

Environmental observatories as data providers: the contribution of Slovenian marine station to LifeWatch-ERIC

National Institute of Biology
Marine Biology Station Piran
Slovenia

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LifeWatch ERIC Scientific Community Meeting
Rome, 27-29 May 2019 | CNR



MORSKA BIOLOŠKA POSTAJA PIRAN
MARINE BIOLOGY STATION PIRAN



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LIFEWATCH-SI CONSORTIUM (ESTABLISHED IN 2015)

LifeWatch SI – Slovenian Consortium



- Research Centre of the Slovenian Academy of Sciences and Arts (ZRC SAZU)



- National Institute of Biology – Marine Biology Station Piran (NIB -MBP)



- Park Škocjanske jame: Škocjan Caves Park Public Service Age (PSJ)



Park *Škocjanske jame*,
Slovenija

- Slovenian Museum of Natural History (PMS)



- Society for Cave Biology (TULAR Cave Laboratory)



- University of Maribor (UM)



- Slovenian Forestry Institute (GIS)



- University of Ljubljana – Biotechnical Faculty (UL-BF)



- University of Nova Gorica (UNG)



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LIFEWATCH SLOVENIA ACTIVITIES

Co-organizer of the 1st working meeting
LifeWatch-ERIC on “**Data Centres**” in Ljubljana,
Slovenia (13-14 December 2018)



National Institute of Biology
MARINE BIOLOGY STATION PIRAN

RESEARCH CHALLENGES

Coastal Sea Research (*P1-0237, 2015-2019*)

improve the
fundamental
understanding of the
functioning of
marine coastal
ecosystems

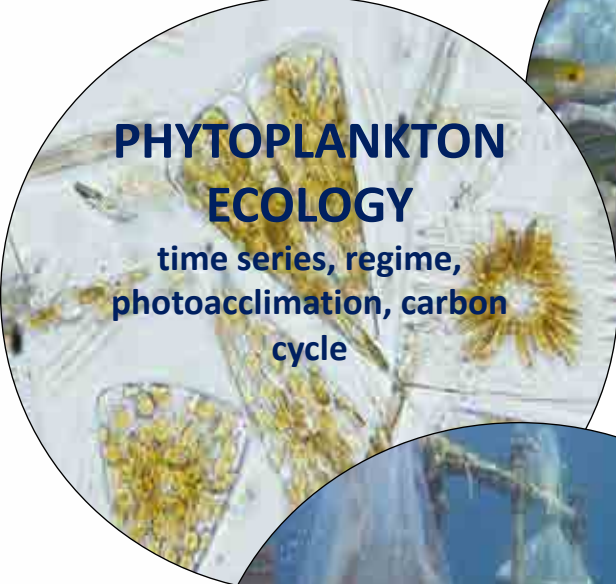
development of
efficient sea
observation systems

study of
anthropogenic
impacts on the
coastal area

Marine Biology Station Piran (MBS) is the leading unit for marine ecosystem research in Slovenia working on basic and applied research

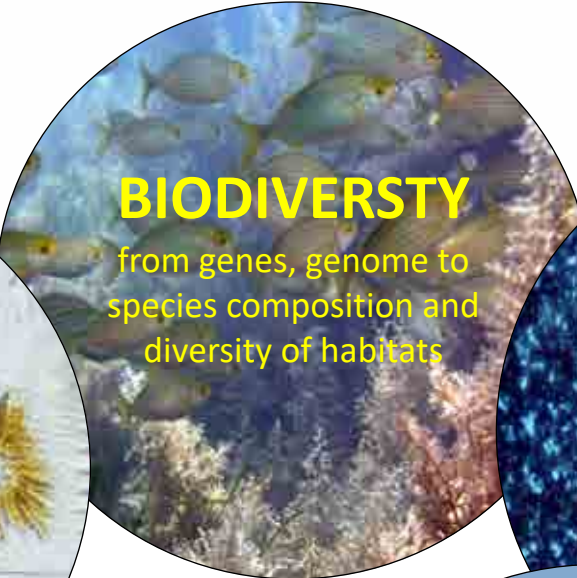


RESEARCH



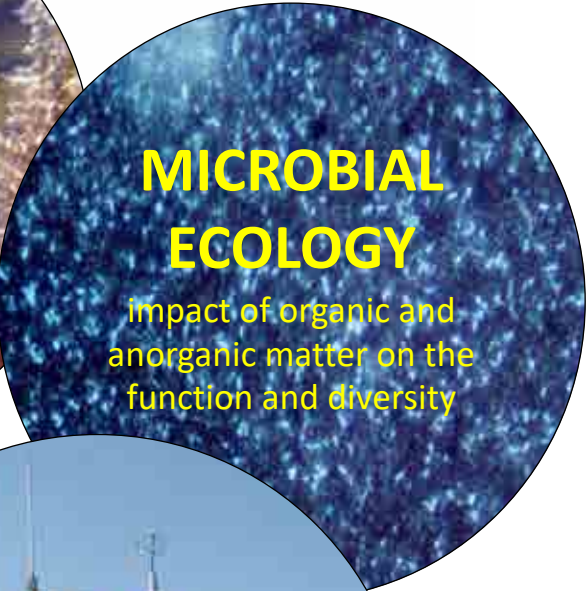
PHYTOPLANKTON ECOLOGY

time series, regime,
photoacclimation, carbon
cycle



BIODIVERSITY

from genes, genome to
species composition and
diversity of habitats

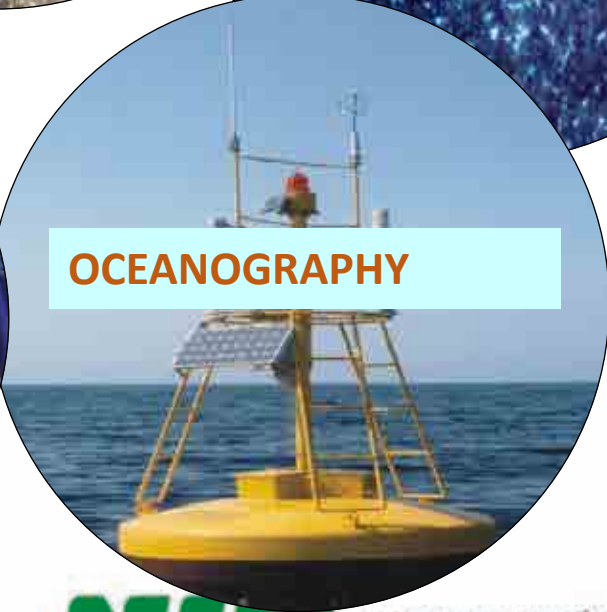


MICROBIAL ECOLOGY

Impact of organic and
anorganic matter on the
function and diversity



ANTHROPOGENIC IMPACTS



OCEANOGRAPHY

Marine Biology Station Infrastructure

Infrastructure

instrumental centre IC MBP

diving base (scientific diving, sampling, underwater video)

Laboratories (molecular biology, microbiology, chemistry lab, wet lab, growth chambers...)

Dormitory and lecture rooms

dormitory (13 rooms, 2 apartments)

congress hall (75 seats)

lecture rooms



Infrastructural centre MBP (IC MBP)



Oceanografic bouy Vida



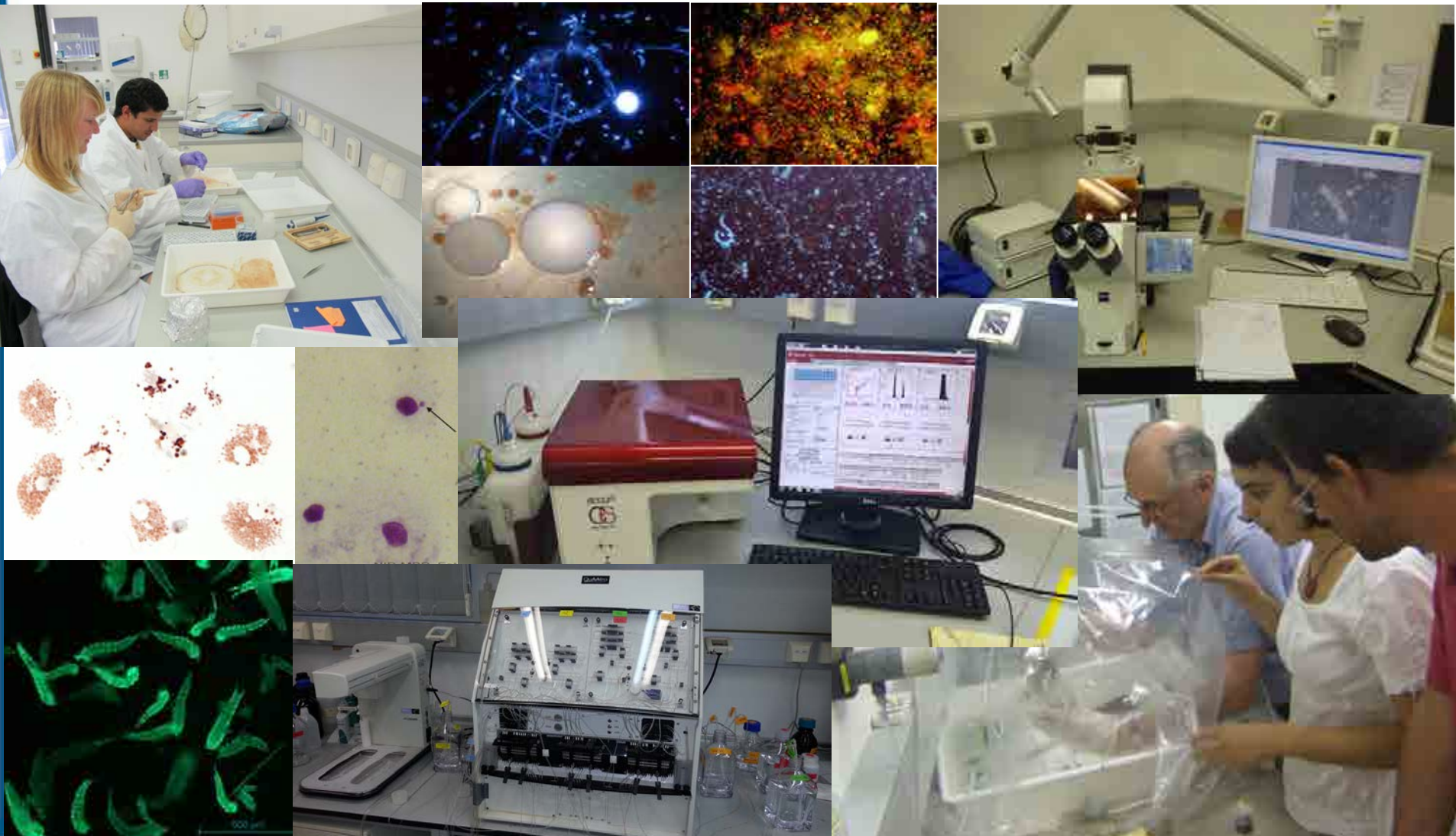
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Infrastructural centre MBP (IC MBP) Sampling and data collection services



