Book of Abstracts

BOOK OF ABSTRACTS

VLIZ MARINE SCIENCE DAY

VIVES, BRUGES
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PREFACE

This is the 'Book of Abstracts' of the 17th edition of the VLIZ Marine Science Day, a one day event that was organised on 3 March, 2017 in VIVES, Brugge.

This annual event has become more and more successful over the years. With more than 400 participants and more than 100 scientific contributions, it is fair to say that it is the place to be for Flemish marine researchers and for the end-users of their research. It is an important networking opportunity, where scientists can meet and interact with their peers, learn from each other, build their personal professional network and establish links for collaborative and interdisciplinary research.

Marine scientists from all Flemish universities and scientific institutes – and representing all marine science disciplines – have contributed to this volume. The book thus illustrates the diversity, quality and relevance of the marine sciences in Flanders (and Belgium): it provides a beautiful and comprehensive snapshot of the state-of-the-art of marine scientific research in Flanders.

Pre-doc and post-doc scientists present their research in an exciting way and communicate their fascinating science – and its importance to society – to the wider public. We thus hope to demonstrate the excellence of Flemish marine science and to increase its national and international visibility.

The volume of research that is presented here holds a great promise for the future. It shows that marine science is a very lively discipline in Flanders, and that a new generation stands ready to address the grand challenges and opportunities that our seas and oceans represent.

New this year are the Brilliant Marine Research Ideas, an initiative sponsored through the philanthropy scheme of VLIZ. We are proud to announce that an initial batch of 4 ideas will be sponsored. We'll hear about the results in the next edition of the Marine Science Day.

I want to congratulate all participants with their contributions, and I invite them all to become members of VLIZ and to actively participate in our events and activities in the future.

Bruges, 3 March 2017
Prof. Dr Jan Mees
General Director VLIZ

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ORAL, DEMO AND POSTER PRESENTATIONS

Development of diffusive gradients in thin films (DGT) passive samplers for simultaneous measurement of Platinum, Palladium, Rhodium and Mercury in surface water

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Increasing anthropogenic activity often has detrimental effects on human health and the environment due to the accompanying emissions of toxic compounds. The increased application of Platinum Group Elements (PGEs) (Pt, Pd, Rh, Ru, Os and Ir) in the last decades, especially as car catalysts but also in other applications, makes it necessary to monitor the concentration of these elements in the environment, investigate their environmental transformations and bioavailability. Mercury (Hg) is also recognized as one of the most toxic trace elements, whose natural cycle has been altered by anthropogenic activities. Complex biogeotransformations result in different chemical species, with varying toxicities and mobility, which need close monitoring. The concentration of these elements usually extremely low in the aquatic environment, which makes the analysis challenging.

As the toxicity, bioavailability and the cycle of environmental contaminants can strongly be influenced by their chemical speciation, in recent years, the importance of speciation analysis has been recognized by the environmental monitoring and assessment community, leading to the development of an increasing number of speciation techniques. The in situ passive sampling technique diffusive gradients in thin films (DGT) as a speciation tool is based on the binding of labile metal species on a resin gel layer via the diffusion through a diffusive hydrogel (agarose or polyacrylamide) using Fick's Law. The concentration gradient built between the bulk solution and the resin gel makes pre-concentration of solutes possible. Using Fick's law, the time-weighted average concentrations of labile metal species can be obtained in situ. This technique has been widely used to assess trace elements such as Cd, Cu, Ni, Pb, Co, Zn in aquatic systems, but never been applied to test PGE elements until now.

The aim of this study was to develop the DGT technique for the assessment of PGE and Hg using two novel resins R14 and R20, which were designed specifically for above elements. This implies that the binding of the PGEs to the resin is strong, irreversible, almost instantaneous and the accumulated metals amounts are well below the capacity of the resin. The method development involves several different steps: 1) selection of an appropriate diffusive gel 2) the selection of an appropriate resin or binding phase for the PGEs and Hg, 3) development of an efficient elution method for the PGEs and Hg from the resin gel, 4) evaluation of the linear response in function of the deployment time, 5) determination of diffusion coefficients for the PGEs and Hg in the diffusive gel, 6) study the selectivity of the tested resins gels, 7) the accumulated metal amount is well below the capacity of the binding gel, 8) fast kinetics of the resins gels.

Agarose diffusive gel (AGA) (1.5% agarose) was chosen for lower interaction with PGEs and Hg, adequate blank values and linear response (R2 = 0.99) in function of the time were obtained for the new resins gels and diffusion coefficients could be determined. An aqua regia and thiourea in hydrogen chloride elution methods gave a recovery for PGEs and Hg over 90% and 80% for the R20 and R14 resins gels, respectively. The selectivity test showed these two resins have higher selectivity to PGE and Hg than other trace elements even though they are at very high concentration level and the analysis of PGEs and Hg by sector field ICP-MS optimized. The new resin gels showed capability of accumulation concentration of PGEs and Hg of each hundred times higher than their reported concentrations in the aquatic environments.

Preliminary deployments in the Zenne River and UZ hospital effluent, Brussels, Belgium, showed that Pt, Pd, Rh and Hg can be quantified by the DGT technique using both evaluated resins in fresh water.

Keywords: DGT; PGEs; Hg; SF ICP-MS; diffusive coefficient; surface water; speciation

Consumption of discarded plaice (*Pleuronectes platessa*) by epibenthic scavengers: common starfish (*Asterias rubens*) and hermit crabs (*Pagurus bernhardus*)

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Beam trawl fisheries catch a large amount of fish and benthic invertebrates in the North Sea. The unwanted catches, called discards, are returned to the sea and provide a food source for several marine organisms such as benthic scavengers. The European Commission established the landing obligation in the Common Fisheries Policy, which prohibits fishermen to discard quota-regulated fish species. The reduced availability of discards in the sea floor means a reduction of feeding opportunities for benthic scavengers, and may result in knock-on effects on scavengers that are competing for the decomposing fishery discards. The aim of this study is to quantify the biomass of discarded plaice (*Pleuronectes platessa*) that is consumed over a period of time by two epibenthic scavengers, hermit crabs (*Pagurus bernhardus*) and common starfish (*Asteria rubens*). The effect of intra and interspecific competition between scavengers and the effect of live infaunal prey as feeding alternative to the discarded plaice was simulated in laboratory conditions.

Epibenthic scavengers, infaunal community and sediment were sampled at a coastal station in the Belgian part of the North Sea, between January and April 2016. Three experiments were carried out. In the first experiment, the objective was to test the intra and interspecific competition between hermit crabs and starfish by comparing differences in weekly consumption of discarded plaice during a seven week period. In the second experiment, the objective was to test the effect of starfish density and the presence of an additional food source (infauna) on the decomposition of discards. In the third experiment, the objective of the previous two experiments was combined. We compared the intra and interspecific competition between hermit crabs and starfish and the effect of an additional food source (infauna) on the weekly consumption of discarded plaice.

The biomass of dead plaice was significantly reduced when hermit crabs were present, but was unaffected by the presence of starfish. Hermit crabs were not influenced by competition with starfish and consumed on average 12 % more biomass than starfish. More surprisingly, the consumption of dead plaice by starfish did not differ significantly from the control treatment, where scavengers were absent. The experiments also suggested that infauna was an alternative food source for hermit crabs, as the consumption of dead plaice was significantly higher when laboratory tanks were deprived of infaunal prey.

Our laboratory experiments indicate that dead plaice is consumed more by hermit crabs than by starfish in a one-week period. If plaice will no longer be discarded as a consequence of the landing obligation, the food availability for hermit crabs will decrease more than for starfish. The reduced food availability may be counterbalanced by infaunal prey as a food alternative for hermit crabs.

Keywords: discards; competition; hermit crab; starfish; plaice

Benthos response to physical disturbance: "The case of deep-sea trawling at the Portuguese Margin off Sines"

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Paucity of data on the ecological significance of deep-sea bottom trawling still exists. An effort to contribute information on the response of benthic communities to bottom trawling disturbance at the SW Portuguese margin, Off Sines was made by conducting two ROV dives: dive 1 (trawled) and dive 2 (non/low-trawled) and through sampling at two stations: Trawled (st 7) and non/low-trawled (st 6) both occurring at depths range (290-356m). Megafaunal densities (ind.100m) and meiofaunal abundances (ind.10cm-2), their structural distribution and composition were determined between dives and stations respectively. Sediment characteristics (i.e. grain size, %TN and %TOC) from the studied areas were investigated and correlated to these benthic functional assemblages. Results of megafaunal abundances and taxa composition significantly differed ($P < 0.0001$) between the two dives, depicting the impact of bottom trawling gear. Meiofaunal community did not show any significant differences ($P < 0.4516$) in abundance (per group) and community composition ($P < 0.1077$) between stations, although their vertical profiles significantly differed ($P < 0.0001$) suggesting the influence of oxygen requirement, quality and quantity of food supply e.g. fresh phytoplankton and phytodetritus, bottom currents flow and higher concentration of Sulphide compounds that trigger sediment biogeochemistry. Top sediment layers (0-3cm) recorded higher abundance and diversity in relation to the deeper sediment depths (3-5cm). Sediment grain size in st 7 (T) had the highest percentage of coarse sand whilst st 6 (LT) showed high percentage of fine sand. Both stations recorded low %TN and %TOC contents in the sediments.

