton studies in situ faces many challenges related to the remoteness and hostility of the environment. There is difficulty in maintaining equipment, sustaining, and implementing an evolving observation system. Challenges include longevity of equipment, pressure resistance, power communication, timing of sample collection, miniaturization, corrosion, drift detection, deployment, communication, and recovery costs. Sampling bias due to larval avoidance and selectivity must also be addressed when developing sampling tools /strategies.

Solutions to the technological challenges exist (as proved by developments presented in theme 2), although adaptations are still needed regarding sampling size. The main scientific questions must be defined to drive technological development. For example, many questions identified above need time-series data and could push for the development of carrousel-type multi-samplers. Nevertheless, effective sampling strategies and larval tracking will not be possible until we have a sense of timing and direction of larval release. Focusing research first at sites where a large amount of baseline reproductive data is available thus may be a good strategy.

Theme 2. Recent developments in plankton observation technology and approaches

The keynote talk by Jim Birch first provided a brief introduction to the team of engineers and scientists of the Monterey Bay Aquarium Research Institute (MBARI), along with their goals for development and innovation in deep-sea observation and technology, followed by the state-of-the-art technologies and new approaches for ocean observations.

Conceptually, studying processes that change both spatially and temporally seems relatively straightforward - one needs to sample in many locations synoptically over time, or follow a coherent water mass and sample it repeatedly. However, implementing either approach presents many challenges. Jim Birch identified the major long-standing challenge for deep-ocean observations as “Being there!”, more specifically, the need for high resolution, broad spatial and temporal scale strategies for ocean observing. This can only
be achieved if human effort required for the acquisition and processing of samples can be reliably replaced by autonomous systems, for instance by the application of molecular probe technology in situ, by implementing extended, unattended operations outside of a laboratory setting, and by developing tools for data visualization and decision support.

The MBARI approach is to develop a new generation of robotic systems with a focus on microbial-mediated processes. The ecogenomic sensors, their components and evolution were presented with examples of Environmental Sample Processors (ESP) capable of sample collection and processing, real-time probe analyses, toxin monitoring, and sample preservation. Analytical modules, such as in situ specialised detection qPCR for sequencing species composition in tandem with environmental data (Harvey et al. 2012, Preston et al. 2011), can be added optionally. Lessons learned from the second-generation ESP, a “lab-in-a-can” allowed for the system to be re-engineered for use on a Tethys-class Long Range AUV (LRAUV). The new instrument is called the third-generation ESP (3G-ESP), and its integration with the LRAUV provides mobility and a persistent presence not seen before in microbial oceanography. Deep-sea versions of the ESP are housed in a large titanium sphere with pressure control for incoming water samples. This robotic microbiology “lab-in-a-can” can be used to monitor temporal changes in a body of water (e.g. diel cycles in gene expression of microbiota, Ottesen et al. 2014), in stationary deployments at ocean floor observatories (MBARI 2017), or for mooring-based data collection (Seegers et al. 2015), and can be coupled with drifters and a range of autonomous vehicles (gliders, AUVs, benthic rover). Several technical details and application examples were provided to illustrate the operational capabilities of these systems.

The importance of mobility was highlighted, particularly in the case of adaptive sampling strategies where targeted sampling can be performed autonomously over periods of days to weeks. For instance, capturing the dynamics of an oceanic front or of the deep chlorophyll maximum can be very laborious and difficult using traditional CTD rosette casts from a ship, but robotic assets can be set for locating and chasing these water masses with particularly defined characteristics (Pargett et al. 2015, Zhang et al. 2019). Such exercises require seamless autonomy and navigation and can be optimised by the use of networked vehicle systems that operate independently while still working towards a common goal. These systems consist of a constellation of coordinated, interoperable, communicating robotic assets and platforms, offering never-before-seen ocean exploration and research capabilities.