Infrastrukturni center MBP (IC MBP)

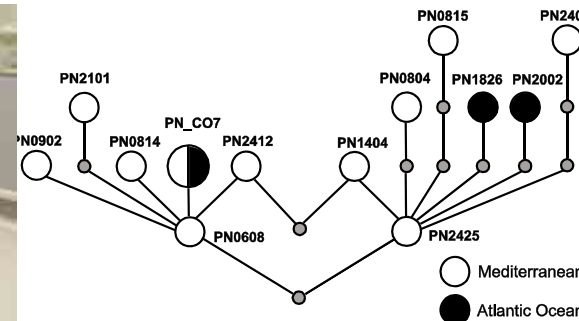
Laboratory analyses and services



MARINE BIOLOGY STATION PIRAN

Access to biological resources

Access to Biological Resources			
Taxon	note	Geographical Coverage	Temporal Coverage
Macrofauna (soft bottom)	specimens	Slovenian coastal sea	2006-ongoing
Macrofauna (hard bottom fouling community)	specimens	Slovenian coastal sea	2008-ongoing
	specimens	Slovenian coastal sea	2010-ongoing
Macrofauna (epifauna of coastal detritic bottom)	specimens, DNA	Slovenian coastal sea	2010-ongoing
Opisthobranchia	specimens, DNA	Slovenian coastal sea, Northern Adriatic	2006-ongoing
Elasmobranchs	Specimens	Slovenian coastal sea	2006-ongoing
Macroalgae	specimens	Slovenian coastal sea	
Fish	fixed environmental samples	Slovenian coastal sea	2010- ongoing
Phytoplankton		Slovenian coastal sea	
Zooplankton	DNA samples	Atlantic Ocean, Mediterranean Sea	2004-ongoing
Scyphozoa	DNA samples	Mediterranean Sea	2005-ongoing
Scyphozoa		Baltic Sea, Atlantic Ocean, Mediterranean Sea	
Scyphozoa	DNA samples	Mediterranean Sea	2005-ongoing
Cnidaria	tissue and DNA samples	Adriatic	2010-ongoing
bacteria		Gulf of Trieste, northern Adriatic	2009-2012



Research of the fouling communities

Structure (species composition and coverage) of different fouling communities:

- Mussel cultures, underwater defence structures etc.

Comparing different taxonomic **methods**:

- morphology based, photography (complementary), DNA and metabarcoding



Photography as a complementary method



Determination of organisms using standard morphological approach

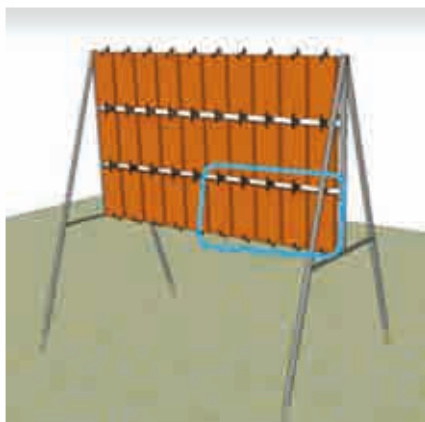


Epifauna of the mussel cultures



Experiments on structure, succession and function on artificial underwater structures:

- clay tiles,
- mesh cylinders with different properties (cotton, plastic, different opening sizes)
- ARMS (Autonomous Reef Monitoring Structure)



Metal constructions with clay tiles



Mesh cylinders with different properties

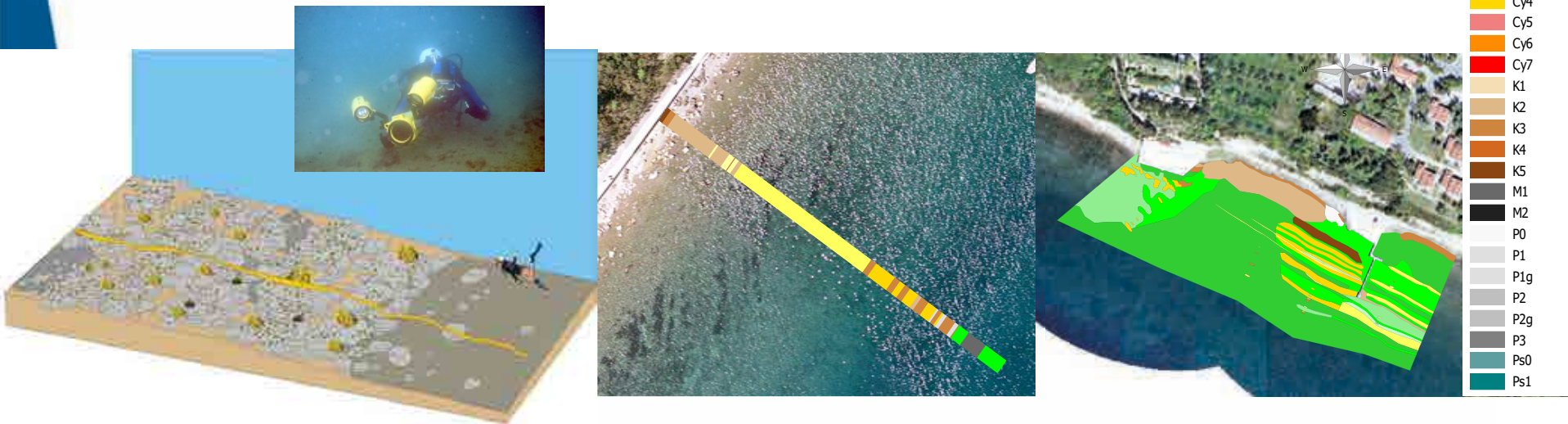


ARMS (Autonomous Reef Monitoring Structure)



Underwater visual census survey and habitat mapping

- Ongoing since 1998
- Parallel and vertical transects mainly in infralittoral zone
- Data on habitat types, microhabitat types, fish community, benthic invertebrates, algae
- Used for research and environmental management (e.g. MSFD, Habitat Directive)



Soft bottom benthic invertebrates

- Ongoing since 2005
- Van veen grab (0,1 m²)
- Used for research and environmental management (e.g. WFD, MSFD, Habitat Directive)



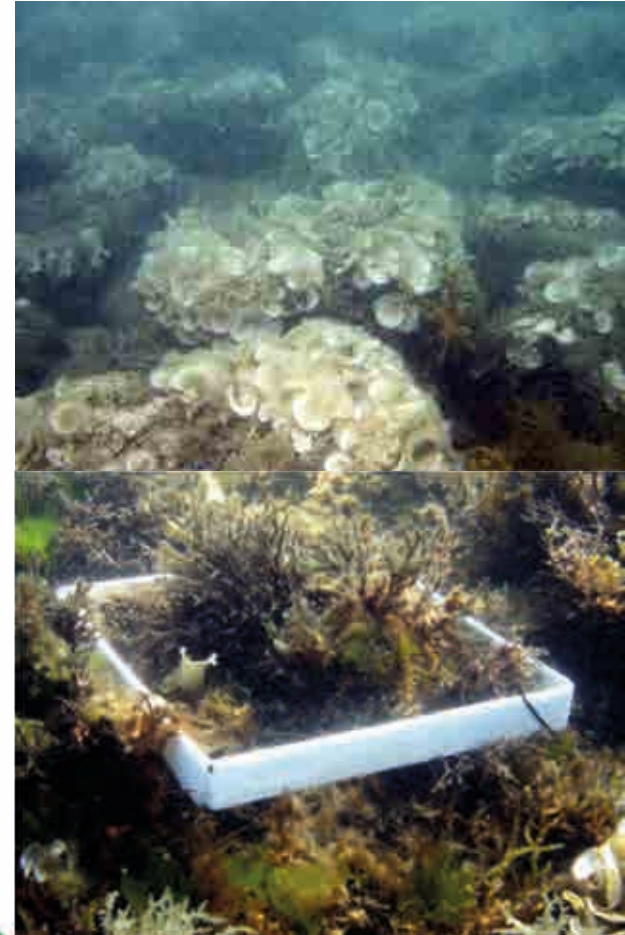
Macroalgae

- Ongoing since 2005
- Scraping within frame 20x20 cm
- Used for research and environmental management (e.g. WFD, MSFD, Habitat Directive)