Findings of the current study indicates that the response of megafaunal communities to the impacts of bottom trawling are more pronounced whereas no effect of trawling was observed on meiofaunal communities, thus knowledge of their interaction with fisheries will promote deep-sea habitat conservation.

Keywords: trawling impacts; deep-sea; meiofauna; megafauna; grain size; conservation

Carbon-neutral silviculture in Matang Mangrove Forest Reserve (Malaysia): a becoming reality?

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There is a growing interest in mangrove forests because of the variety of ecosystem functions, goods and services they provide (*e.g.* buffer against coastal erosion, habitat for fish and shellfish, timber and non-timber forest products). Mangroves are the most carbon-rich forests in the world (Donato *et al.2011*) and have a high economic potential yet they are facing increasing anthropogenic threats.

Matang Mangrove Forest Reserve (hereafter referred to as MMFR) is located on the northwest coast of Peninsular Malaysia (State of Perak at 04°45'N, 100°35'E) and covers approximately 40000 ha along 52 km of coastline. A large part of the area is covered by *Rhizophora apiculata*, which is the most important mangrove species in terms of commercial timber in the Asia - Pacific region (Ong *et al.* 2004). This reserve is under silviculture management by the Perak State Forestry Department since 1902. More than 30000 ha are exploited for charcoal (in 30 year cycles) and pole production (in 20 year cycles) for national and international trade. After harvesting, the area is replanted with *Rhizophora apiculata* and *R. mucronata* seedlings (Azhar *et al.* 2003).

This management process in Matang involves a series of exploitation activities that emit carbon, whereas other natural processes result in carbon sequestration.

The objective of this study is to calculate the emission of carbon produced by the exploitation activities as well as calculating the carbon sequestered by MMFR mangroves (through a meta-analysis of vegetation growth data). The final result will indicate whether or not the carbon release is outbalanced by carbon sequestration. Preliminary results will be presented, including the relative importance of various management activities with regard to carbon emissions, and the method & findings for calculating carbon sequestration by the mangrove forest.

With this project we aim to enhance the understanding of the potential of mangrove forests as key systems in effective climate change mitigation.

Keywords: mangrove forest; carbon emission; carbon sequestration; charcoal and pole production

Determination of the best otolith preparation method for aging of dab (*Limanda limanda*)

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Age determination of fish is key in fisheries biology and the management of stocks. Indeed, one of the management tools for marine biological resources is the use of stock assessments, which utilize fish length-at-age data to estimate mortality, recruitment and recommended harvest levels. Age studies can also supply other important information such as stock age structure, age at first maturity, spawning frequency, individual and stock responses to changes in the habitat, recruitment success, etc. Obviously, it is imperative to develop accurate age determination methods to be able to perform correct stock assessments. Age can be determined by different methods of which counting the growth increments formed in hard calcified tissues (such as e.g. otoliths or scales) is most commonly used. We examined and compared two different otolith preparation techniques for age determination in dab with the aim of assessing the best method for the species. The included methods were based on reading of whole (WH) and stained sections (S&S) of otoliths.

Otoliths of 296 dab from the North Sea (ICES region IVb and IVc) were collected during 2015 and analysed by two experienced readers for each method which resulted in four readings for each otolith. Readers of the otoliths were provided with metadata such as the month of capture (to assist in edge interpretation) and individual fish length at time of capture. For both methods, otoliths were read from digital images, with annual growth increments being annotated on the image using in house developed software, i.e. OtolithManager. Comparisons of the methods were made for accuracy (the proximity of the determined age to the modal age) and precision (the reproducibility of results between readers). Accuracy was evaluated by means of bias by comparing reader derived ages against the modal age. In absence of calcified structures of known age, the age readings can be compared to modal age, which is here defined as the age agreed upon between both readers for an individual structure. Precision was determined using coefficients of variation (CVs) with data calculated for each reader for both whole and sectioned otoliths. Results were also calculated for the percentage agreement to modal age.

Fish were between 1 and 9 years old and individual length was between 15 and 30,5 cm. There was no significant difference between the results of both methods (p=0,27). Bias for both methods was small for both readers, however it was slightly higher for the sectioned otoliths (-0,08) than for whole otoliths (-0,06) read by analyst 1. The results for coefficients of variation and percentage agreement demonstrated better results for the sectioned otoliths for both readers. A source of age determination error was the disagreement in the identification of the first annual ring as a false ring when reading whole otoliths. Based on these results, it was agreed to continue age readings of dab using the stained sections as best preparation method.

Keywords: age reading; dab; preparations methods; otoliths

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Carbon cycling in Antarctic benthic communities subject to glacier retreat

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The western Antarctic peninsula (WAP) is one of the fastest warming regions on Earth. At the northern tip of the WAP, on the southern coast of King George Island, lies Potter Cove, a fjord-like small embayment (about 3 km^2), influenced by the dynamics of the Fourcade Glacier. This glacier has been actively retreating since the 1950s and completely lost its water tongue in the past decade and now lays completely on land. The retreat released huge portions of the underlying soft sediments from the ice, exposing them to glacier calving disturbances (e.g. brash ice and ice scouring), the increased discharge of sediment-laden melt waters and to wave action. Several contrasting shallow benthic habitat types are present within the bay. Although bathymetric and granulometric characteristics are relatively similar within the cove, the sediment-inhabiting fauna community composition is very patchy and variable, ranging from colonist to medium-developed benthic assemblages as a result of the locally altered conditions.

Efficient carbon cycling is especially crucial in this very productive bay. The large annual primary producer biomass (mainly benthic microalgae and large macroalgae) needs to be recycled to the basic nutrients. Since benthic communities are - through their feeding and burrowing activities - strongly involved in the degradation of organic matter, it can be expected that the gradient in development of benthic communities in Potter Cove will be somehow reflected in the local patterns in carbon cycling.

In 2015-2016, we had the unique opportunity to measure carbon cycling *in situ* over a seasonal cycle (summer, winter *under ice* measurements, and spring). To this aim, skilled divers deployed a set of benthic chambers over the sediment and measured fluxes of oxygen, inorganic carbon and nutrients at the sediment-water interface. At the same time, the sediment was sampled to assess environmental variables and benthic assemblage structure. Preliminary results show that despite the low water temperatures (0-2°C), benthic carbon cycling rates were similar to those of temperate regions, which highlights the productivity of the area. Carbon cycling in winter was remarkably lower than in spring and summer, which probably relates to a lower activity and/or biomass of the benthos. The most recent ice-free site, also most frequently disturbed by glacier calving, was characterized by the least developed communities and lowest carbon cycling.

This seasonal set of carbon cycling measurements along a gradient of benthic assemblage statuses in Potter Cove will provide an example dataset for direct and indirect effects of glacier retreat on benthic ecosystem functioning, representing a unique study in the Western Antarctic Peninsula region.

Keywords: Antarctica; benthos; carbon cycling; glacier retreat

One year of monthly beach morphological changes in Mariakerke (Oostende) related to their forcing factors

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Sea level rise and an increase in extreme weather events due to climate change are expected to change the hydrological and meteorological conditions at the Belgian coast in the coming decades. At present, more than half of it is already in an erosive state. To protect the beach from further erosion in the future, a profound understanding of coastal processes is needed. For the Belgian coast there is a good understanding of storm thresholds for erosion impact and long-term morphological evolution. However, a relation between forcing and morphological response on a short- or medium-term is still lacking. Therefore, the aim of this study is to assess the morphological variability of the beach at Mariakerke over the year 2016 and to relate morphological changes to meteorological and marine forcing factors.

The study site is at Mariakerke near Oostende (Belgium). The beach is oriented SW-NE, with prevailing winds and waves from the WSW to NW. The wide, sandy (D_{50} = 170-250 μm) beach is flat and ultra-dissipative, typical for a macro-tidal environment. The beach is protected with groins, a dike and sand nourishments. At Mariakerke, beach topographic profiles have been measured at two different sections with an RTK-GPS (Real Time Kinematic GPS) every month from January to December 2016, except for July and October. From these profiles the morphological and volumetric changes of the intertidal and dry beach were analyzed. Wave height, water level and wind speed have been measured continuously near the research area.

The volumetric changes of the beach from the mean low water line to the dike are in the order of 0-18 m^3 per meter beach width. No clear seasonal cycle is observed, but it seems that accretion dominated in April, June/July and September, that the beach was stable in January, February and May and that erosion occurred in March and August. The beach (from the dike to the mean low water line) narrowed with up to 24 m in April and widened with up to 25 m in May. This is remarkable compared to the changes in beach width over the rest of the year, which were only up to 12 m. The beach narrowed in March, August and September/October and was stable in the other months.