Finally, it was concluded that robotics can be a powerful tool to address the primary ocean observation challenge of acquiring samples over relevant spatial and temporal scales. However, the use of robotic assets in biological observations is being somehow hindered by the slow development of operational sensors incorporating molecular analyses, which remains a processing bottleneck. The near-future direction is to adjust existing robotic assets into the various areas of meroplankton research, namely by reaching deeper waters, increasing sample volumes, and increasing sampling durations.
The discussion following the keynote talk focused on the technological obstacles that must be overcome to answer the needs of meroplankton investigations. Anna Metaxas mentioned that, considering the enormous dilution factor of larvae in the deep ocean, sampling large volumes of water must be reconciled with mobile, portable sensors. Florence Pradillon added that the dilution makes locating and chasing larvae a greater challenge, even in cases when proxies (e.g. turbulence, vent plumes) may be used. Another major limitation in deep-ocean meroplankton research, besides finding the larvae, is the substantial expertise required for their identification. Stace Beaulieu inquired about new developments regarding testing and mounting meroplankton imaging systems in autonomous vehicles. Jim Birch responded that meroplankton imaging systems are still not efficient enough to be used in autonomous vehicles or other robotic assets because the image data still requires human processing at this time, whereas the current system takes advantage of nearly complete autonomy, communicating the main species present in the form of a tiff image or a genetics-derived array. In the future, supervised machine-learning methods for image analysis might be possible, but this obviously would require extensive validation and the existence of a certified image database. Rob Young indicated that a similar problem persists with genomic data, but the use of NGS-multi-loci indicators is beginning to improve the resolution of genetic analyses. Luciana Génio suggested focussing on adult metagenomics as a precursor to larval genetics-based identification as a way forward, which, however, is currently hampered by a lack of reference genomic data.

Synthesis and discussion of the questionnaire’s responses

Marina Cunha presented the synthesis of the pre-workshop responses to three questions concerning Theme 2.

Question 2.1. What research or new technologies do we need for deep-sea meroplankton observations?

The answers to this question were structured around the main research questions in meroplankton studies, the methodological approaches to tackle these questions, and their expected outputs Table 2. The need for accurate larval identification (integrated taxonomy), building up genetic and image reference libraries, mapping distributions (from fine to large scale) and combining ecological and genetic data to understand life histories was consensual among the workshop participants.

<table>
<thead>
<tr>
<th>Approaches</th>
<th>Data and outputs</th>
<th>Research questions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metabarcoding, eDNA/eRNA</td>
<td>Genetic databases</td>
<td>Taxonomy, biodiversity, distribution, biogeography</td>
</tr>
<tr>
<td>Gene expression</td>
<td></td>
<td>Larval development</td>
</tr>
</tbody>
</table>

Table 2.
Main methodological approaches for meroplankton research.
### Approaches

<table>
<thead>
<tr>
<th>Laser Capture Microdissection combined with molecular analysis</th>
<th>Larval gut contents</th>
<th>Trophic ecology</th>
</tr>
</thead>
<tbody>
<tr>
<td>Synchrotron, micro-CT</td>
<td>Internal anatomy and physiology</td>
<td>Ontogeny, larval development</td>
</tr>
<tr>
<td>Elemental fingerprinting, stable isotope analyses</td>
<td>Individual biogeochemical signatures</td>
<td>Ontogenetic migration, dispersal trajectories, source-sink dynamics</td>
</tr>
<tr>
<td>In situ and ex situ imaging</td>
<td>Image databases</td>
<td>Taxonomy, biodiversity</td>
</tr>
<tr>
<td>In situ imaging combined with automated identification</td>
<td>Faunal composition datasets for a given water mass</td>
<td>Taxonomy, biodiversity, distribution, biogeography</td>
</tr>
<tr>
<td>In situ imaging combined with environmental data</td>
<td>Ecological databases</td>
<td>Fine-scale distribution, ontogenetic migration</td>
</tr>
</tbody>
</table>

New methodologies are being increasingly used to gain more knowledge on trophic ecology and internal structure of larvae. A combination of Laser Capture Microdissection (LCM) and DNA metabarcoding was successfully applied to analyse the gut contents of wild-collected cephalopod paralarvae (Fernández-Álvarez et al. 2018). Synchrotron- and micro-CT can be used to observe internal structural (physiological or functional) change during ontogenic development (Chen et al. 2018). These methodologies become more powerful when combined with genetic (e.g. gene expression) and ecological data (e.g. larval, juvenile, adult distributions). Information on life-history traits is crucial to provide adequate data for modelling and advance our knowledge of connectivity.

Other concerns raised during the discussion included insufficient genetic resolution and the need to gain more accurate knowledge on larval sources. Also debated was the extent to which taxonomy is currently a limitation for meroplankton research and related topics, such as the usefulness of taxonomical vs functional information (e.g. ecosystem function, response to stressors), and the large-scale sequencing data quality issue.

Obtaining more larval samples and improving the spatial and temporal coverage of sampling is indisputably the fundamental step for further advancing meroplankton research. But quantification is definitively a major challenge. Lombard et al. 2019 carried out an extensive review on the technologies available to make globally quantitative ocean observations of particles in general and plankton in particular, detailing a variety of relevant technologies and measurements, including bulk measurements of plankton, pigment composition, uses of genomic, optical and acoustical methods, as well as analysis using particle counters, flow cytometers and quantitative imaging devices. However, there are many deep-ocean meroplankton-specific constraints that remain to be resolved.