Slovenian experience in applying WFD and MSFD: the case of benthic macroalgae

- ❖ Samples were collected at a depth range of 1 to 4 m at 51 sampling stations (151 samples)
- ❖ Macroalgal sampling on hard bottoms-on a quadrat 20x20 cm, the minimal sampling area for the Mediterranean infralittoral communities (Montesanto & Panayotidis, 2001)
- ❖ identification of taxa – most were identified to the species level
- ❖ Indices (MediSkew frequency distributions of ln-transformed leaf lengths of adult and intermediate leaves (n=1200) for *Cymodocea nodosa*:

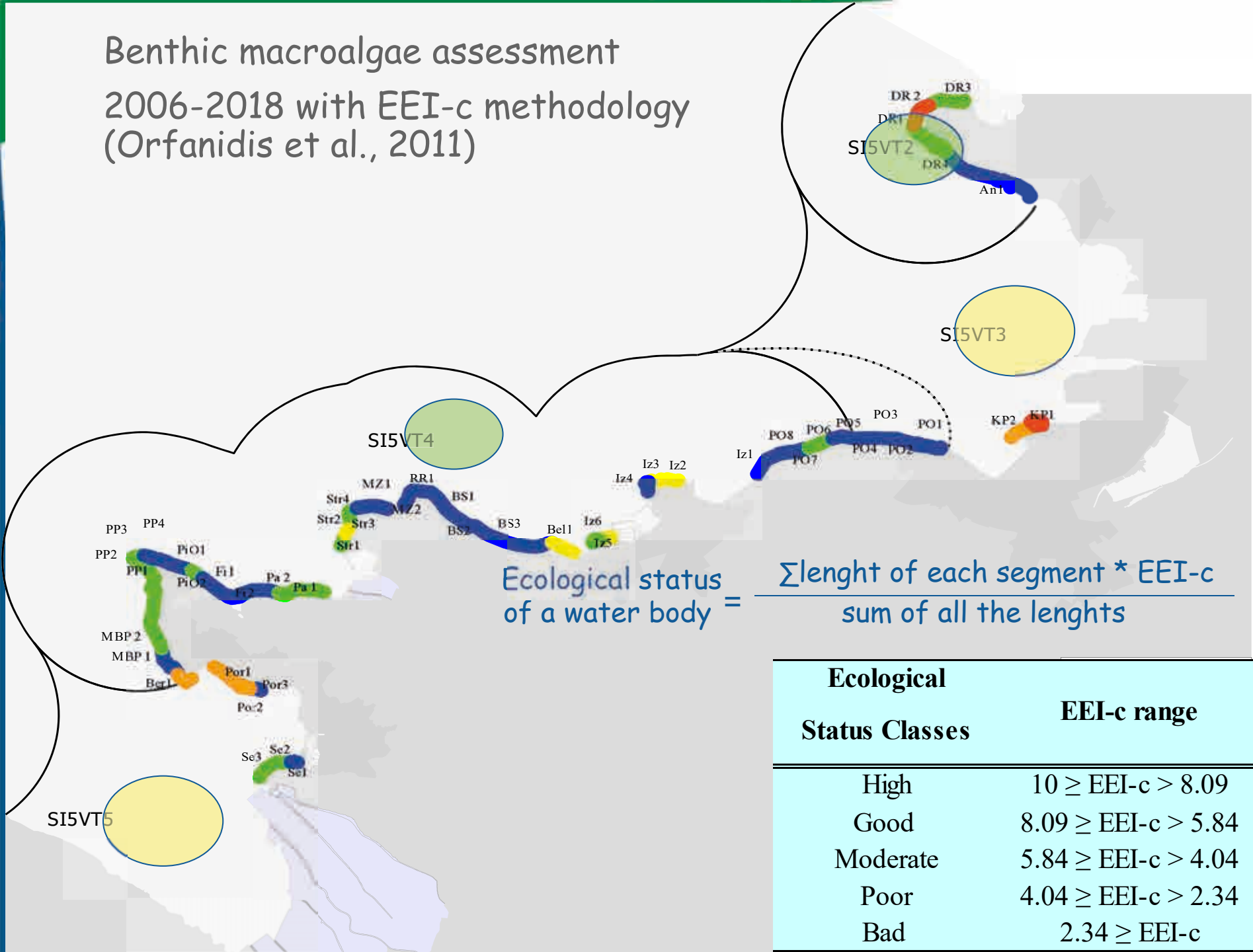


Benthic macroalgae - reference conditions

Strunjan Nature Reserve

- high ecological status of macroalgae;
- low pressures and impacts -
- natural coastal environment well preserved;
- no sources of anthropogenic pollution;
- no non-indigenous species that can affect autochthonous species and habitats.

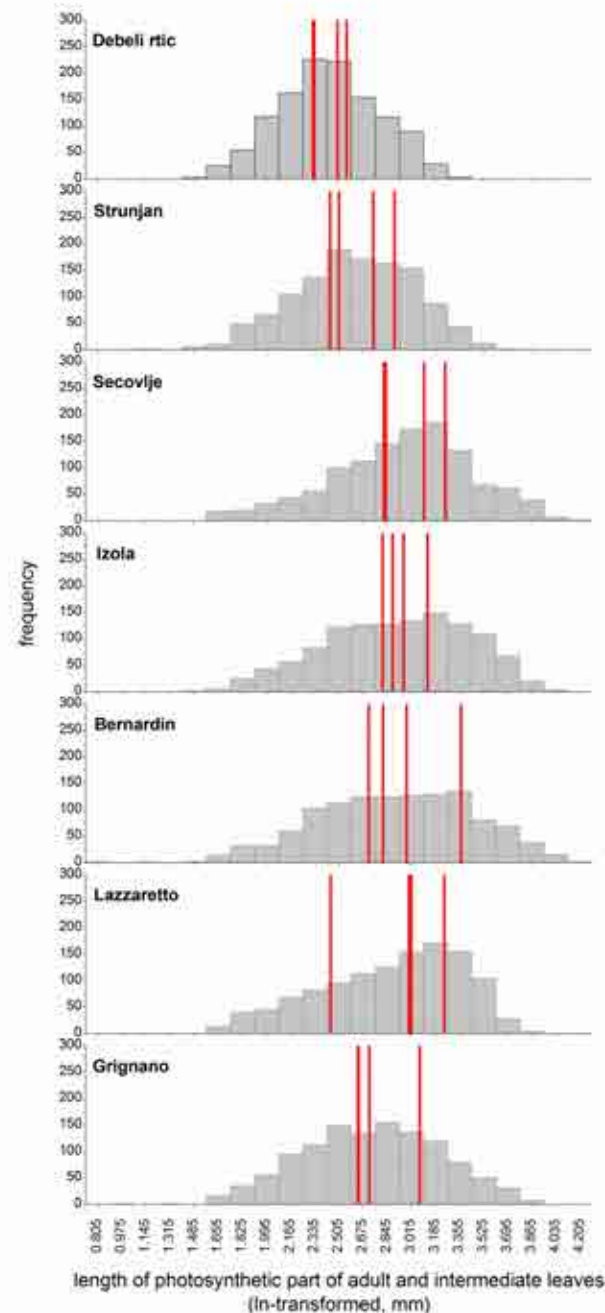
Benthic macroalgae assessment 2006-2018 with EEI-c methodology (Orfanidis et al., 2011)



MEDISKEW DEVELOPMENT - 1ST STEP

Frequency distributions of ln-transformed leaf lengths of adult and intermediate leaves (n=1200) for *Cymodocea nodosa*:

different shapes,
different dispersion →
median values quite diverse
among areas.