At the beginning of 2016, the waves were small (< 2 m high), but during a storm in January peaks in wave height of 4 m, with a water level reaching 5.3 m TAW, were observed. The marine conditions were rather calm between March and April, followed by a more energetic period in May, when a storm occurred characterized by wave heights up to 3.9 m and a water level of 4.4 m TAW. After this storm, the sea state returned to calm.

Although the hydrodynamics show a clear seasonal cycle with more energetic wave conditions in winter, the beach morphology does not. Nevertheless, the storm event in May resulted in a lowering and widening of the intertidal beach for all the profiles. Interestingly, the intertidal beach was not affected by the storm event in January. Although the wave characteristics were similar between the two storm events, the lower water level in May resulted in a larger wave impact in the intertidal zone. Moreover, it seems that a narrowing of the beach occurred during the months with calm wave conditions.

Keywords: storm impact; beach recovery; hydrodynamics; wind

Abiotic modelling options of estuarine areas as building blocks for ecological predictions

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Most current ecological models focus on the improvement of underlying biological processes which are of course inherent to any model trying to simulate ecosystem components and processes. However, the importance of both the physical and chemical dimensions of an environment cannot be overlooked as ecological model performances are often limited by oversimplified, not representative or mismatching abiotic data of current velocity, DO, water temperature, conductivity, etc.

Estuaries, being characterized by their complex interplay of tidal movements, freshwater discharges, topography, geometry and atmospheric influences, show a high variability in their physical and chemical properties over both time and space. Researchers of estuarine phytoplankton dynamics, larvae dispersal and fish movement patterns are faced with a complex ecosystem and a wide range of data-driven and process-based methods to choose from. This choice is most often guided by limited data availability, previous modelling efforts and required temporal and spatial resolutions but often goes without a profound justification or recognition of the unknown uncertainty. This could therefore lead to the identification of key processes and variables based on artefacts, resulting from bad coupling between hydrodynamic, biogeochemical or ecological components of the model, rather than identifying real ecological trends.

In this study, we explored different modelling approaches in the Scheldt Estuary. Namely, within the framework of the European LifeWatch Observatory, a network of acoustic receivers was installed in the Scheldt Estuary to track fish with an acoustic tag. To analyse their behaviour, it is necessary to relate knowledge about the abiotic environment to the tracking data. Nevertheless, to deal with the major concern of the limited use of a wide spectrum of modelling approaches, the review is expanded to other estuaries and the simulation of their abiotic dimensions as a reference for the current set of modelling options in the Scheldt estuary. This setup allows us to provide a general overview of existing modelling approaches in estuaries and to characterize them in the light of different objectives.

Keywords: abiotic models; ecological models; estuaries; Scheldt estuary

Cable bacteria protect coastal waters from toxic nightmares

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Cable bacteria are long, multicellular, filamentous bacteria that form dense populations in marine and freshwater sediments (up to 1km of cable bacteria length per cm^3 sediment). These cable bacteria have a unique "electrogenic" metabolism, where electrons are first harvested from sulfide at centimeters deep in the sediment, then transported cell to cell along the longitudinal axis of the bacteria, and finally shuttled to oxygen near the sediment-water interface. Moreover, the activity of cable bacteria strongly impacts the pore-water pH, and this way, they substantially influence the cycling of various elements, such as iron, calcium and manganese.

Recently, a multi-year study of a coastal reservoir in the Netherlands demonstrated that cable bacteria activity not only influences the sediment geochemistry locally, but rather has a basin-wide impact. Every summer stratification of the water column in this reservoir leads to oxygen depletion in the bottom waters. During seasonal oxygen depletion, toxic sulfide produced as an end-product of anoxic metabolisms in the sediment could diffuse into the bottom waters and finally lead to depauperate toxic waters. However, Seitaj et al., (2015) proposed that cable bacteria activity in spring, before the onset of oxygen depletion, induces a chemical "firewall" in the upper millimeters of the sediment which effectively protects the overlying water from sulfide effluxes from the sediment in the oxygen depleted summer.

In this study, we have tested this so-called "firewall hypothesis" by seasonally sampling sediment cores, inducing anoxia in the overlying water, and assessing the sulfide release by the sediment. In these closed sediment incubations, we weekly traced major fluxes (e.g. sulfide and iron) until sulfidic conditions were reached in the water column. Cores which were collected during spring, with intensive cable bacteria activity, delayed sulfide release up to 80 days of anoxic incubation. These results combined with the proposed global distribution of cable bacteria indicate that the "firewall" might even be stronger and more prevalent than previously thought.

Keywords: cable bacteria; seasonal hypoxia; Lake Grevelingen; firewall hypothesis

Electrogenic sulfur oxidation drives trace metal cycling in sediments from the Belgian Coastal Zone

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Recently, a novel process of microbially mediated sulfide oxidation has been discovered [1]. In this process, named electrogenic sulfur oxidation (e-SOx), long filamentous cable bacteria couple the reduction of oxygen near the sediment-water interface to the oxidation of free sulfide and iron sulfides in deeper sediment layers via electrical currents that run over centimetre-wide distances [1,2]. This spatial decoupling of redox reactions generates a distinct pH profile with an alkaline peak just below the sediment water interface and an acidic pH minimum in the deeper sediment layers. These strong alkalinity gradients have been shown to significantly influence the elemental cycling of iron, manganese and carbonate minerals in the upper ~5 cm of the seafloor through the acidic dissolution of pH-sensitive minerals such as iron sulfides and carbonates [3,4]. Metal oxides and sulfide minerals are primary drivers for trace metals cycling [3,5] and thus, e-SOx will likely affect trace metal cycling. Yet, the impact of e-SOx on trace metal cycling is still largely unknown

Here we present results from *in situ* sampling (November 2014), and laboratory sediment incubations from the Belgian Coastal Zone (BCZ, Station 130). By combining the results of the microsensor profiling, pore water analysis and sequential solid phase extractions for trace metals and inorganic sulfur, it is shown that e-SOx is the primary driver of the biogeochemical cycles of cobalt and arsenic in the BCZ. Trace metal cycling is driven by the dissolution of sulfide minerals at depth and the reprecipitation of iron and manganese oxides near the sediment-water interface.

Keywords: electrogenic sulfur oxidation; cable bacteria; trace metal cycling

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Connectivity the Southern Ocean: population genomics of icefish

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The teleost fauna of the Southern Ocean is dominated both in terms of abundance and biomass by icefish of the suborder Notothenioidei. These fish are a rare example of a marine adaptive radiation with more than 120 extant species. Their diversification concurred with key adaptations to a cold, stable environment. Rapid environmental changes and increasing anthropogenic pressures, however, may affect their diversity and call for sound management and conservation measures. Characterizing genetic structuring in time and space is an important prerequisite to define targets for such measures. The oceanographic system of the Southern Ocean is dominated by the Antarctic Circumpolar Current that is connected to local gyres and fronts. While this current in conjunction with long pelagic larval phases may act as homogenizing force between populations, other factors such as habitat discontinuity or local adaptation can increase genetic differentiation. Current reduced genome representation approaches such as restriction-site associated DNA (RAD) sequencing enable characterization of markers on a genome wide level even in non-model species. We use these techniques to infer large and fine scale patterns of contemporary population structure in icefish and relate observed signals in genomic variability to potential driving forces of connectivity, such as ocean current systems.

Keywords: Antarctica; population structure; fisheries management; conservation genetics

Mercury stable isotopes discriminate different populations of common Seabass around Europe and provide insight on mercury cycle

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In a context where worldwide emission of mercury, a global pollutant, are increasing, research for new tools and data enabling a deeper understanding of mercury fluxes and sources are crucial. Over the past few years, the analysis of stable isotopes of mercury has emerged as a new promising technique affording to explore the Hg cycle, somewhat like what is being done for the carbon and nitrogen cycles. Hg can exhibit both mass-dependent (MDF, reported as $\delta^{202}Hg$) and mass-independent fractionation (MIF, reported as $\Delta^{199}Hg$). While MDF may occur during biological cycling *inter alia* and could be used to understand bioaccumulation processes, MIF provides a unique fingerprint of specific chemical pathways, such as photochemical transformations. In this context, information provided by Hg isotopes would help to improve environmental management strategies. However, so far, few studies considered Hg isotopes in marine vertebrates.

Our study reports the first data on Hg isotopic composition in marine European fish, for seven distinct populations of the common seabass, *Dicentrarchus labrax*. The combination of $\delta^{202}Hg$ and $\Delta^{199}Hg$ values enabled us to successfully discriminate several populations and recursive partitioning analyses demonstrated their relevance as discriminating tools. Moreover, mercury isotopic values provided insight on Hg contamination sources for biota and on MeHg cycling. We showed that $\delta^{202}Hg$ in seabass muscle is probably a good integration of the $\delta^{202}Hg$ of MeHg in their diet, except when concentrations are low, in which case in vivo processes would significantly influence the $\delta^{202}Hg$ in fish muscle. The $\delta^{202}Hg$ was also linked with known Hg point sources in several sites and the overall range of $\delta^{202}Hg$ around Europe was suggested to be related to global atmospheric contamination. $\Delta^{199}Hg$ in seabass was shown to reflect the level of contamination of fish and their habitat but not only. MIF was also clearly influenced by ecological characteristics of fish and their habitats, and therefore could be used to identify and investigate peculiar Hg environments such as in the Black Sea. Throughout this study, results from the Black Sea population stood out, underlying the particularities of Black Sea Hg which seemed to display a Hg cycling similar to what is observed in fresh water lakes.