The main technological challenges identified by the workshop participants in relation to sampling and sampler type are summarised in Table 3. The debate was focused around bulk water samples and individual organism samples and continued around some of the topics already addressed in the discussion following the keynote talk, such as automated plankton imaging and identification, operational autonomy and versatility of samplers,
duration vs resolution of time-series. The biggest challenge is imposed by the dilution factor and the subsequent large volume needed for obtaining representative samples of deep-ocean meroplankton. Morane Clavel and Jim Birch suggested that one way to mitigate the dilution factor, while waiting for future technological breakthroughs, is to identify physical ‘pinch-points’ for sampling where larvae are moving through a physically constrained space (e.g. straits, oceanographic fronts).

<table>
<thead>
<tr>
<th>Sampler type</th>
<th>Technologies</th>
<th>Technical challenges</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bulk water samplers</td>
<td>general samplers (nets, pumps)</td>
<td>increase adaptability and versatility for operation with autonomous, land-or vessel-based control, or mounted on different underwater vehicles (ROV, manned submersibles, gliders); resolve spatial vs temporal resolution and coverage</td>
</tr>
<tr>
<td></td>
<td>large volume pumps for sequential, long term sampling</td>
<td>resolve volume vs mesh size and long term vs temporal resolution constraints; improve specimen preservation</td>
</tr>
<tr>
<td></td>
<td>autonomous sampling gear with geolocation, controlled speed and water flowmeter</td>
<td>control geolocation (both vertical and horizontal); adjust filtering speed to the size of target organisms</td>
</tr>
<tr>
<td></td>
<td>multitraps with open-closure system</td>
<td>control/monitor from land</td>
</tr>
<tr>
<td>Sensors</td>
<td>ecogenomic sensors (automated, miniaturized in situ environmental nucleic acids samplers)</td>
<td>increase depth range, sample volume, and sampling duration; decrease cost; resolve lack of reference databases to feed supervised machine learning methodologies</td>
</tr>
<tr>
<td>Individual-based methodologies</td>
<td>in situ imaging</td>
<td>increase sample volume; resolve in situ optic constraints; enable simultaneous larval collection for sequencing</td>
</tr>
<tr>
<td></td>
<td>automated identification</td>
<td>resolve lack of reference databases to feed supervised machine learning methodologies</td>
</tr>
<tr>
<td></td>
<td>larvae-specific tools (e.g. polarization of mollusc shells, elemental fingerprinting, stable isotopes)</td>
<td>resolve lack of geolocated reference databases on target elements; improve resolution of source populations (reference datasets with larva with known origin/trajectories</td>
</tr>
</tbody>
</table>

Scientists need to establish the minimum requirements for enabling sampling gear to effectively answer their research questions. This is an important step to overcome the technological challenges of meroplankton sampling - as Jim Birch stressed, communication between scientists and engineers must be improved so that engineers can comprehend the scientific goals and design the necessary fit-for-purpose equipment.
Question 2.2. What do we expect from an observing system for deep-sea meroplankton?

An observing system for deep-ocean meroplankton is expected to improve the temporal and spatial coverage and resolution of sampling. In the pelagic ecosystem, characterized by its high variability at various spatial and temporal scales (e.g. Game et al. 2009), the dynamics of meroplankton cannot be adequately monitored, or even captured, by isolated or opportunistic sampling efforts. The observing system should be sustained, i.e. provide long-term time series (10 years or more), and of high temporal resolution (ideally one week or less). The spatial resolution is most likely constrained by available funds, and therefore continuous fine-scale and high resolution data should target specific habitats and ecosystems (biodiversity hotspots) or focus on specific processes, such as source-sink dynamics. The observing system should provide not only qualitative but also quantitative data, which are the basis of the mechanistic understanding of ecological processes, enable the parameterization, validation, and predictive capacity of modelling efforts, and are also required for effective adaptive management of ecosystems. The detection of larval vertical distributions was considered a priority for which large-volume gear, such as precision pumps (SyPRID) and vertical multinet plankton samplers (e.g. VMPS-6000D with frame opening area of 1m$^2$ and 8 nets of 100 $\mu$m mesh size), which must be optimised.

