



Workshop of WP5 to present the results of the application of innovative monitoring

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1. Objective of the Workshop

The main objective of WP5 in DEVOTES is to develop innovative monitoring systems to refine and deepen our ability to detect environmental/biotic variables utilized in marine monitoring, including both, instrumental and methodological approaches. This will fill the gaps identified in WP1 in which current monitoring systems in the Regional Seas and Member States (MS) were interrogated. The innovative approach using instrumental tools include earth observation and ecosystem model output, automated ship-board instruments and underwater image analysis devices, whilst the approach using innovative methodologies are based on the selection and application of the latest developments in molecular biology (second generation sequencing, high-throughput approaches) to identify species (from microorganisms to large invertebrates) or to detect genes indicating, for instance, the presence of toxins. These innovative approaches based on instruments and methods are directed to the identification of key variables and indicators at the three main levels of biodiversity investigated: a) species and functional groups, b) habitats and c) ecosystems.

The objective of the WP5 workshop was to present the innovative techniques tested and methodologies applied until now by the partners to determine the convenience of the use/application of the newly developed instruments and methods to environmental monitoring.

The Workshop of WP5 was organized by the partner ICM-CSIC with the aim to present the results on the application of innovative monitoring techniques that are developed in DEVOTES. The Workshop was held in Barcelona, from 13-14 October and was conceived as a forum for discussion among partners working in different methodologies. The main goals were to: 1) present the current knowledge on the methods, focusing on biodiversity quantification; 2) identify which methodologies are more promising; 3) define future research directions; and 4) debate the most appropriate approaches and methods. Twenty participants took part of the workshop which was comprised of 16 presentations covering the most relevant innovative methods in marine monitoring. Seven presentations were done by DEVOTES partners (i.e. CSIC, AZTI, CONSIMA, CIMA, CEFAS, Akvaplan-niva) and four presentations were done by invited speakers of different institutions (i.e. CSIC, EMBL, NEIKER-Tecnalia). The invited speakers were experts in molecular methodologies. After each talk, discussion focused on the main gaps of knowledge, methodologies drawbacks and research needs.

2. Summary of the methods presented

The methods presented at the workshop were mainly based on:

1. Molecular approaches for plankton biodiversity quantification in monitoring systems
2. Microarrays applied to the identification of Harmful Algal Blooms
3. Metabarcoding macroinvertebrates for assessing benthic communities
4. The use of chemotaxonomic techniques for the retrieval of phytoplankton diversity and size structure
5. Flow cytometry as a tool to monitor the status of aquatic environments and the use of On-line Flow cytometry for high frequency measurement of phytoplankton diversity
6. Visual monitoring of sea-floor biodiversity
7. Biosensor to monitor bivalve behavior / Underwater hyperspectral imaging

A nice summary on Plankton indicators and marine policy: new developments and opportunities for DEVOTES was also presented.

The utility of molecular approaches for biodiversity quantification in monitoring systems were the dominant topic in the workshop. Several partners involved in DEVOTES and invited speakers described sampling area, sampling strategy and sample treatment. Detailed protocols about the method used for DNA extraction, PCR and primers used have been provided along with a description of sequencing techniques and bioinformatic tools chosen. The other dominant methodology was the flow cytometric approach to classify phytoplankton and bacterioplankton according to their size and optical properties

3. Conclusions

Partners discussed the convenience of the different techniques and methodologies. Common for most traditional monitoring initiatives is a labour-intensive and often very time-consuming methodology. Advantages in the new methods, as detection of a higher diversity of organisms, cost-effective and possible automatization in the near future were discussed. Some of the new technologies have become more readily available and affordable and now we have an enormous technological development for environmental and biodiversity assessment and monitoring

Some disadvantages in the new methods, such as incompleteness of databases for some taxonomic groups, definitions and delimitation of OTUs cut-off (in the molecular methods), methodological biases, training requirement, and dependence on offhouse sequencing services, were also discussed.