Data on Hg sources and levels in Europe are scarce and Hg cycling is still poorly understood. Our findings constitute the first large scale isotopic analyses of Hg in the area. They bring out the possibility to use mercury isotopes in order to discriminate distinct populations, to explore the global Hg cycle on a large scale (Europe) and to identify particularities in the Hg cycle of several sites. The interest of using mercury isotopes to investigate the whole European Hg cycle is clearly highlighted by our results.

Keywords: mercury sources; stable isotopes; Europe; fish

Recombinant sea bass Transferrin inhibits and kills *Vibrio anguillarum*

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Transferrin, a glycoprotein with molecular weight of 80 kDa, is considered to be a natural anti-microbial agent in fish due to its iron deprivation activity leading to growth reduction of of bacterial pathogens. However, many bacterial pathogens, like *Vibiro anguillarum* have siderophore-dependent and/or siderophore-independent iron-acquisition systems allowing them to use iron for growth even in the presence of the iron binding, innate immunity related fish proteins like transferrin. In this study, recombinant European sea bass transferrin (Tf) was produced by transient transfection of COS-7 cells using pcDNA4::*transferrin.* Recombinant Tf was purified by column chromatography. Next, we showed that *Vibrio anguillarum*, which is the causative agent of vibriosis in aquaculture, was inhibited in its growth and was also killed by recombinant sea bass Tf. Recombinant Tf blocked the growth of both a virulent (V11) and a non-virulent (V14) *V. anguillarum* strain. Significant growth inhibition (OD measurements) of Vibrio (10^6 CFU) was observed from 4 hours onwards in the presence of 0.5mg Tf per ml culture medium. Taken together, these findings add extra knowledge on our understanding of the anti-microbial activity of transferrin, which apparently is not only caused by iron deprivation but also due to direct contact with the bacteria.

Keywords: sea bass; transferrin; vibrio anguillarum

A meta-analysis of isotopic compositions of North Sea marine mammals

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For over a decade, the North Sea has been undergoing significant changes due to global changes and overfishing. We conducted meta-analyses of previously published data on marine mammals sampled in the North Sea to test the competition for food sources and spatial variations. The overall objective of this study was to assess the potential trophic changes of the grey seal, the harbour seal and the harbour porpoise. Data included $\delta^{13}C$ and $\delta^{15}N$ values measured in blood cells and muscles from the three species. SIBER, a trophic niche overlap quantification approach, highlighted potential competition between marine mammal species. The ellipse drawn for harbour seal data showed the highest $\delta^{15}N$ values, reflecting its trophic position at the top of the food web. But the ellipse overlapping between the harbour seal and the grey seal of Germany was very important, showing a potential strong competition for food sources may be due to the overfishing. The harbour porpoise displayed a lower trophic position and a wide range of $\delta^{13}C$ and $\delta^{15}N$ values compared to harbour seal and grey seal as seen from its extended ellipse size. This may be due to a more opportunistic behaviour following the decline of some fish population in the North Sea. Surprisingly a group of grey seals sampled in Scotland present a very small ellipse size, presumably more selective in their prey choice, and show the lowest $\delta^{15}N$ values. Caution should be taken before comparing the trophic position of the groups of grey seals as the baseline differed between the two sampling areas. Low nitrates concentrations, higher latitudes, colder temperatures, deeper waters and rocky soils of the Scotland's coasts of the North Sea cause a stratification phenomenon of the water column explaining the lower $\delta^{15}N$ baseline in this area and so the spatial variation between these two groups of grey seals living in the North Sea.

Keywords: North Sea; marine mammals; stable isotopes

Extremely loud and incredibly close: *in situ* exposure of Atlantic cod to pile driving

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Underwater noise related to human activities is an increasing source of pollution in the marine environment. Although offshore wind farms (OWFs) do create green energy, they adversely affect the marine ecosystem by introducing different types of underwater noise. Especially during the construction phase, high impulsive sound is generated when the steel foundation piles are driven into the sea bottom. Impulsive underwater noise can be detrimental for marine life. Several laboratory experiments on fish and marine mammals showed behavioral changes, physiological stress, internal and external injuries, sometimes leading to mortality. However, a recent *in situ* study in the Belgian part of the North Sea (BNS) only showed short term physiological effects in larval and juvenile seabass (*Dicentrarchus labrax*) after exposure to high impulsive sound in the direct vicinity (<50 m) of a real pile driving event. Still, during that field experiment adult whiting (*Merlangus merlangus*) was seen floating at the surface at the moment of pile driving. Next to the need to further determine solid sound thresholds to be used in international guidelines, this anecdotic observation was the immediate reason for the current *in situ* experiment. In the summer of 2016, a field experiment was undertaken in the Nobelwind OWF on the BNS, to determine the direct effect of pile driving on the health status of Atlantic cod (*Gadus morhua*). Cod is classified as a vulnerable species on the IUCN list, it has a closed swim bladder (physoclistous fish) which makes it more vulnerable to swim bladder injuries, and it is known to aggregate around OWFs in the BNS. Large netted cages, each holding 10 cod individuals (avg. size 30 cm), were submerged at 8 m under the water surface. The cages were placed at increasing distances (75 m, 400 m, 1400 m and 1700 m) from the sound source, being the offshore installation vessel *Vole au vent*. All cages were submerged for on average 16 hours before pile driving, after which all fish were exposed to one pile driving event (lasting on average 2 hours). A similar control experiment was repeated in the same period when no pile driving took place. Underwater noise levels were measured at different distances during pile driving, while background measurements were made to determine ambient noise levels. Average single strike sound exposure levels (SEL_{ss}) decreased from 181 dB re 1µPa²s at 100 m distance to 168 dB re 1µPa²s at 1700 m distance. Ambient sound pressure levels (SPL) varied between 114 and 138 dB re 1µPa.

On average, 28 hours after exposure, the cages were retrieved onboard RV Simon Stevin, and all cod individuals were evaluated for buoyancy in water tanks. Shortly afterwards, all fish were euthanized and examined for swim bladder barotrauma and internal bleeding. Overall, 11 % cod were retrieved death, most probably due to handling stress, as no direct relation could be found with distance to the sound source. On the other hand, a steep increase in swim bladder barotrauma was detected with decreasing distance to the pile driving source: no swim bladders were ruptured at 1700 m nor at the control treatments, 20 % were ruptured at 1400 m distance, 40 % at 400 m distance and up to 90 % of the swim bladders were ruptured at 75 m distance. Although most fishes in the cages in the direct vicinity of the piling source (50-100 m distance) did survive this short term experiment, they all showed many internal bleedings and a high degree of abnormal swimming behavior, indicating they would probably not survive on the longer term. However, these immediate detrimental effects seem to be only a local effect, close to the high impulsive sound source, as swim bladder injuries rapidly decreased with increasing distance from the pile driving source. Based on these results, a sound threshold of 165 dB re 1µPa²s for SELss and 185 dB re 1µPa for SPLz-p may be suggested as precautionary approach to avoid internal injuries.

Keywords: cod (gadus morhua); in situ cage experiment; offshore wind farm; pile driving noise

Phaeocystis blooms in the Belgian coastal zone - Routine phytoplankton monitoring with the Cytosense flowcytometer

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Phytoplankton is a diverse group of photosynthesizing organisms which are accountable for roughly 50% of the primary production on earth. A subset of these phytoplankton species is capable of forming harmful algal blooms (HABs). During these HABs, phycotoxins can be produced, which can result in massive fish kills and pose a risk for human health. Furthermore, HABs can cause depletion of oxygen and block available sunlight for other organisms, through their rapid growth. Increasing our knowledge on phytoplankton dynamics, and specifically on factors involved in HABs, will provide the opportunity to initiate protective measures to ensure a healthy marine ecosystem. The prymnesiophyte Phaeocystis is such a bloom forming species, occurring in the Belgium coastal zone (BCZ). The blooms of Phaeocystis seem to be initiated by a change in nutrient load and the exceedance of an irradiance threshold. In 2016, the phytoplankton composition, including Phaeocystis, in the BCZ was monitored year-round, providing information on the location, timing and intensity of HABs.