The workshop participants established that the observing system must ideally have the capability to deliver relevant data for the following research fields:

- Biodiversity: quantitative data on community composition, abundance and biomass;
- Source-sink dynamics: data on fecundity, timing and synchronicity for reproductive output assessments and data on larval inputs and post-metamorphic processes for net recruitment assessments;
- Individual-based traits: size, life-history stage, and genetic data supported by accurate taxonomic identifications;
- Bio-physical modelling: environmental and biological Essential Ocean Variables.

It is foreseeable that the development of low-cost, automated, and miniaturized in situ environmental nucleic acid (eDNA/RNA) samplers (Gan et al. 2017), together with advances in metagenomics (Ruppert et al. 2019, Sinniger et al. 2016), new approaches to the processing of molecular data (Stat et al. 2017), and the trivialization of supervised machine-learning methodologies (Cordier et al. 2017) will be able to deliver large volumes of molecular data. Marina Cunha commented that while accurate taxonomic identifications will be always irreplaceable when investigating life-history traits and population processes, our traditional approach to biodiversity studies may have to change to deal with the expected volume of data, especially with regards to undescribed species. We may need to move away from a static approach to biodiversity based on the current “biological” species concept towards a more flexible approach based on evolutionary lineages and how they change over time. Rob Young added that increasing genetic data outputs will allow streamlining taxonomic (lineage) composition, detecting trends and more easily identifying changes over time in a semi-automated way. Functionality can also be used as an add-on, to identify whether or not a change in species composition is paralleled by a change in
ecosystem functioning (or vice versa). Other discussion topics related to genetic data outputs involved several participants and were summarized by Rob Young as: i) data quality issues associated with sequencing choices; ii) the need to develop multi-gene methodologies to overcome the insufficient resolution of genetic data for tracking larval origins and for connectivity studies; and iii) the advantages of using RNA vs DNA, with RNA being a better proxy for the presence of living organisms and also more effective in revealing epigenetic changes and physiological responses to stress.

Several workshop participants raised the point that molecular identifications of deep-sea larvae are presently hindered by the limited amount of data on adult molecular sequences. Species-level identifications of larvae necessitate large reference databases of sequences from described species in order to match larvae to adults. Basic research to build up public databases would significantly enhance the utility of deep-sea larval observations. In the meantime, larvae can be identified morphologically. Many Classes and Orders of invertebrates have distinct larval forms (see Young et al. 2001), so larvae can be matched to these higher taxa.

**Question 2.3. How can we develop an observing system for deep-ocean meroplankton?**

The participants agreed on the following important steps for developing a sustained observation system for deep-ocean meroplankton:

- Identify priority scientific questions for meroplankton research and define a strategy with long-term goals but short-term actions that can be implemented relatively quickly.
- Communicate the scientific and societal value for sustained observations of meroplankton to secure support from potential stakeholders and funding entities. The involvement of stakeholders working across multiple sectors is fundamental for the successful implementation and continuity of integrated ocean observing systems. Mackenzie et al. (2019) identified convergence on common goals, effective communication, co-production of information and knowledge, and the need for innovation as the most important overarching principles for stakeholder engagement.
- Develop autonomous robotic assets and other innovative meroplankton-specific sampling and observation technologies. For this, it is crucial to improve interdisciplinary collaboration and communication between scientists and engineers. Networked robotic systems for adaptive sampling (Zhang et al. 2019) and fixed observatories for continuous monitoring of the water column and seafloor (Aguzzi et al. 2019) are equipped with interoperable cutting-edge technology and rely on technological innovation for the constant enhancement of their performance.
- Establish synergistic collaborations with active ocean observation initiatives and infrastructures at national and international levels. For a successful implementation of sustained meroplankton observations, it is imperative to foster partnerships, converge on common goals, and gain access to the distributed infrastructures and...
a range on *in situ* elements under the auspices of the Global Ocean Observing System (GOOS; IOC 2019, Weller et al. 2019) and more specifically to their regional (e.g, EOOS) and deep ocean (DOOS) counterparts.

Figure 2. doi

Sustained meroplankton observations - a basin-scale approach. Low-cost samplers for deep-ocean observations (colonization modules coupled with larval traps) deployed at moorings or mounted on landers of the EMSO-ERIC distributed observatories and covering different water masses (Project LO3CATED; Génio, Cunha and Young). NACW: North Atlantic Central Water; AIW: Antarctic Intermediate Water; MOW: Mediterranean Outflow Water; NADW: North Atlantic Deep Water; MABW: Modified Antarctic Bottom Water.