In conclusion, the challenge for the monitoring systems lies in using these new methodologies efficiently, and in such a standardized way as to complement existing methods, to attain at new

monitoring system, to understand biodiversity and its fluctuations in time and space, and how it is influenced by human beings.

4. Agenda

Day 1

1. **Isabel Ferrera – CSIC, Spain.** Meeting presentation. WP5 partner member.
2. **Esther Garcés – CSIC, Spain.** CSIC goals and contributions to DEVOTES WP5. WP5 partner member.
3. **Ramiro Logares – CSIC, Spain.** An intro to high-throughput sequencing and its application in metagenetics, meta(genomics), single-cell genomics and RNA-seq; perspectives for monitoring. Invited speaker.
4. **Shinichi Sunagawa – EMBL.** Towards Ocean Eco-Systems Biology. Invited speaker.
5. **Naiara Rodríguez-Azpeleta – AZTI-Tecnalia, Spain.** Genomics in Monitoring. WP5 partner member.
6. **Anders Lanzen – NEIKER-Tecnalia, Spain.** Current-generation Amplicon Sequencing for environmental monitoring - experimental design, a simple example and data analysis tools. Invited speaker.
7. **Pablo Sánchez – CSIC, Spain.** Metagenomics: How much should we sequence? Resolution implications for marine microbial diversity monitoring. Invited speaker.

Day 2

8. **Ramon Massana – CSIC, Spain.** Microbial eukaryotes include a wide diversity of targets for monitoring. WP5 partner member.
9. **Isabel Ferrera – CSIC, Spain.** Comparison of different high-throughput sequencing methodologies for marine planktonic biodiversity monitoring and assessment of ecosystem health status. WP5 partner member.
10. **Marco Berzano – CONSIMA, Italy.** Innovative monitoring tools in marine monitoring: Applying microarrays (phylochips) to investigate the biodiversity of Harmful Algal Blooms (HABs). WP5 partner member.
11. **Priscila Goela – CIMA, Portugal.** The use of chemotaxonomic techniques for the retrieval of phytoplankton diversity and size structure in the Southwest Coast of Portugal. WP5 partner member.
12. **Rodney Forster – CEFAS, UK.** Plankton indicators and marine policy: new developments and opportunities for DEVOTES. WP5 partner member.
13. **Sabine Cochrane - Akvaplan-niva, Norway.** Industry-linked visual monitoring of sea-floor biodiversity in areas of environmental concern. WP5 partner member.
14. **Josep M Gasol – CSIC, Spain.** Flow cytometry as a tool to monitor the status of aquatic environments. WP5 partner member.
15. **Veronique Creach – CEFAS, UK.** On-line Flow cytometry for high frequency measurement of phytoplankton diversity. WP5 partner member.
16. **Lionel Camus - Akvaplan-niva, Norway.** Biosensor to monitor bivalve behaviour / Underwater hyperspectral imaging. WP5 partner member.

All participants. Group discussion and closure.

5. List of participants

Name	Institution
Albert Reñé	CSIC (Spain)
Anders Lanzen	NEIKER (Spain)
Caterina R. Giner	CSIC (Spain)
Esther Garcés	CSIC (Spain)
Eva Flo	CSIC (Spain)
Isabel Ferrera	CSIC (Spain)
James Strong	UHull (UK)
Jordi Camp	CSIC (Spain)
Josep M. Gasol	CSIC (Spain)
Lionel Camus	Akvaplan-niva (Norway)
Marco Berzano	CONISMA (Italy)
Naiara Rodríguez-Ezpeleta	AZTI Tecnalia (Spain)
Pablo Sánchez	CSIC (Spain)
Priscila Goela	CIMA (Portugal)
Ramiro Logares	CSIC (Spain)
Ramon Massana	CSIC (Spain)
Rodney Forster	Cefas (UK)
Sergei Danchenko	CIMA (Portugal)
Shinichi Sunagawa	EMBL (Germany)
Veronique Creach	CEFAS (UK)
Sabine Cochrane (by webcam)	Akvaplan-niva (Norway)