Monitoring data was collected on board of the research vessel Simon Stevin, between February and December 2016, using the Cytosense flowcytometer (FCM), which is capable of semi-continuous automatic sampling. This device was used to determine species size and fluorescence characteristics of the phytoplankton present. This monitoring dataset was analyzed using specialized clustering software (Easyclus) and spatial packages in R, to determine the spatial and temporal phytoplankton composition. The 2016 monitoring data revealed an intense Phaeocystis bloom in spring, but also a small bloom at the end of summer. This innovative monitoring in the BCZ produces data of phytoplankton composition with a high spatial and temporal resolution, which is important for identifying potential high-risk areas and time periods for aquaculture and recreation. Furthermore, this research will improve our understanding of the environmental factors driving these HABs.

Keywords: phaeocystis; routine monitoring; cytosense flowcytometer; HABs; Belgium coastal zone

Diving response: an argument for the Aquatic Ape Theory in human evolution?

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Introduction: In contrast to the widely accepted evolution theory where apes left the trees to live in the African savanna, the Aquatic Ape Theory suggests human ancestors came to ground-level to live in aquatic environments close to riverbeds and in coastal regions[1]. Humans are believed to have developed adaptations to an aquatic lifestyle. One of those adaptations is the diving response (DR). DR is characterized by bradycardia (slow heart rate) and peripheral vasoconstriction (narrowing of blood vessels), resulting in a blood pressure increase and a blood flow redistribution to the hypoxia sensitive regions (i.e., O_2-conserving effect)[2]. Japanese fisher women, called Amas, are still utilizing this feature to dive for seafood. The aim of this study was to explore which physiological characteristics influence the heart rate (HR) drop during dynamic facial immersed apneas in apnea-trained and non-apnea-trained (controls) athletes.

Methods: This experimental study recruited 20 healthy athletes of which 10 apnea-trained (height: 1.64±0.08 m, body weight: 57.6±6.7 kg, body fat: 20.6±3.5 %, VO_2max:2457±354 ml/min) and 10 matched controls (height: 1.65±0.05 m, body weight: 59.3±5.6 kg, body fat: 18.5±4.5 %, VO_2max: 2396±318 ml/min). All subjects passed lung function tests (forced vital capacity (FVC), forced expiratory volume in 1s (FEV1), Tiffeneau-Pinelli index (FEV/FVC*100)), 5 maximal static apnea trials in air and a maximal incremental ramp cycling test. All subjects then executed a 40min cycling endurance test at 25% of their maximal power output. Every 4 minutes, a 30s apnea while cycling with facial immersion (FIA) in cold water (15°C) was performed. HR was monitored continuously. Absolute (bpm) and relative (%) HR drop represent the difference between the HR plateau during cycling and the lowest HR reached during apnea. Two-way Repeated Measures Anova, followed by Paired Samples T-tests were executed to analyze HR drop during dynamic apnea. One-way Manova was used to explore differences in apnea-trained athletes and controls. Pearson correlations were calculated to investigate possible relationships between physiological characteristics and HR drop. Significance was set at p<0.05.

Results: Maximal breath hold time (BHT) was significantly longer in apnea-trained athletes compared to controls (161±29s vs. 113±39s; p=0.005). No significant differences in lung function, maximal ventilation and breathing frequency were found between apnea-trained subjects and controls. During the 40min cycling test, no differences in HR drop appeared between both groups. The length of the sustained dynamic apnea was significantly longer in the apnea-trained group (30±0s vs. 24±5s; p=0.002). During every apnea, the absolute average HR of the entire group dropped (48±15 bpm) significantly compared to the HR plateau (p<0.001 for every apnea). Significant correlations between the Tiffeneau-Pinelli index and the relative HR drop during dynamic apnea (R=0.508, p=0.022) and between breathing frequency and the length of the sustained dynamic apnea (R=-0.632, p=0.004) were found in the entire group.

Conclusion: The decrease in HR during dynamic apnea with facial immersion, suggests that the parasympathetic stimulus overrules the sympathetic stimulus caused by cycling[2]. This phenomenon can be explained by the evolutionary O_2-conserving effect of DR. In contrast to previous studies, no differences in average HR drop were found between apnea-trained and control subjects[3]. Moreover, the apnea-trained group had a significantly longer maximal BHT and a longer sustained apnea during cycling. The lack of differences in lung function between the groups suggests the apnea trained group was not as specifically trained as in other studies. Correlations found between Tiffeneau-Pinelli index and HR drop and between breathing frequency and length of sustained dynamic apnea, suggest that lung function plays a role in DR.

Keywords: human evolution; aquatic ape theory; diving response; dynamic apnea; bradycardia

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Late Pleistocene palaeogeographical evolution of the southern North Sea

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During the late Middle to Late Pleistocene (ca. 160-11.7ka) the Belgian Continental Shelf (BCS) and the wider southern North Sea (SNS) experienced drastic and complex landscape transformations initiated by palaeoclimate changes related to the ice ages, causing ice sheet formation and large sea-level variations (up to 140 m). During periods of high sea-level, like today, the BCS and the SNS were completely submerged with the sea invading low lying coastal plains and river valleys. During low sea-level previously submerged plains were transformed into river landscapes with forests and open plains marked by a large biodiversity. One of these nearby river landscapes is the large Rhine-Meuse river system which has left a large imprint on the Dutch sector but also on the BCS. Until recently it was believed that all Pleistocene deposits on the BCS date from the Eemian and Holocene interglacials and are *in casu* marine deposits. New research however has shown that most of the preserved deposits actually are fluvial in origin and have, next to a Rhine-Meuse imprint, also a northern fluvial source from the central North Sea, with a possible input from North German Rivers like the Elbe and Weser.

Evidence for this is a.o. provided by gravel deposits found on the middle and outer BCS. Petrographic analysis shows that the larger part of these gravels originate from Scotland and the east coast of the UK (Dusar et al., 2016). Most likely ice sheet scouring of the rock basement has transported these gravels south and released them into either a lake or a river that transported calved off pieces of the ice sheet towards the Dover Strait. By unravelling the palaeogeographic evolution of the SNS, which is still poorly understood, we may get a good understanding of the context of future archaeological and palaeontological finds and how climate change impacts the landscape. One of the main outcomes of this study is to provide an archaeological potential map (APM) of the BCS. This map will help increase a proactive approach (e.g. archaeological impact reporting) as well as the cost-efficiency of activities at sea.

Keywords: Late Pleistocene; North Sea; palaeogeography

Apnea trained athletes: rather marine mammals than humans?

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Introduction: Diving mammals are unable to breathe underwater. In order to remain and survive underwater, they developed several physiological mechanisms, e.g. the diving response. The diving response is characterized by bradycardia (slow heart rate), peripheral vasoconstriction (narrowing of blood vessels), a rise in blood pressure and a redistribution of the blood flow to regions that are most sensitive to hypoxia, such as the brain and the heart (i.e., O_2-conserving effect). Humans show a similar response during apnea. This response, which is initiated by apnea and augmented by facial immersion in (cold) water, is more pronounced in apnea-trained athletes than in untrained humans[1,2]. We hypothesized that there would be a difference in average heart rate (HR) fall between dynamic apneas with facial immersion and apneas in air during exercise. In a second hypothesis, a larger HR fall was expected for the apnea-trained athletes compared to non-apnea-trained athletes (controls).

Methods: In this experimental study, the bradycardic response of 10 female apnea-trained athletes (height: 1.64 ± 0.08 m, body weight: 57.6 ± 6.7 kg, body fat: 20.6 ± 3.5 %, VO_2max:2457 ± 354 ml/min) was compared with the response of 10 matched controls (height: 1.65 ± 0.05 m, body weight: 59.3 ± 5.6 kg, body fat: 18.5 ± 4.5 %, VO_2max: 2396 ± 318 ml/min). All subjects performed two 40-minutes endurance tests on a cycle ergometer in a random order at 25% of their maximal power output. Every 4 minutes, they performed a 30 seconds apnea while cycling, with facial immersion (FIA) in cold water (15°C) or in air (AA) (18°C). This was repeated 7 times every test. During both conditions, the HR of the subjects was recorded continuously. Absolute (bpm) and relative (%) HR drop represent the difference between the HR plateau during cycling and the lowest HR reached during apnea. Statistical analysis was done using a repeated measures (M)ANOVA. Significance was set at $p < 0.05$.

Results: For the entire group, FIA as well as AA resulted in a significant fall in HR ($p < 0.001$). Both average absolute (FIA: 48 ± 11 bpm vs. AA: 36 ± 15 bpm; $p < 0.05$) and relative (FIA: 37 ± 7 % vs. AA: 28 ± 11 %; $p < 0.05$) fall in HR were significantly larger during FIA compared to AA. Apnea-trained athletes were able to sustain 92 % of all the apneas completely. Controls only completed 36 %. No significant differences were found in average absolute and relative HR drop during the apneas with and without water between the apnea-trained athletes (FIA: 50 ± 11 bpm; 39 ± 6 %; AA: 40 ± 16 bpm; 31 ± 12 %) and the controls (FIA: 45 ± 10 bpm; 34 ± 7 %; AA: 32 ± 14 bpm; 24 ± 10 %).