Figure 3. doi

Sustained meroplankton observation systems. envisioned approach at regional scale.
Two different but complementary schemes for sustained observing systems were envisaged: i) a global approach with a large spatial coverage network using simple, low-cost sampling methodologies (Fig. 2); ii) a more specialised approach at regional scales using more complex observing and sampling assets with high spatial and temporal resolutions (Fig. 3).

**Theme 3. Data integration and oceanographic modelling**

Biophysical oceanographic modelling is a predictive tool for connectivity studies that integrates physical oceanography and life-history traits. The models enable estimates of the temporal dynamics of dispersal trajectories (Lagrangian particle tracking models) and inference of source and sink regions, pointing to areas that should be prioritized for conservation. They can be used to materialize scenarios of future potential impacts from human activities on deep-sea populations, and they provide visual outputs that facilitate the communication with managers and other stakeholders. There are currently many examples of modelling exercises for larval dispersal that are based on the comprehensive knowledge of the ocean surface circulation. However, the use of biophysical modelling applied to deep-sea larvae is lagging behind its application to their shallow-water counterparts. Accurate and realistic outputs from those models can only be achieved if sufficient qualitative or quantitative information is provided. In the case of the deep ocean, information on life-history traits is lacking, and currents are far less studied. Therefore the influence of biological and physical processes on larval dispersal and connectivity among populations is still poorly understood.

Jonathan Gula presented a unique case study for realistic high-resolution modelling of larval dispersal from the Lucky Strike hydrothermal vent. Because Bathymodiolus vent mussels are one of the best-studied taxa at the Mid-Atlantic Ridge (e.g. Breusing et al. 2017), the species *Bathymodiolus azoricus* was selected as the model organism. The deep-ocean circulation in the rift valley along the Azores sector of the North Mid-Atlantic Ridge was described (Lahaye et al. 2019). The deep currents exhibit significant variability triggered by mesoscale and submesoscale turbulence and interaction between the currents, and the topography adds significant complexity to the flows. The impact of meso- and submesoscale turbulence has been shown for the dispersion of materials in the surface layer (e.g. Poje et al. 2014), and several studies also demonstrated the importance of turbulence in the deep ocean (Adams et al. 2011, Gula et al. 2016, McWilliams 2016, Vic et al. 2018). In fact, submesoscale turbulence (1-10 km) and high-frequency forcing (tides) need to be taken into account to get a realistic approximation of the deep circulation over the Mid-Atlantic Ridge. Long-range transport of larvae and other materials is partially performed by submesoscale coherent vortices, while internal tides and associated mixing facilitate crossing topographic barriers by particles. The combined effects of the tidal and submesoscale currents and finer scales of topography significantly impact simulations of larval paths and overall make connectivity between remote vent sites more likely at biologically-relevant time scales (Vic et al. 2018). Jonathan Gula highlighted the importance of ongoing and future transatlantic projects (e.g. DEEPER, iAtlantic) that
provide free online access to the modelling products through hub servers for fully realistic simulations at large basin scales.

The discussion following the talk focused on details of the topographic and oceanographic parameters and of the biological traits included in the model simulations. The workshop participants acknowledged that the paucity of data on the life histories of deep-ocean organisms increases the uncertainty associated with model predictions. PLD and ontogenic vertical migration are particularly important, but in fact, we also know very little about reproduction parameters (e.g. seasonality of larval release - differences in the time of the year when larvae are released may lead to completely different outcomes in terms of dispersal trajectories).

**Synthesis and discussion of the questionnaire’s responses**

Morane Clavel presented the summary of the pre-workshop responses to the questions concerning Theme 3.

**Question 3.1. What methodologies or approaches do we need for data integration?**

Comprehensive ocean observing systems must be interoperable to enable studies across different science domains and observing regimes (Benedetti-Cecchi et al. 2018). However, the multi-dimensional nature of biological and ecological data results in multiple and heterogenous approaches for quantitative measurements and sampling techniques, which create difficulties for data management and interoperability. Currently, there is a pressing need to overcome these limitations and improve data dissemination according to the best practices and standards (Pearlman et al. 2019). Data integration requires that datasets share the same type of data, comply with well-established and sustainable file formats, and use common vocabulary and well-documented information. The outputs must ultimately address the needs of intermediate- and end-users (e.g., scientists, modellers, managers) and support advice to policy-makers. Moreover, data should be easily understandable by humans and interpretable by machines in compliance with the FAIR (Findable, Accessible, Interoperable, Reusable) data principles (Wilkinson et al. 2016).