Red vertical
lines: median
values for 4
areas in every
meadow

Phytoplankton

Data:

- **biomass – Chlorophyll a concentrations** (several stations in the Gulf of Trieste, monthly data, different periods, the longest dataset from 1984 onwards for 1 station)
 - Use of Chl a metrics (Giovanardi et al. 2017) for the assessment of the ecological status of coastal waters (WFD, MSFD)
- **community composition and abundances** (5 stations, different periods, monthly data, the longest dataset from 1989 onwards for 1 station)
- **Toxic phytoplankton - HAB** (3 mussel farming areas, from 1994 onwards)
 - National nominee for uploading HAEDAT database of HAB events
- **primary production and photosynthetic parameters** (one station, period 2009 – 2011, fortnightly data)
 - Talaber et al. 2014, 2018
- **phytoplankton pigments** (one station, HPLC, fortnightly data, since 2005)

Projects:

- **MedCis:** Activity 1- Develop common approaches and methodologies towards GES and targets in Mediterranean regional/subregional scale - Subtask 1.1.1 Towards GES definition of *pelagic habitats* according to plankton communities
 - Common database on phytoplankton communities from Adriatic, Ionian and Aegean Seas
 - analysis of the behaviour of a set of diversity indices across spatial and trophic gradients
- **MedRegion:** Activity 5- Implementation of the new GES Decision: Biodiversity

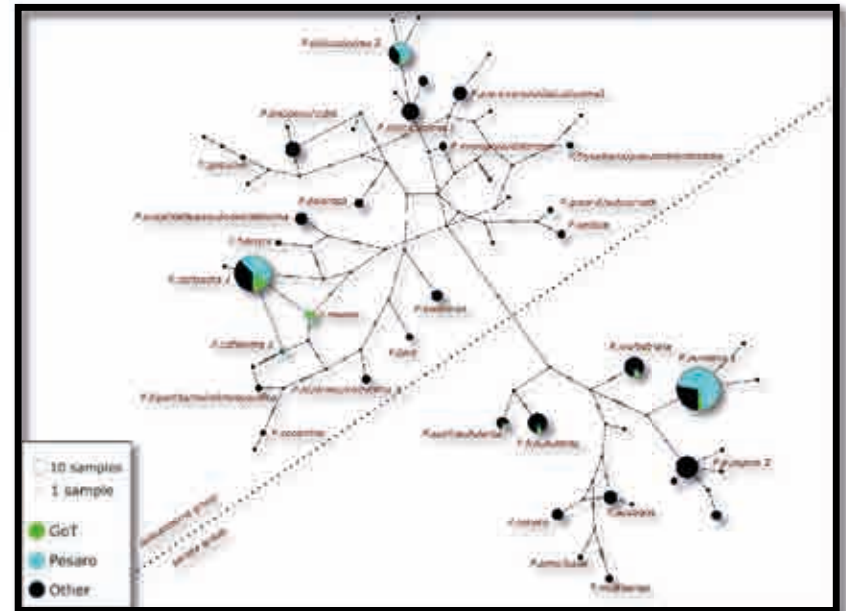
Pseudo-nitzschia: cultures, morphology, genetics



P. calliantha



P. mannii

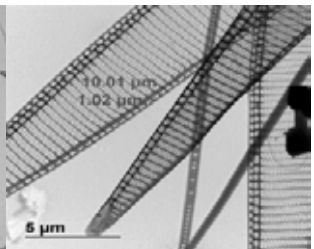
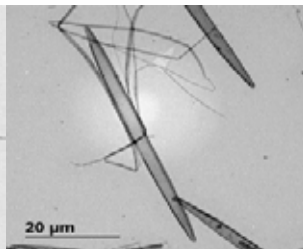
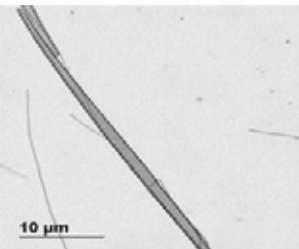
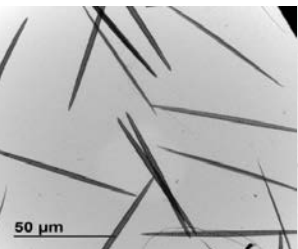
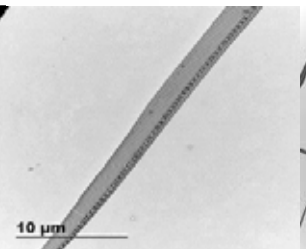
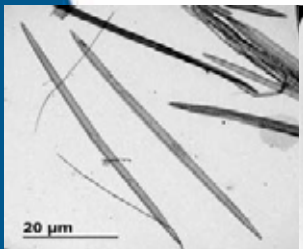
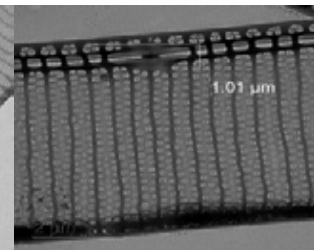
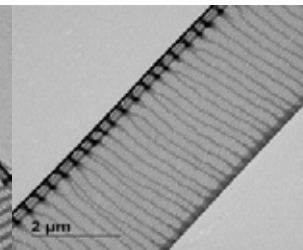
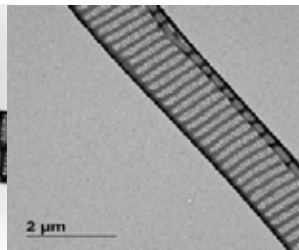
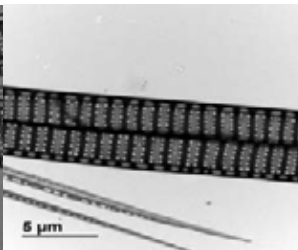
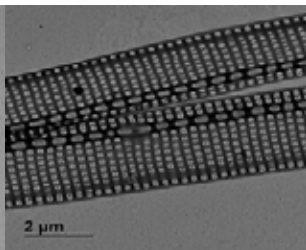
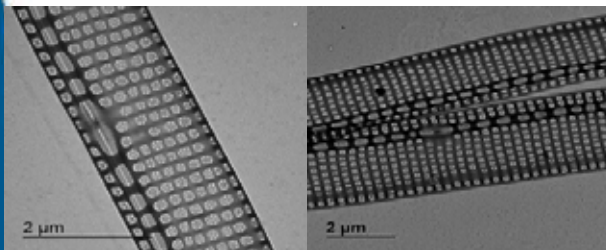


P. pungens

P. delicatissima

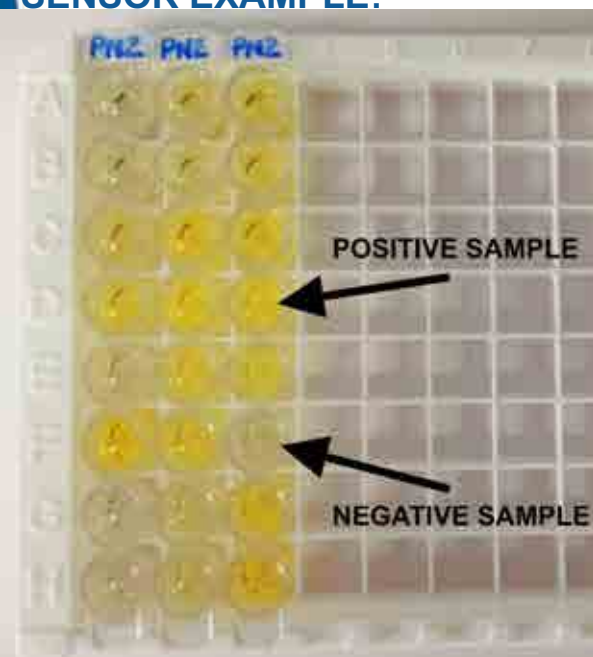
P. multistriata

P. fraudulenta

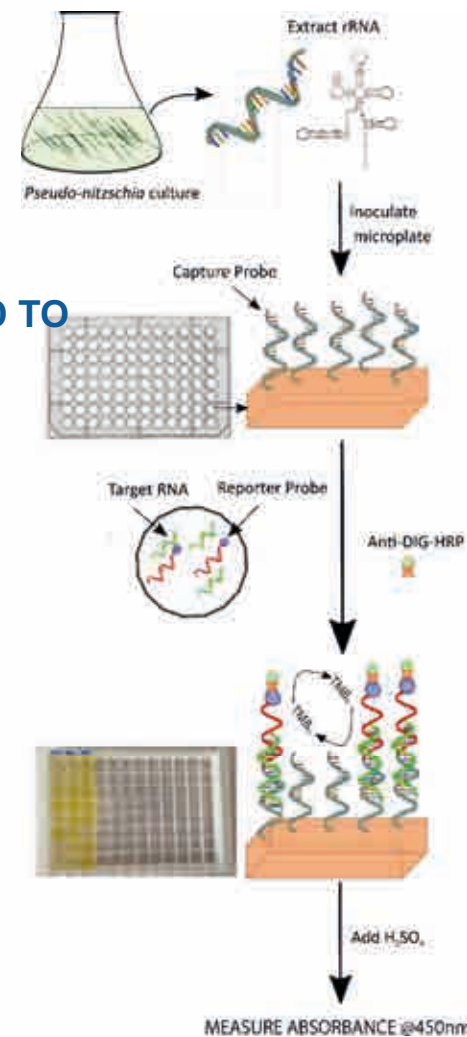


ELISA for detecting *Pseudo-nitzschia* rRNA in environmental samples

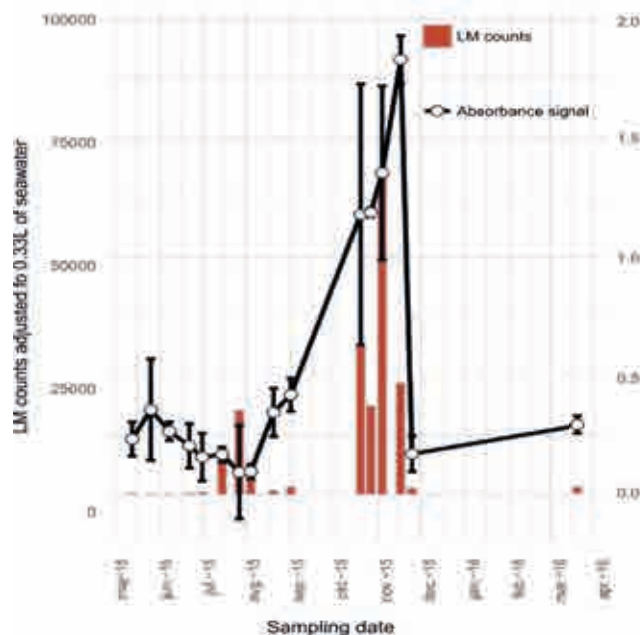
SENSOR EXAMPLE:



PROCEDURE:



SENSOR PERFORMANCE COMPARED TO MICROSCOPY:



Marine microbial ecology

- ▶ **community dynamics → specific bacterial groups** involved in specific mechanisms and interactions to study:
 - ▶ function of selected bacterial groups
 - ▶ bacterial physiology
- ▶ **Interactions:**
 - ▶ marine microbes – primary or secondary producers
 - ▶ marine microbes - anorganic & organic matter pool
 - ▶ marine microbes – oceanographic conditions
- ▶ **Microbes and response to antropogenic impacts:**
 - ▶ selected pollutants,
 - ▶ pathogens (viruses and bacteria)
 - ▶ testing new detection methods

Methods

Molecular methods:

- taxonomic identification of specific bacterial groups (16S bacterial rRNA sequence)
- DGGE
- RT-PCR
- High-throughput sequencing

Experiments:

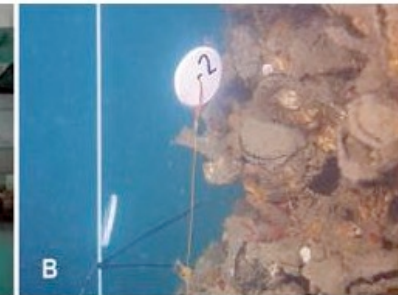
- micro- and meso-cosm experiments (*in vitro* and *in situ*) in which specific bacterial groups are exposed to different regimes
- use of diffusion chambers

Structure → function of specific bacterial groups:

- bacterial ecto-enzymatic activities
- bacterial ecto-enzymatic activities on single cell level (using microscopy techniques)
- biochemical transformations of organic matter (proteins, lipids, polysaccharides)
- bacterial respiration
- use of radioactive labeled substrates

SCYPHOZOAN RESEARCH

- mass appearances records from 1790 onward
- phylogeography and population studies



Malej *et al.* (2012) Can.Biol.Mar.53:337-342

Historical records of scyphozoan blooms Northern Adriatic

Blooms:

1790 (P. noctiluca)
1875-1922 (R.pulmo)
1909 - 1915 (A. aurita)
1981 - 1986 (P. noctiluca)
2004 - 2007 (P. noctiluca, A. aurita)

shortened periodicity in last decades
(A. aurita, R. pulmo)

Climate associated effects:

Regime shifts
Phenological changes
Distribution expansions

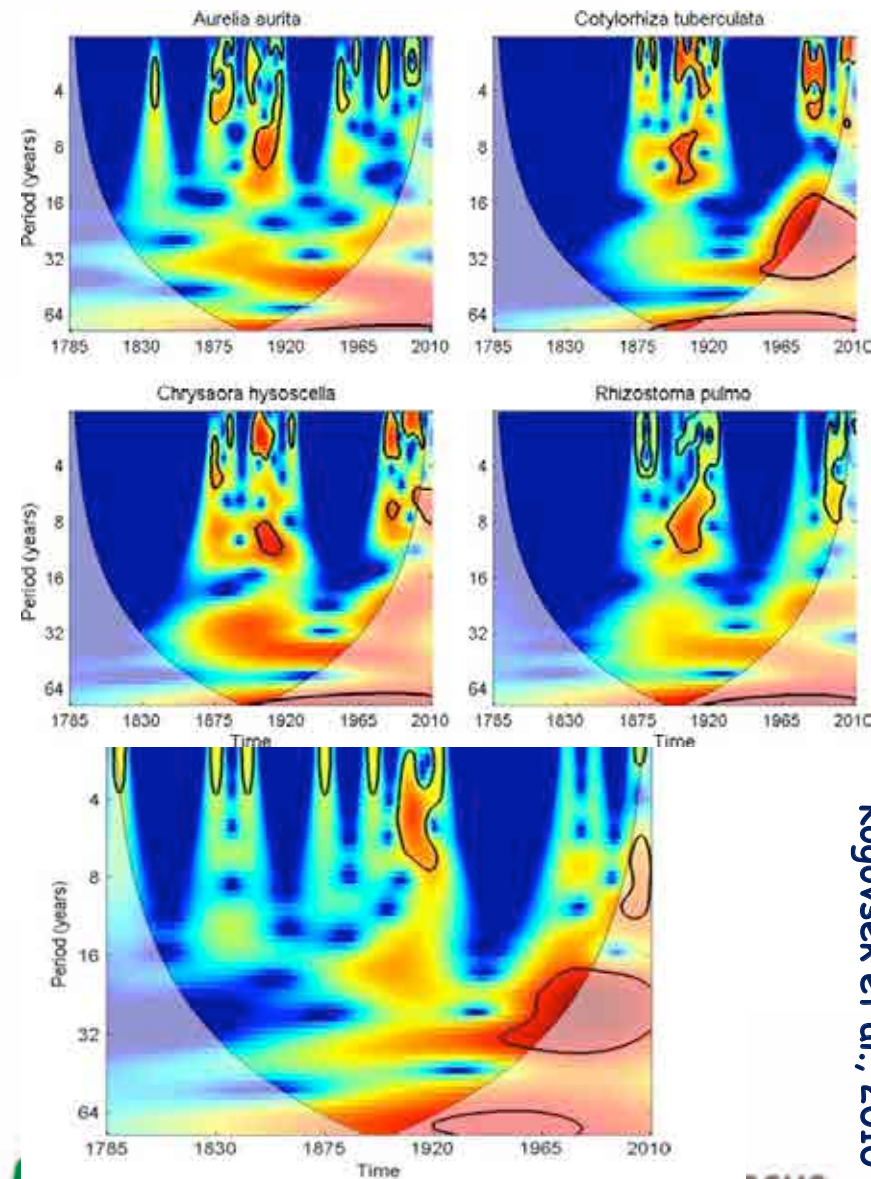
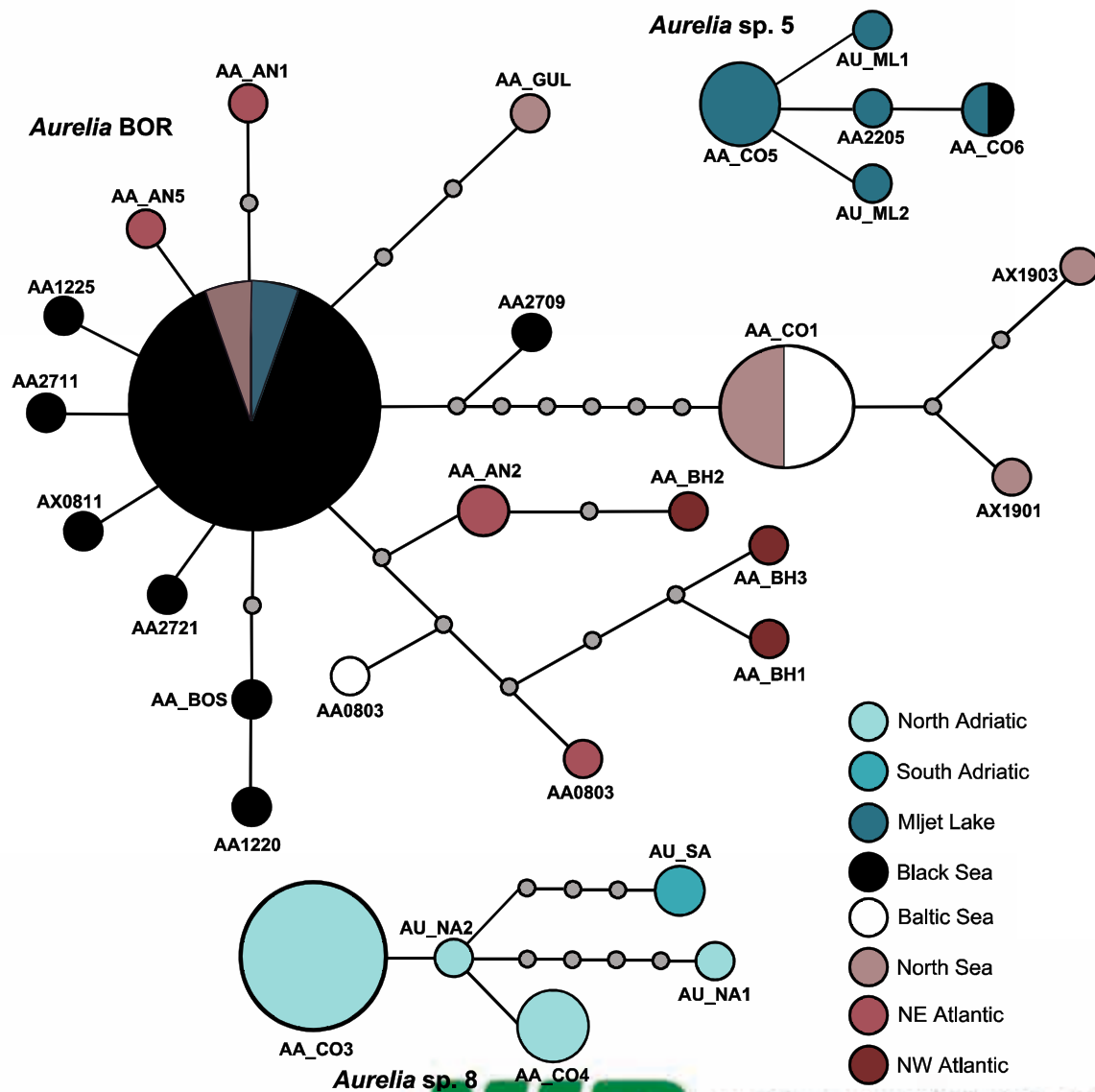
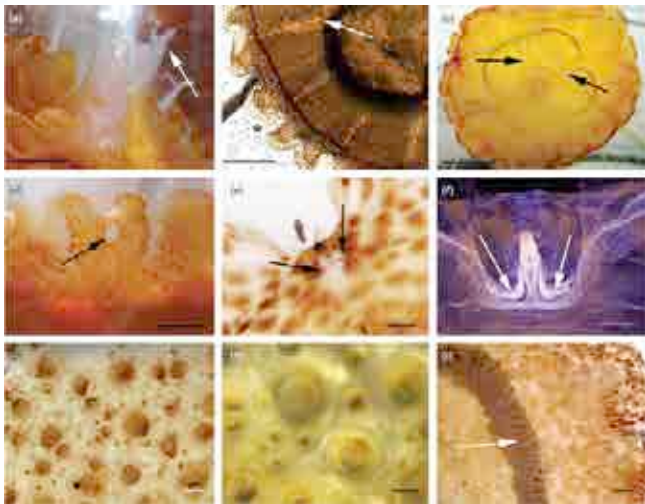