Conclusions: AA is sufficient to elicit a significant bradycardia as a part of the diving response. However, and in line with our first hypothesis, during FIA, where the facial cold receptors and thus the trigeminal nerve are stimulated, this response is more pronounced. In contrast to our second hypothesis, no differences in absolute and relative HR drop were found between apnea-trained athletes and controls. It is possible that breath-holding for 30 seconds was too short to bring out the full response in the apnea-trained individuals. Also, the specific environmental conditions during testing (water and air temperature) as compared to their natural training environment can have a different effect on the responses. Lastly, hydrostatic pressure, which was absent during facial immersion, might be necessary to elicit a complete response.

Keywords: mammals; humans; diving response; dynamic apnea; bradycardia

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Movement of chemical stressors changes the relationship between regional diversity and productivity

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Chemical stressors and nutrients in coastal and marine waters are not homogeneously distributed. As a result, environmental conditions vary among places, which generates differences in community composition. Such among-community variation can be expressed as beta-diversity. Beta-diversity decreases when the movement of individuals among patches, named biota dispersal, homogenizes community composition. Moreover, productivity can decrease when biota dispersal removes species from patches where they are best adapted to the local environmental conditions. However, not only biota but also environmental factors can move, which leads to a homogenization of the environmental conditions. Such homogenization potentially affects beta-diversity, productivity and the relationship between both. How the movement of environmental factors affects the relationship between beta-diversity and regional productivity has, however, hardly been studied. Therefore, this study examined this relationship across a stress dispersal gradient. To do so, we moved marine micro-algae and a toxicant independently (following a full-factorial design) between two patches, of which one patch initially contained the herbicide atrazine. The movement of the algae resulted in a gradient in beta-diversity, while the movement of the toxicant homogenized environmental conditions.

The relationships between beta-diversity and regional productivity were positive in the treatments with stress dispersal on day 8 and without stress dispersal on day 24. In contrast, these relationships were negative in the treatments with stress dispersal on day 24. The positive relationships between beta-diversity and regional productivity were caused by a biota transfer from the more productive non-stressed community to the stressed community. This transfer resulted in a productivity loss of the non-stressed community that could not be compensated by internal growth. Therefore, regional productivity decreased with decreasing beta-diversity. The negative relationships at high stress dispersal were caused by the inflow of biota to the stressed community, leading to an increase in productivity of the stressed community when stress levels decreased. The regional productivity thus increased with decreasing beta-diversity.

Our study shows that the relationship between beta-diversity and regional productivity changes when toxicant movement homogenizes the environmental conditions. Today, there is an increasing interest in how local environmental conditions regulate community composition and productivity. Based on this study, we argue that those studies should also incorporate the dispersal of biota as well as the movement of environmental factors.

Keywords: algae; beta-diversity; productivity; dispersal; toxicant; community

Development of a triad assessment method for brackish sediments in Flanders

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In sediment risk assessment, analysis of the pollutant concentrations is essential in determining the degree and nature of sediment contamination. However, chemical analyses provide no evidence of toxic effects or effects in situ. The Sediment Quality Triad method incorporates measures of various chemical parameters, toxicological effects and benthic community structure in view of conducting an integrated assessment of sediment quality.

In framework of developing a triad assessment method for the quality evaluation of brackish sediments, in 2015 and 2016 40 sediment samples were taken along the Scheldt estuary (Sea Scheldt (Flanders) and Western Scheldt (the Netherlands)) and other brackish aquatic systems in Flanders. For these samples, chemical parameters (e.g. metals and organic pollutants), toxicological effects and benthic community structure are being assessed. In this study 3 bioassays are carried out to test their suitability for uptake in a quality triad method for brackish sediments as indicator of ecotoxicological effects. Two sediment contact bioassays with the polychaete worm *Hediste diversicolor* and the amphipod *Corophium volutator*, and a pore water test with the rotifer *Brachionus plicatilis* are performed to test the toxicity of the samples. For the samples taken in 2015 observed mortalities varied between 0%-33% for *H. diversicolor*, between 1%-22% for *Corophium volutator* and between 10%-43% for *B. plicatilis*. Specific growth rate for *H. diversicolor* ranges from 3 to 4.3% day^{-1}. Control tests with reference sediment showed for the 3 tests mortalities lower than 10%, and for *H. diversicolor* an average growth rate of 5% day^{-1}. For the Scheldt estuary, the 3 tests showed significantly higher mortalities for sediments from the upper part of the estuary in the Sea Scheldt, which was characterized by higher levels of micropollutants, than for the lower part of the estuary (Western Scheldt).

Based on a literature inventory of existing sediment quality standards for brackish waters and on analysis of the ecotoxicological and biological effects of sediment pollutant concentrations, quality guidelines for chemical parameters for Flemish brackish sediments were derived.

For the biological component of the triad method, comprising an evaluation of the benthic invertebrate community, a separate evaluation method is developed for brackish oligohaline more static water systems, and for brackish sediments in the different ecotopes of the Scheldt estuary. For the latter the M-ABMI ('Multivariate AMBI', Bald et al., 2005; Muxika et al., 2007) and the Occurrence Intactness Index are tested for their suitability as biological index in a triad method for the evaluation of Scheldt sediments.

Keywords: sediment quality assessment; triad method; ecotoxicity; bioassays; benthic community assessment

The community structure of deep-sea macrofauna in an area of interest for deep-sea mining

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Deep-sea areas characterized by the presence of polymetallic nodules are getting increased attention due to their potential commercial and strategic interest for metals such as nickel, copper and cobalt. The polymetallic nodules occur in areas beyond the national jurisdiction, regulated by the International Seabed Authority (ISA). Anno 2016, the ISA has allocated license areas for the exploration of potential mining for polymetallic nodules to fifteen contractors. Under these exploration contracts, contractors have the obligation to determine the environmental baseline in the exploration areas. Despite a large number of scientific cruises to the central east Pacific Ocean, still very little published data on the macrofaunal biodiversity and community structure is available for the abyssal fields of the Clarion-Clipperton Fracture Zone (CCFZ). This study focused on the macrofaunal abundance, diversity and community structure in three comparable, mineable sites located in the license area of Global Sea Mineral Resources N.V. A rather homogeneous but diverse macrofaunal community associated with the sediment from polymetallic nodule areas was observed at a scale of 10s to 100s of km. However, slight differences in the abundance and diversity of Polychaeta between sites can be explained by a decline in the estimated flux of particulate organic carbon (POC) along a southeast-northwest gradient, as well as by small differences in sediment characteristics and nodule abundance. The observed homogeneity in the macrofaunal community is an important prerequisite for assigning areas for impact and preservation reference zones. However, a precautionary approach regarding mining activities is recommended, awaiting further research during the exploration phase on environmental factors structuring macrofaunal communities in the CCFZ. For instance, habitat heterogeneity, which was shown to structure macrofauna communities at larger spatial scales, has to be taken into account in future studies. Unless the rather limited number of samples taken in the current study (12 box cores), a large fraction (59-85%) of the macrofaunal genus/species diversity has been characterized in this study.

Keywords: polymetallic nodules; macrofauna; deep sea; CCFZ; biodiversity; community structure

Development of the first standardized biotic challenge model for Dover Sole (*Solea solea*) and its validation by assessing the protective potential of probiotic candidates

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Dover sole (*Solea solea*) is gastronomically highly appreciated and has a high market value. The species is therefore considered to be a very promising candidate for European aquaculture. Furthermore, ensuring a reliable supply of sole would reduce fishing pressure on wild Dover sole populations. However, as for many other fish species, sole production is hampered by amongst others high susceptibility to diseases and larval mortality, justifying the need for more research in this area. Infectious diseases are a major cause of larval mortality thereby decreasing survival and having a negative impact on the development of the fish embryo and larva.

Severe economic losses due to diseases in larviculture may be linked to vibriosis. Various *Vibrio* spp. have been cited as causative agents, with *Vibrio anguillarum* on top of the list. Efforts have been made to reduce infectious diseases, in the past mainly by applying antibiotics. Because of the emergence of acquired antimicrobial resistance, there is a great need for alternative measures to overcome diseases. In this respect, the amount of studies investigating the use and effect of probionts in aquaculture increased drastically. However, the mode of action of these probiotics is largely unknown. In addition, researchers become increasingly interested in studying bacteria-host interactions as understanding these may assist in combatting disease. Reliable experimental models not only enable experiments exploring the mode of action of probiotic candidates but also facilitate research aimed at unravelling the ways pathogens elicit disease and mortality. Nevertheless, only a handful of studies focused on the development of such models for fish larvae and for Dover sole, no such model is available yet.