The first step for data integration is probably the agreement on the critical variables to focus on. GOOS adopted the Framework for Ocean Observing (FOO, *Task Team for the Integrated Framework for Sustained Ocean Observing 2012*) as a foundational document setting the common guidelines that comprise elements from Inputs (e.g., essential ocean variables - EOVs) to Processes (observations and maintenance), to Outputs (data and products) (Tanhua et al. 2019). Eight biological EOVs were prioritized by GOOS, with two others emerging. More recently, the Biodiversity Observation Network from the Group on Earth Observations (GEO BON) identified 22 Essential Biodiversity Variables candidates organized in six classes (Benedetti-Cecchi et al. 2018). DOOS also recommended three biological EOVs (including "connectivity of species") that are relevant for the deep ocean (Levin et al. 2019). Although these sets of candidate variables aim to account for the complexity of marine ecosystems, including some relevant aspects for meroplankton research, their feasibility and consideration for operational needs may be somehow limited.
In the context of meroplankton observations, the workshop participants emphasised the importance of standardizing sampling protocols, ensuring the consistency of types of data and file formats, and implementing quality control procedures. A recurrent issue - meroplankton identification - was widely debated. The use of flow imaging microscopy with particle analyzers (Álvarez et al. 2011) and other integrated analysis systems for acquisition and classification of digital zooplankton images (Gorsky et al. 2010) may enable a faster, semi-automatic processing and identification of meroplankton samples. However, these approaches rely extremely on the existing reference databases (e.g., EcoTaxa - Picheral et al. 2017, World Wide Web of Plankton Image Curation - wwwpic Team 2020) to facilitate the specimens’ classification and minimize errors. The use of the World Register of Marine Species (WoRMS, WoRMS Editorial Board 2020) was recommended as the most up-to-date reference database for scientific names.

Another requirement fundamental to data integration is the existence of publishing platforms and repositories, which improve the interoperability of data (connections to related information). Biogeography online repositories such as OBIS (OBIS 2020) or GBIF (GBIF 2020), provide templates and dedicated publication tools that facilitate the data submission and ensures the maintenance and preservation of data, in an openly and freely accessible manner. The Darwin Core data format, used by OBIS and GBIF, was also recommended as the ideal vehicle for storage of meroplankton data. The Darwin Core facilitates data integration by offering a standard form for proper cataloguing and sharing of biological and environmental data (Wieczorek et al. 2012), and is flexible enough to allow the development of data standards for specific sub-disciplines. Whenever possible, it is desirable to link the biological occurrences to associated environmental data and, in the future, also add information on individual life history traits. In order to maximize data dissemination and use, and besides being open and freely accessible, data portals should provide pre-visualization tools that digest the information available. Yet, the expected large volume of data and the continuous need for accessibility and maintenance require long-term funding and a strong investment in informatics expertise.

**Question 3.2. Who are the potential end-users?**

A facilitated access to data products is a fundamental step of the Framework for Ocean Observing. The ultimate objective is to provide added value to the observation data by processing reusable information (“measure once/use many times”) for a broader group of intermediate and end-users in research, management, and policy making. The availability of ocean observation data in online repositories fosters its use beyond the scope of the primary studies, enabling a broader analysis of ecological, environmental and biological research questions, supporting scientific discovery and addressing societal issues related to global change Achieving this goal is a strong justification for supporting sustained data collection efforts (Levin et al. 2019, Task Team for the Integrated Framework for Sustained Ocean Observing 2012).

A better knowledge of deep-sea meroplankton is beneficial for multiple profiles of intermediate and end-users. In the scientific field, the potential users of meroplankton data include biologists and ecological modellers aiming to address fundamental research
questions including connectivity and resilience to climate change and other natural and anthropogenic disturbances. Modellers have a pivotal function among the end-users because they process the data into presentable and relevant outputs to non-scientific communities (i.e., managers, policy makers, industrial companies). Deep-ocean meroplankton data products may have a particular interest for environmental and regulatory intergovernmental bodies (e.g. OSPAR commission, International Seabed Authority), managers, and policy makers by contributing to environmental impact assessments and supporting marine spatial planning. Meroplankton data may also be used by stakeholders associated with educational and outreach programs.

**Synergies and added value of a sustained observation system for meroplankton**

This session included three presentations that illustrated, on one hand, the importance of sustained meroplankton observations to provide knowledge on connectivity for deep-sea conservation and management and, on the other hand, the importance of existing deep-ocean observation infrastructures as a starting point for implementing the vision of a sustained system for such observations.