Fig. 4 Wavelet spectrum of the *Pelagia noctiluca* time series in the northern Adriatic Sea

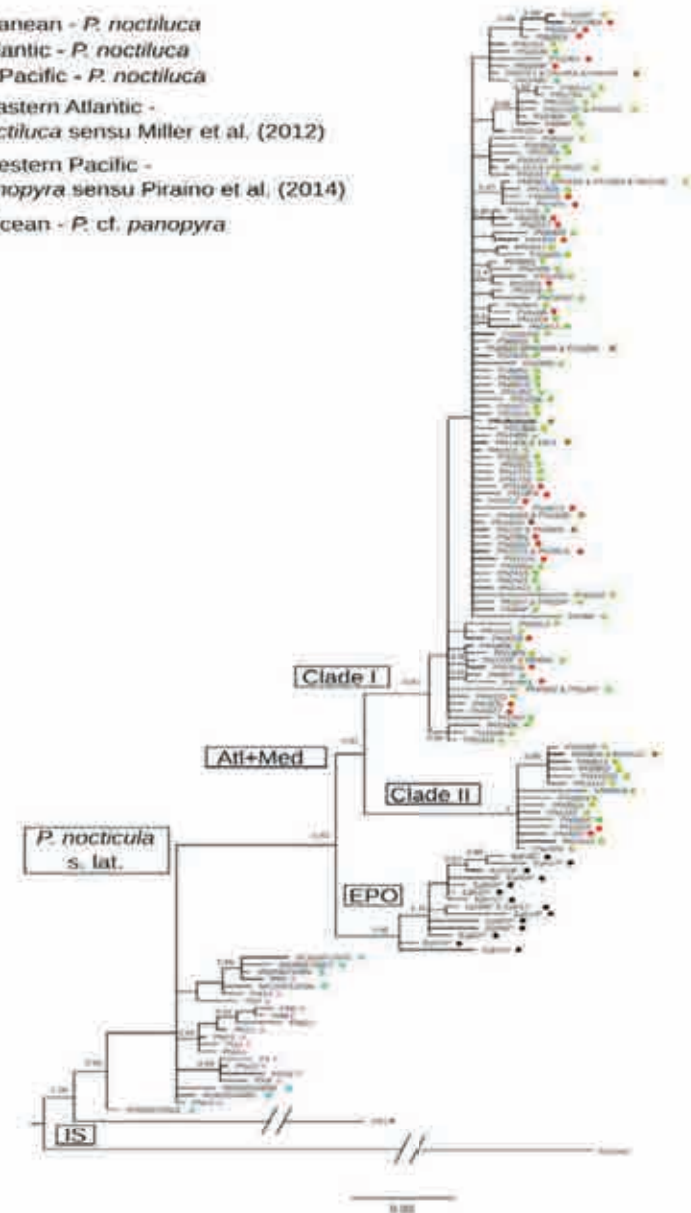
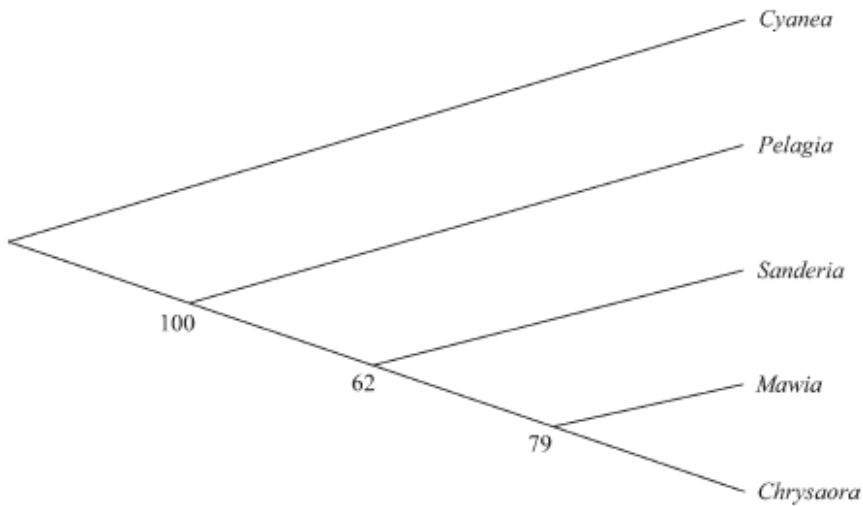
Phylogeography: *Aurelia* spp.,



Pelagiidae: *Pelagia noctiluca*, *Mawia benovici*



- Mediterranean - *P. noctiluca*
- North Atlantic - *P. noctiluca*
- Eastern Pacific - *P. noctiluca*
- South-eastern Atlantic -
P. cf. noctiluca sensu Miller et al. (2012)
- South-western Pacific -
P. cf. panopyra sensu Piraino et al. (2014)
- Indian Ocean - *P. cf. panopyra*



Pelagia noctiluca: niche modelling



Genomic Observatories

OSD 2019 (2015-

Ocean Sampling Day (2019)



COUNTDOWN HAS BEGUN TO OCEAN SAMPLING DAY 2019!
(Event date: June 21)



ARMS (Autonomous Reef Monitoring Structure)



Promotion of science, citizen science

Publishing



Open Day



Prometej znanosti 2011, 2016



25 - 29 September 2017

Piran, , Slovenia



International networking

CIESM



LTER site



Experts for harmful algae bloom (HAB)

UNEP/MEDPOL (Barcelona convention)