Our research group developed a standardized biotic challenge model for Dover sole larvae adopting *Vibrio* spp. as potential pathogens. Five potentially pathogenic isolates were selected: the wild-type *V. anguillarum* HI610, *V. anguillarum* Fr and *V. harveyi* Fr that were both isolated from a disease outbreak in a French sea bass farm, *V. tapetis* isolated from skin blisters and liver of Dover sole and *V. tapetis* 2 originating from active skin ulcers of wild dab (*Limanda limanda*). Each isolate was added to the housing water of larvae at 10 days after hatching in a final concentration of 10^5, 10^6 or 10^7 colony forming units (CFU)/ml. No significant differences in survival were noticed between the larvae inoculated with *V. anguillarum* wild-type, *V. harveyi* Fr, *V. tapetis, V. tapetis* 2 and the control group at any of the concentrations tested. *Vibrio anguillarum* Fr supplied at a final concentration of 10^6 CFU/ml well water resulted in a larval mortality of 36%, while the non-challenged control group displayed 5% mortality. When this isolate was administrated to the larvae at a concentration of 10^7 CFU/ml, 48% mortality was observed.

In addition to proving valuable in many other applications, this model is to be regarded as a powerful tool to evaluate the impact of (a)biotic components on larval health. Different probiotic candidates (administrated via the water and *Artemia* nauplii) were already assessed for their protective potential against *V. anguillarum* challenge and several prebiotic components will be evaluated in the future. In a next step, the impact of algal toxins on larval health will be considered amongst others by evaluating the susceptibility to disease agents.

Keywords: *Solea Solea*; Dover sole; larvae; *vibrio anguillarum*; probiotics

The LifeWatch infrastructure

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LifeWatch was established as part of the European Strategy Forum on Research Infrastructure (ESFRI) and can be seen as a virtual laboratory for biodiversity research primarily, but also for climatological and environmental impact studies, to support the development of ecosystem services, and to provide information for policy makers in Europe. This large European research infrastructure consists of several biodiversity observatories, databases, web services and modelling tools, distributed across Europe, but virtually connected. The LifeWatch e-infrastructure integrates the existing systems, upgrades them were possible and develops new systems where needed. The LifeWatch Marine Virtual Research Environment (VRE) is a product of LifeWatch that was created based on discussions and developments during a number of LifeWatch meetings in 2014. The LifeWatch Marine VRE portal aims to aggregate some highly relevant marine resources, data systems, web services, online tools, etc. into one environment in the context of LifeWatch. The portal can be considered as a first bottom-up development demonstrating potential and capability emulating the LifeWatch objectives. The integration and interaction between the different components will increase in future versions of the portal. The portal will be updated in the framework of upcoming LifeWatch and H2020 VRE developments. The portal is organized into three interlinked sections: Access, Analyze and Develop. Belgium contributes to LifeWatch with varied and complementary in-kind contributions. These are implemented under the form of long lasting projects by different research centers and universities spread over the country and supported by each respective political authority.

The Flemish LifeWatch partners (Flanders Marine Institute (VLIZ)), and Research Institute for Nature and Forest (INBO)) are providing facilitated access to regional and global biodiversity data bases and data systems, through several data services, data publication, data archology activities and the construction of a local marine-freshwater-terrestrial observatory. Furthermore, a central Taxonomic Backbone will bring together taxonomic, biogeographic, trait and genomic data and disseminate this information through web services. The Walloon LifeWatch partners (Earth and Life Institute of the Université catholique de Louvain, and the Biosystems Engineering Department of the Université de Liège/Gembloux-ABT) set up a Belgian ecotopes database to integrate geographic datasets about European biotic and abiotic factors. Furthermore, ecosystem dynamic descriptors are being monitored. For this purpose a geoportal was launched, where information of ecosystem dynamics (vegetation cycle, snow indices, solar energy and fire indices) can be viewed and downloaded. The federal authority, in addition to the in cash annual LifeWatch contribution, supports the Royal Belgian Institute of Natural Sciences (RBINS) to develop an Antarctic Biodiversity Information System (AntaBIS) as a LifeWatch virtual laboratory. A second project under the guidance of RBINS and in collaboration with the Royal Museum for Central Africa (RMCA) is the development of a Barcoding facility for Organisms and tissues of Policy Concern (BOPCo). The federal authority also supports the Belgian Biodiversity Platform to set up and animate a LifeWatch scientific node.

During the VLIZ Marine Scientist Day 2017, the (Belgian) LifeWatch infrastructure will be presented through an informative poster series, and through interactive demo sessions:
- Studying plankton diversity in the Belgian Continental Zone: research opportunities (Mortelmans *et al.*)
- The acoustic receiver network: a sea of opportunities (Reubens *et al.*)
- GPS tracking network of Lesser Black-backed Gulls and Herring Gulls breeding at the southern North Sea coast (Stienen *et al.*)

Keywords: biodiversity research; e-infrastructure

Where goes the flow? – Tracing sole of the North Sea with genomics and otolith shape

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Fisheries managers should monitor fish stocks at an ecologically meaningful scale. Better understanding of population structure and movements of individuals between populations would be helpful. However it is difficult to measure connectivity between populations, especially because early-life stages cannot be tagged physically. Therefore we resort to information embedded in the genome and the otoliths. Population structure of adult sole *Solea solea* seems to be low but meaningful at the European scale. However this is less clear at the regional level of the North Sea. Our research focuses on Young of the Year sole caught off the Belgian coast and in the Wadden Sea in 2013 and 2014. Using Single Nucleotides (SNPs) molecular markers we trace the spawning origins of juveniles reaching the Belgian nursery. What is the contribution of the different spawning grounds to the Belgian nursery? Is there a reproductive strategy favouring different cohort origins throughout the season? We also compare the assignment of juveniles caught in the Belgian nursery with the ones caught in the Wadden Sea nursery. In order to assess connectivity, 200 juveniles have been genotyped using ddRAD sequencing and Fourier descriptors have been used to describe the otolith shape of the same individuals. First, we computed population genetic measures and observed variation in population structure between years. Then, the genomic data were compared with otolith shape. Our research reveals origins of fish contributing to the Belgian nursery stock, hence improving management perspectives.

Keywords: connectivity; traceability; early-life stages; genomic; otolith shape; flatfish; North Sea; Wadden Sea

EMBRC.be – The Belgian node of the European Marine Biological Resource Centre

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The European Marine Biological Resource Centre (EMBRC) is a pan-European research infrastructure providing a strategic delivery mechanism for excellent and large-scale marine science in Europe. EMBRC interconnects European marine research communities by upgrading and harmonizing their current infrastructures and services. It supports both fundamental and applied research based on marine bio-resources and marine ecosystems and as such encourages the development of blue biotechnologies. EMBRC provides the suitable research environment for a variety of users from both academia and industry.

Presently, EMBRC has nine European and associated countries as full members. EMBRC strives to establish a legal structure as a European Research Infrastructure Consortium (ERIC) by the end of 2016, which will also lead to a restructuring of the organization of the Belgian node of EMBRC (EMBRC.be) with potential new partners. EMBRC.be is at date represented by Ghent University, University of Hasselt, Royal Belgian Institute for Natural Sciences and the Flanders Marine Institute.

EMBRC builds on existing marine biological facilities, equipment and human capital. It aims for long-term collaboration, development of common strategies and standards, and sharing of best practices related to the use of marine biological resources. Services, tailored to users from academia, industry and governments, include access provision to European marine ecosystems and biodiversity, marine model organisms, culture collections, technology platforms, e-infrastructure services and training facilities.

For more information: www.embrc.eu

Keywords: blue bio-economy; EMBRC; research infrastructure; biological resources

Join the International Master of Science in Marine Biological Resources (IMBRSea) now!

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The IMBRSea programme is supported by the Marine Research Institutes belonging to the European Marine Biological Resource Centre (EMBRC).

Keywords: Master of Science; marine biological resources; EMBRC; Erasmus+; international

How to cope(pod) with a multistressor environment?

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The rapidly expanding human population and associated pressure on the climate and the environment are leading to multiple disturbances in marine ecosystems worldwide. Climatic change leads to physical stressors such as changes in sea water temperature, salinity, and pH, while dissolved nutrients and pollutants in the marine ecosystem are important chemical stressors resulting from human activities. There is still a lack of quantitative data and understanding on how these chemical stressors and stressors resulting from climate change interact in marine ecosystems. To guarantee a sustained biodiversity and ecosystem functioning in the future, the understanding of the relative importance of the main drivers of change within marine ecosystems is crucial.

Our study aims to get better insight in the relative contribution of various environmental drivers to changes at the base of the pelagic food web in the Belgian Part of the North Sea (BPNS). In the framework of LIFEWATCH monthly sampling campaigns were conducted at twelve stations within the BPNS and the Belgian harbors from February 2015 to February 2016. Zooplankton samples were collected and environmental variables such as sea water temperature, salinity, pH, chlorophyll a concentration, nutrient concentrations and a selected set of priority pollutants were measured at each site. The samples are being analyzed through a multimethodological approach including stereomicroscopic identifications combined with Zooscan analysis, fatty acid profiling and toxicological measurements.