**Added value to conservation**

Understanding the extent to which populations are connected by larval dispersal is critical for the development of strategies to sustain biodiversity and preserve deep-sea ecosystems. A growing commitment to protect the ocean's biodiversity in both national waters and areas beyond national jurisdiction requires the development of concepts for the observation and valuation of marine biodiversity and ecosystem services and their integration into conservation efforts, as well as development of scientific and technical solutions relevant to environmental impact assessments in marine areas.

Anna Metaxas first presented the criteria of the Convention on Biological Diversity (CBD) for the selection of Marine Protected Areas (MPAs) and the design of Networks of Marine Protected Areas (MPAn). MPAn are increasingly being designed and implemented for deep-ocean ecosystems. Some of the criteria provided by CBD require an understanding of processes in early life history. However, while "maximising connectivity" is considered a critical design element for all MPAn (WCPA/IUCN 2007), it ranks very low in the scale of criteria most often used by managers (Balbar and Metaxas 2019). Its implementation to date has been extremely limited and hindered by inconsistencies in the terminology and concepts considered in management, which favours landscape connectivity metrics, compared to scientific research, which uses predominantly demographic and genetic connectivity estimates and modelling approaches (Balbar and Metaxas 2019). Various techniques (e.g. network theory, Treml et al. 2008) exist to then use these estimates for the development of metrics (e.g. betweenness centrality) that can used for the design of MPAn. Overall, the data needs for conservation are similar to those needed to address fundamental ecological questions, but the data outputs differ between the two.
The discussion that followed Anna Metaxas' talk was focused on effective ways of measuring connectivity among MPAs. Although connectivity is being proposed to integrate the biological set of EOVs (Levin et al. 2019), the types of measurements, scalability and temporal variation need to be specified. The limited feasibility and associated operational issues may be responsible for the difficulty in applying this CBD criterion, often discounted in favour of socioeconomic criteria when selecting the areas for protection or resource exploration. The added value of an observation system for meroplankton research capable of providing science-based evidence to support the design of MPA networks is therefore unquestionable.

**Synergies with deep-ocean observation infrastructures**

The up-to-date map of the main in-situ components of the Global Ocean Observing System, easily obtained online, shows a variety of assets including drifters, Go-ships, buoys, moorings and other mobile and fixed platforms that cover significant areas of the oceans. However, for the deep ocean both in the seafloor and the water column, the number and distribution of such elements are dramatically fewer (Levin et al. 2019). Deep-ocean observatories with the capability to sample environmental and biological variables on a systematic and regular basis are the keystone for long-term monitoring ecosystem changes and environmental trends. There are currently few examples of infrastructures with such capabilities. Two of these infrastructures were represented in the workshop: the EMSO-Azores Observatory, a component of the EMSO-ERIC distributed observatory, and the Neptune Observatory from Ocean Networks Canada. The invited talks by their representatives Jozée Sarrazin and Fabio De Leo, respectively, provided a detailed overview of the infrastructures, the equipment and types of measurements and observations available, as well as the science focus, data products and data access policy (more details in Suppl. material 1). The contributions of the observatories to the knowledge of deep-ocean ecosystems were illustrated with various examples of scientific outputs and pathways for future collaborations in the field of meroplankton research were outlined.

Following the course of the discussions from previous sessions, it was agreed that taking advantage of the synergies resulting from the collaboration with existing ocean observation infrastructures is the way forward and will support the vision of a sustained system for meroplankton observations. Fixed observatories are crucial elements of the envisaged regional approaches for obtaining high spatial-temporal resolution and long-term temporal datasets. They are also visited regularly for maintenance purposes, providing cruises of opportunity for sampling at the site or during transit to the observatory nodes. Both talks raised the interest of the workshop participants in relation to funding and visiting opportunities and the possibility of applying for add-on experimentation (e.g. larval traps, recruitment plates, colonization modules).

**Conclusions**

The final discussion began with a synthesis of the main points raised during the previous workshop sessions. Central to the debate were the steps for developing a sustained...
observation system for deep-ocean meroplankton identified during Theme 2 session. In summary, recommendations were:

- Identify priority scientific questions for meroplankton research
- Communicate the value of meroplankton observations to stakeholders and funding entities
- Design and develop autonomous sampling/observation technologies
- Establish synergistic collaborations with active ocean observation initiatives and infrastructures

In the face of the large knowledge gap on fundamental scientific questions on meroplankton and the lack of even baseline data, one can argue whether the focus on more applied (e.g. management) issues is a priority. Nevertheless, in order to communicate the importance of meroplankton studies and engage other sectors of the society, it is crucial to highlight the relevance of meroplankton for maintaining biodiversity, replenishing populations, and ensuring connectivity of the deep ocean and other marine ecosystems, as well as for the recovery of impacted environments (e.g. by resource exploitation). We need to identify objectives that will make sustained meroplankton observations achievable. Part and parcel to this is targeting stakeholders who can support this type of baseline long-term studies. Some environmental and regulatory agencies are already mentioning connectivity as a necessary component of ongoing policy and conservation.