EmodNET

national expert boards



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Acknowledgement

Dr. Borut Mavrič

Dr. Janja France

Dr. Martina Orlando Bonaca

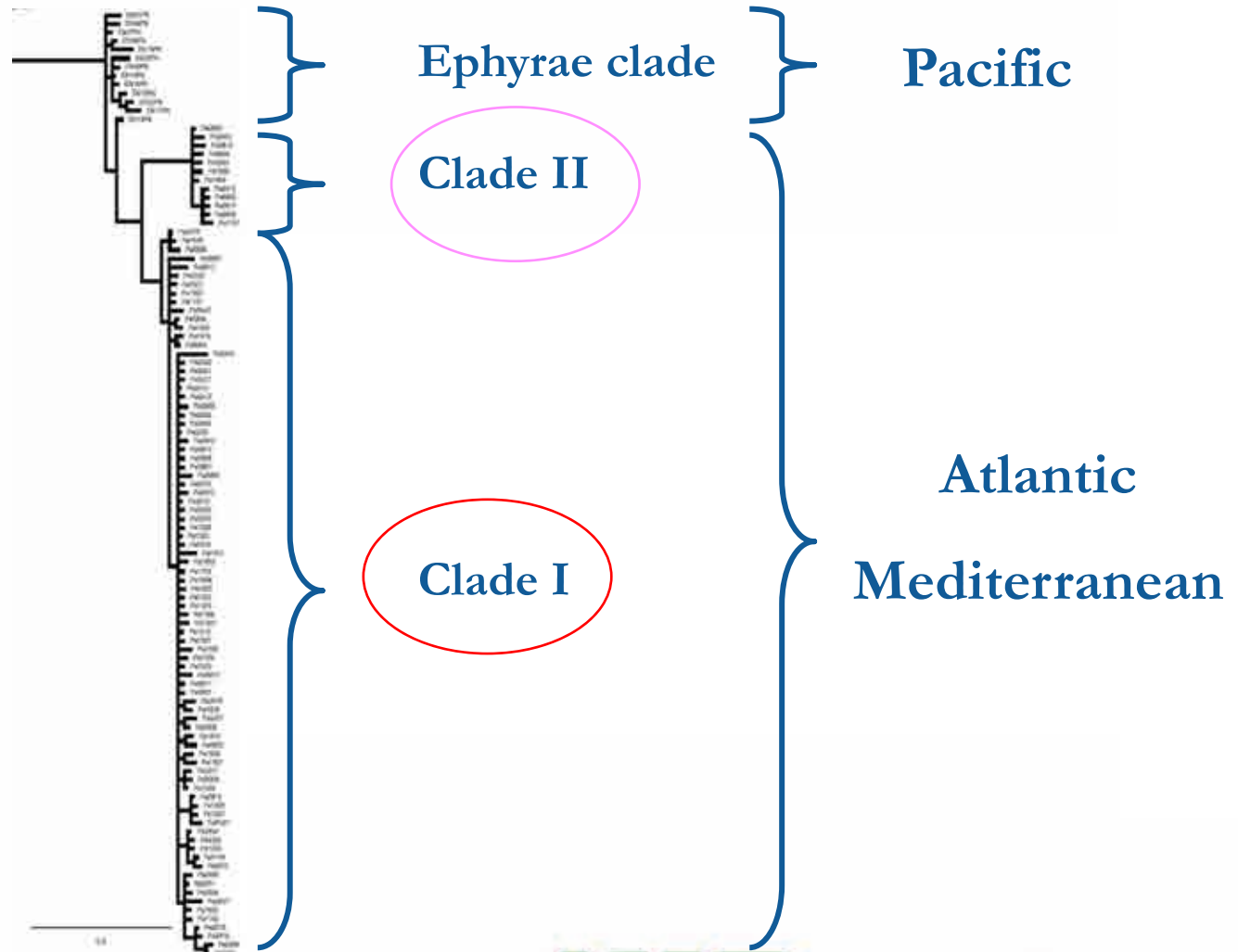
Dr. Patricija Mozetič

Timotej Turk Dermastia



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Phylogenetic relationship between Pacific and Atlantic-Mediterranean populations



Consensus tree of Bayesian phylogenetic inference for COI

Analysis of the most recent blooms was complemented with environmental descriptors (temperature, salinity, pH, chlorophyll a, zooplankton dry weight and major river discharges)

Adriatic over the last 200 years, with two major periods of jellyfish proliferations. The first period in the years around 1910 was characterised by significant periodicity of 8–12 years for each species, while the second period from the 1960s onwards was characterised by a shortened significant periodicity of less than 8 years.

Pelagia noctiluca fluctuations were analysed in greater detail for the last four decades, revealing significant periodicities of *10 years, 2.5 years, 8–14 months, and 8 months. The significantly marked periodicity of about 10 years in the *P. noctiluca* spectrum indicates a pattern similar to that observed in the western Mediterranean. Wavelet analysis showed that the periodicity of occurrence of five jellyfish species has shortened in recent decades and the recurrence of blooms has increased, particularly for *A. aurita* and *R. pulmo*

Semi-quantitative

data are available from fishery surveys that record jellyfish as by-catch (Brodeur et al., 1999; Graham, 2001) and have proved useful in assessing trends in jellyfish distribution, biomass, and abundance



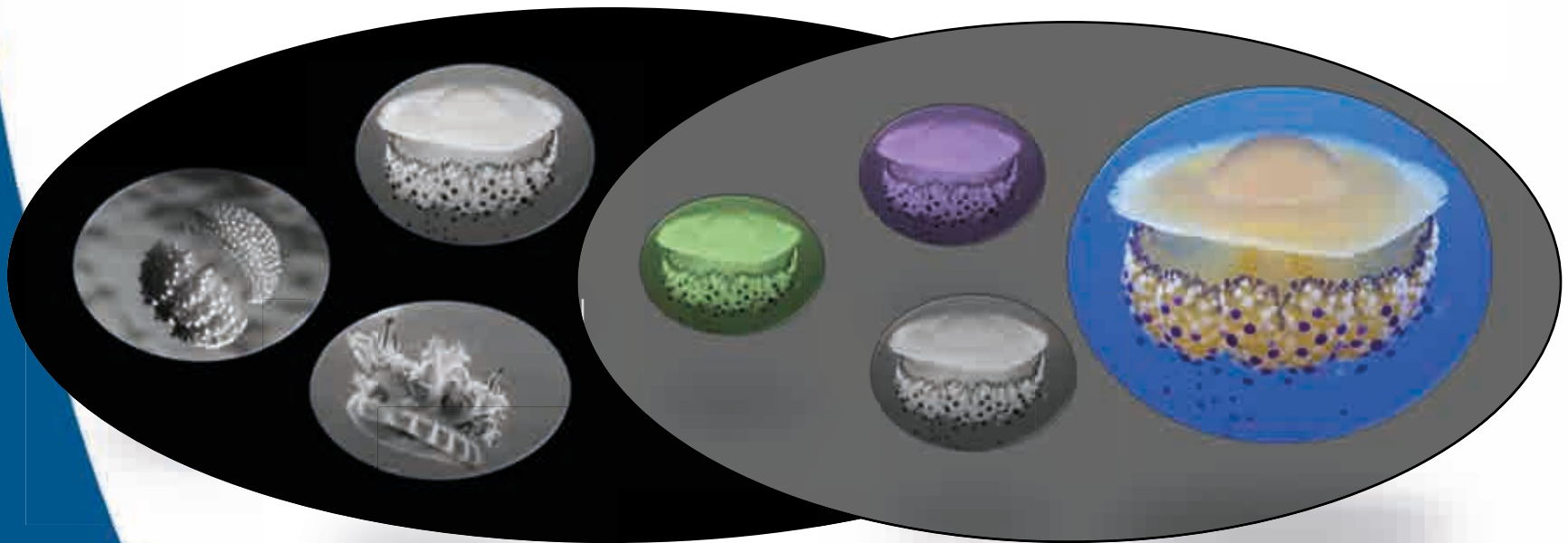
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Identification and variability of symbionts in Scyphozoa

Scyphozoa species

Within
species

Individual



SYMBIOTIC JELLYFISH SYMBIODINIUM

Mastigias sp.



A₃

LaJeunesse 2001

*Cassiopea
xamachana*



(A₁, A₃, B₁, C₃ in
juvenile)

A₁

*Cassiopea
frondosa*



A₁

LaJeunesse 2001

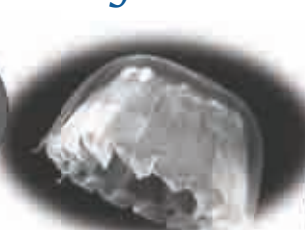
*Cassiopea
andromeda*



A₁

Lampert et al., 2011

*Linuche
unguiculata*



A₄

LaJeunesse 2001

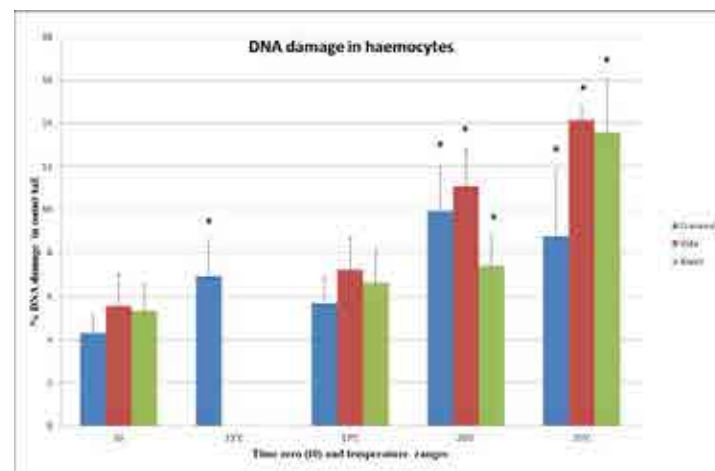
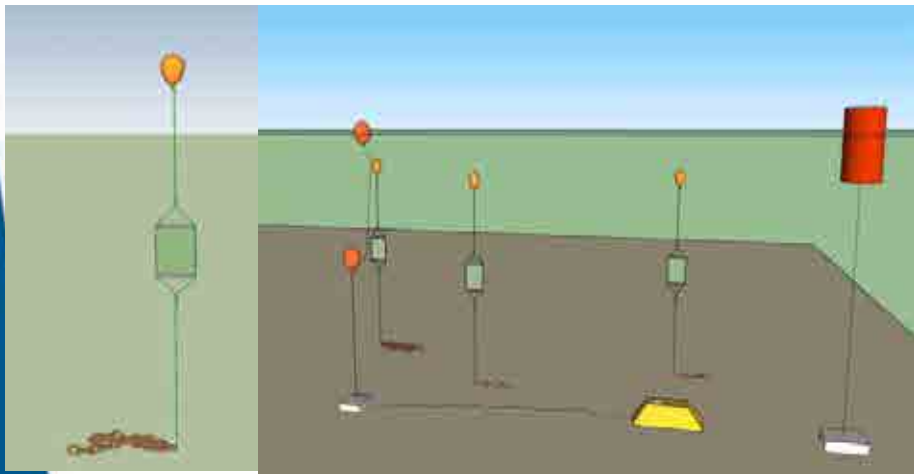
*Cotylorhiza
tuberculata*