Our claim for the imminent need of technological developments to study meroplankton may also be attractive for innovation and by economically-driven players. Autonomous samplers for continuous monitoring, networked robotic systems for adaptive sampling, integrated systems for semi-automated processing and identification of meroplankton samples, as well as supervised machine-learning methodologies for imagery and metagenomics data processing were identified as some of the most relevant technological advances needed to underpin the irreplaceable humanpower in meroplankton research.

The existing in situ components and land-based infrastructures of the Global Ocean Observing System are indispensable for implementing the envisaged global and regional schemes for sustained observing systems, as are many other initiatives for ocean observations at national and international levels. The EMSO-ERIC and the Ocean Networks Canada observatories were identified as the most approachable consortia for collaborations in the short term.

After a lively debate, the workshop participants agreed in the continuity of the working group, namely by writing a collaborative manuscript with the focus first and foremost on (i) scientific questions, and (ii) on how technological innovation can assist in building up a sustained deep-ocean meroplankton observation system to gain a better knowledge of fundamental ecological processes such as connectivity and resilience. Nevertheless, the manuscript should also be relevant to potential stakeholders by showcasing examples of applications and presenting potential technological solutions.
Hands-on session

The workshop continued with a hands-on session to define the scope and main outline of the foresight paper, assign lead and writing assignments for the envisaged chapters, and establish expected deadlines. The participants worked in small breakout groups to draft initial contributions and further refine the structure of each chapter. Finally, the progress made by each breakout group was briefly presented by their respective rapporteurs.

Wrap up

The workshop ended with a wrap up by the conveners. Coincidentally, the paper "Global Observing Needs in the Deep Ocean" (Levin et al. 2019) was published in the last day of the workshop, and we could verify that the results of our discussions were in line with the recommendations issued by this paper. In fact, this is one of the many publications recently issued with a focus on ocean observations that are creating a momentum for the implementation of ocean observing systems (see reference list). A sustained deep-ocean meroplankton observation system will contribute to an enhanced knowledge of the oceans within current international frameworks and directives (e.g. EC 2019, UN 2020). It was also stressed that it is an opportune time to launch this enterprise which fits well into the Vision of the UN Decade of Ocean Science for Sustainable Development: “Ocean Science that is fit for purpose - The Decade will address both deep disciplinary understanding of ocean processes and solution-oriented research to generate new knowledge. This knowledge will support societal actors in reducing pressures on the ocean, preserving and restoring ocean ecosystems and safeguarding ocean-related prosperity for generations to come. Building on existing research and initiatives, the Decade will boost international cooperation to developing scientific research and innovative technologies, connecting ocean science with societal needs” (UNESCO 2020).

Funding program

This report is an outcome of the Euromarine Foresight Workshop Advances in Ocean Biological Observations: a sustained system for deep-ocean meroplankton (1804call_FWS_Meroplankton). The workshop was co-funded by the Euromarine Consortium, InterRidge and the French National Agency for Research (ANR) under the programme "Investissement d'Avenir" IsBlue(the Interdisciplinary Graduate School for the Blue planet), grant number ANR-17-EURE-0015. Universidade de Aveiro contributed in kind while Ifremer and CSIC sponsored the co-conveners (FP and MCH, respectively). Participants from Universidade de Aveiro were funded through Centro de Estudos do Ambiente e do Mar (CESAM, UID/AMB/50017/2019) and by Fundação para a Ciência e Tecnologia (FCT/MCTES) through national funds. The workshop and report are contributions to the project EMSO-PT (Observatório Europeu Multidisciplinar do Fundo do Mar e Coluna de Água – Portugal, PINFRA/022157/2016, proposal n.º 01/SAICT/2016) funded by Programa Operacional Competitividade e Internacionalização (POCI), through ERDF, and by FCT/MCTES through national funds.
Hosting institution

Universidade de Aveiro - Departamento de Biologia & Centro de Estudos do Ambiente e do Mar (CESAM)

Conflicts of interest

None

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Supplementary material

Suppl. material 1: Abstracts of keynote talks doi

Authors: Birch J., Gula J., Metaxas A. and De Leo F.
Data type: abstracts of keynote talks
Download file (23.69 kb)