A₁,

**Temperate
A, B**

Ecotoxicology data



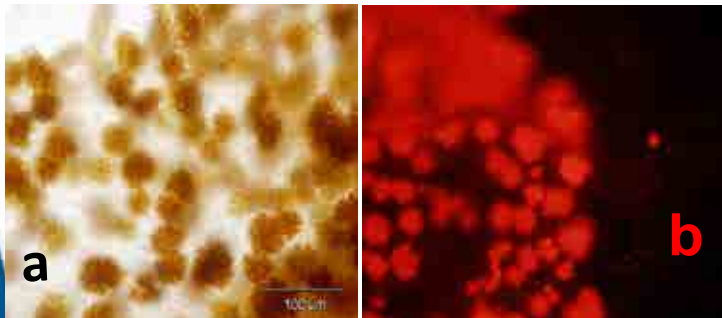
Molecular characterization of symbionts in Scyphozoa



Cotylorhiza tuberculata



Cassiopea xamachana



Symbiodinium isolated from *Cotylorhiza tuberculata*

- a) under light microscopy and
- b) under epifluorescent microscopy (photo V. Turk).



a

b

- a) Cultivated motile *Symbiodinium* strain 4 fixed in 1% acid Lugol,
- b) cysts in culture (photo A. Beran).

Genomic Observatories – metabarcoding distributed Genomics Observatory across the partnership and beyond of which the data are available for virtual access (VA).

-cross cutting with other research

Barcoding, metabarcoding

Verifying databases for taxonomic reference barcodes

inter-calibration of classical biodiversity data and genomics data (metabarcoding, meta-transcriptomics, etc.).

Joint Research Activity 1 (JRA1) will foster the application of genomics technologies at Long-Term Ecological Research Network (LTER) sites.

Research encompasses populating and verifying databases of taxonomic reference barcodes, harmonisation of metabarcoding standard operating procedures (SOPs) across the consortium so that the resulting data can be compared across the partnership, and inter-calibration of classical biodiversity data and genomics data (metabarcoding, meta-transcriptomics, etc.). The final objective will be the establishment of a distributed Genomics Observatory across the partnership and beyond of which the data are available for virtual access (VA).



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The northern Adriatic is a shallow sea with large salinity and temperature variations and strong seasonal vertical stratification.

The physical properties and dynamics of the northern

Adriatic water masses are characterised mainly by

atmospheric forces due to wind stress, heat flux, and

fresh water inputs by major rivers along the Italian

coast (Cushman-Roisin et al., [2001](#)). The northern

The most detailed early records of jellyfish in the northern Adriatic probably were of *R. pulmo* blooms, because its high abundance during blooms interfered with fishing. During the first period (1875–1922), blooms of this species showed significant periodicities of 4 and 6–8 years; it has been present every decade after the 1950s, with increasing blooms in the last two decades. It was most abundant during the last decade from 2003 to 2006, after which its numbers decreased markedly.

Collaboration with universities



Jožef Stefan International Postgraduate School, doctoral study Biosensors

University of Maribor

University of Ljubljana

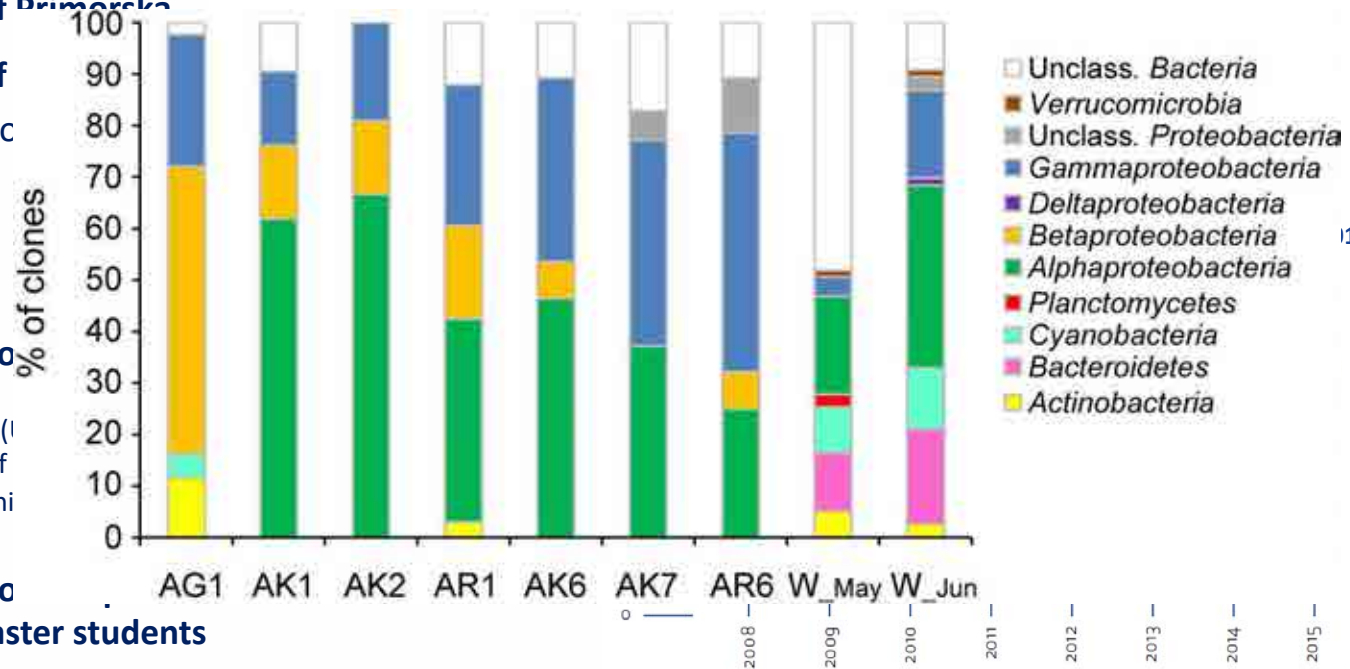
University of Primorska

University of
(marine ecol

Student's pro

- Fish fraud (I
- Methods of
- Microorgani

Erazmus scho
IMBRSea master students



(17)



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DEVELOPMENT OF A NEW INDEX FOR THE ASSESSMENT OF THE *CYMODOCEA NODOSA* (UCRIA) ASCHERSON MEADOW'S STATUS → MEDI-SKEW

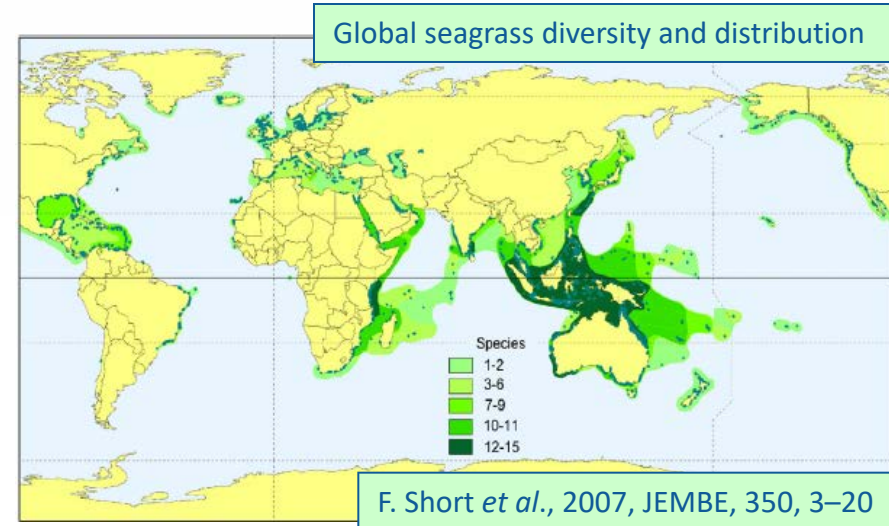
Worldwide deterioration of coastal areas

Assessment of the status in EU waters:

WFD, 2000/60/EC

MSFD, 2008/56/EC

HD, 92/43/EEC



Quality Elements → seagrasses → indicators
→ biotic indices

- CYMOX index (Oliva *et al.*, 2012) – composed of several metrics
- CymoSkew (Orfanidis *et al.*, 2007, 2010) – only photosynthetic leaf length