Generalized additive modelling (GAM) in R was used to determine the main drivers of change in the abundance and distribution of the dominant copepod species Temora longicornis and Acartia clausi. T. longicornis and A. clausi show different dynamics in their spatial distributions and abundances and appear to be driven by different factors. By means of GAM we succeeded to explain a large proportion of the variability in both species. When optimized, these GAM will provide important tools to identify and quantify the relative contribution of multiple stressors on zooplankton species within the BPNS. The developed models will be validated in controlled lab experiments with selected zooplankton species, providing for possible model species to be used as bio-indicators in the future.

Keywords: multiple stressors; zooplankton; generalized additive modelling

Numerical modelling of wave interaction with coastal and offshore structures using a CFD solver

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Sustainable development of coastal and offshore structures – from conceptual layouts, optimisation, stability and environmental assessment to detailed design – requires a thorough understanding of coastal and offshore processes. Numerical modelling is a suitable tool to investigate these physical processes and to deliver valid, accurate, efficient and usable results and solutions. Parallel to the increase of the available computational power, the use of numerical models is growing rapidly, also in the design of all kinds of coastal and offshore structures, such as breakwaters, wave energy converters, etc.

In this work, the Computational Fluid Dynamics (CFD) software OpenFOAM (http://openfoam.org/) has been used to study wave interaction with coastal and offshore structures. The numerical simulations are performed in a numerical wave tank, which is the equivalent of a physical wave tank used for experimental model testing at a smaller scale. However, the idea behind both wave tanks is the same: reproducing the physics as observed in the ocean or the sea in a controllable environment to study particular physical processes in detail. The numerical wave tank is characterised by a length, width and height. Boundary conditions are needed at the sides of the tank, for example to generate and absorb waves. The CFD software solves a set of partial differential equations, the Navier-Stokes equations, representing the physics with a very high accuracy. Therefore, the numerical wave tank is split up in a large amount of small grid cells and the governing differential equations are discretised to algebraic equations using a finite volume method. In each grid cell, the algebraic equations are solved numerically using linear systems, taking the boundary conditions into account.

In this study, we present several simulations of coastal and offshore structures using a CFD solver:
- Wave propagation in a two-dimensional wave flume,
- Wave breaking on a sloping beach,
- Wave run-up on a monopile,
- Modelling of wave energy converter arrays.

The first author is Ph.D. fellow of the Research Foundation – Flanders (FWO), Belgium (Ph.D. fellowship 1133815N).

Keywords: waves; numerical modelling; CFD; OpenFOAM; numerical wave tank

With a pinch of salt: microplastics and trace elements in sea salts for human consumption

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Are commercial sea salts on the European market contaminated with microplastics or trace elements? And if so, what are the implications for food safety? To give an answer to these questions asked by the Belgian Federal Ministry for Public Health, 18 brands of commercial food graded sea salts were collected from Belgian supermarkets and speciality stores. As sea salts are harvested from seawater, they might contain contaminants present in the marine environment, but also the production and food processing methods may lead to increased concentrations of microplastics and trace elements. Therefore, sea salts from different origins and with varying harvesting, production and processing methods were selected. Microplastics were extracted from the sea salt through a hydrogen peroxide treatment to dissolve the organic matter, and were further identified and quantified after filtering by means of a stereo microscope. Trace elements (As, Cd, Hg, Pb and Br) were determined by Inductive Coupled Plasma Mass Spectrometer (ICP-MS).

The analyses showed a varying contamination of microplastics in the investigated food graded sea salts. The majority of the observed microplastics were categorized as synthetic fibres. The amount of microplastics seemed mainly to be influenced by the brine processing of the sea salts. Traditionally hand harvested sea salts, which are harvested by scraping the salt of the salt pan or marsh without brine washing after crystallisation, contained highest number of microplastics, with a maximum of 180 microplastics/kg salt. Taking into account the advised daily salt consumption, we estimated a maximum uptake of 330 microplastics per year or a daily intake (DI) of 1 microplastic particle through the consumption of sea salt. Microplastics are non-regulated emerging contaminants, with no food safety levels identified yet. Therefore, the potential human health risks posed by (this amount of) microplastics in food items cannot be assessed yet.

The concentrations of trace elements Hg and Cd could not be detected in any of the sea salts (i.e. less than the detection limit of 0.002 mg/kg salt for both elements). On the other hand, the elements As (detection limit: 0.004 mg/kg) and Pb (detection limit: 0.03 mg/kg) were found in most of the investigated sea salts. Concentrations of As were significantly lower than the maximum safety values for food graded salts as given in the Codex Alimentarius (0.5 mg/kg) and Belgian regulation (KB 17/09/1968, 1 mg/kg). Similarly, Pb concentrations were consistently below the maximum allowed level of 2 mg/kg as stated in the Codex Alimentarius of 1995, although 40 % of the samples exceeded the more strict Belgian limit of 0.1 mg Pb/kg. Concentrations of Br amounted to several hundreds of mg/kg in all sea salts, which is consistent with the fact that seawater naturally contains on average 65 mg Br per litre seawater.

As conclusion of this pilot study it can be stated that the current contamination of food graded sea salts with trace elements is not hazardous to public health. However, the higher Pb concentrations partly reflect the presence of this trace element in the marine environment, probably as an effect of human activities. Although the presence of trace elements and the DI of microplastics via sea salt are low, it is important to add these amounts to the overall intake via the complete food basket for further risk assessments.

Keywords: sea salt; microplastics; trace elements; heavy metals; food safety

Population structure and connectivity patterns of Giant Clams (*Tridacna* spp.) along the Western Indian Ocean

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Connectivity among populations reflects the influence of natural processes, such as community dynamics, community structure, and genetic diversity, but also plays a significant role for the resilience of populations to natural and anthropogenic impacts. Understanding the population structure of a species can give an insight to the connectivity of populations and it offers an opportunity to implement effective conservation and management priorities. *Tridacna* spp. are protected under Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) and many species are locally extinct in many regions due to overexploitation. Only a few studies have been conducted so far to understand their gene flow in the Western Indian Ocean (WIO) and hence critical information is still missing. This study aims to address this data inadequacy by assessing the genetic population structure of three *Tridacna* spp; *Tridacna maxima, Tridacna costata* and *Tridacna squamosa* and their differentiation in the WIO. 251 individual tissue samples were collected from Red Sea, Kenya, Tanzania, Mozambique and Madagascar. Genomic DNA was extracted with Chelex method and cytochrome oxidase I gene was used as the molecular marker. The obtained sequences will be compared with sequences in GenBank using BLASTN to verify the orthology and will be combined with the published sequences. Haplotype diversity, nucleotide diversity and statistical tests will be performed by using the program Arlequin to determine the genetic differentiation among populations. Based on the results, the possible patterns of ecological and evolutionary processes of the species will be discussed which is necessary for their protection.

Keywords: population structure; connectivity; gene flow; Western Indian Ocean

The relationship between oysters and microscopic benthic algae revealed by satellite remote sensing

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Primary production in estuarine and coastal mudflats is largely provided by assemblages of unicellular, benthic algae commonly referred to as microphytobenthos (MPB). Despite their microscopic size, these tiny algae form photosynthetic biofilms visible by satellite remote sensing (RS), thanks to their special optical characteristics. This makes RS a great tool to analyse MPB spatio-temporal variability, bringing to light the mechanisms behind it. Yet not all processes, e.g. interactions with higher-level benthic communities, are fully understood. In the context of the growing impact of invasive species, we analysed the influence of feral *Crassostrea gigas* oysters on MPB biofilms. A life-size experiment was conducted to evaluate the effect of oyster elimination on surrounding MPB, by burning the oysters from a reef within a mudflat. The impact of the experiment was assessed thanks to a unique 30-year time series (1985-2015) combining Landsat and SPOT satellite data, using the normalized difference vegetation index (NDVI). Seasonal signals were also extracted from this extensive time series. Our results clearly showed the promoting influence of oyster reefs on MPB spatial distribution. Thanks to the high resolution (30 m) of the historical time series, persistent, highly concentrated MPB patches surrounding oyster reefs were first highlighted. Secondly, the field experiment revealed oyster elimination's negative impact on both MPB biofilm structure and concentration. This confirmed that the relationship between MPB and oysters is not limited to a bottom-up control, but that oysters actually play a positive top-down control on MPB biofilms. The hypothesis of nutrient inputs has been advanced as an explanatory factor, where oyster organic matter released through excretion and biodeposition would stimulate MPB growth. MPB also showed marked seasonal variations associated to changes in patch shape, size and degree of aggregation around oyster reefs. These variations, with higher NDVI occurring during spring and fall, were consistent with those observed at broader scales in other European mudflats. This study provided the first evidence of oyster reefs' positive effect on MPB biofilms using a RS satellite multi-sensor time series.

Keywords: microphytobenthos; satellite remote sensing; oyster reefs; NDVI

Wave overtopping on steep low-crested structures: another climate change challenge

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Climate change is responsible for sea level rise and increase in the storminess (i.e. more frequent and more severe storms), posing risks to coastal communities. Floods on the coastline are expected to increase, leading to potential human life losses and significant economic damage. In this climate change scenario, the existing coastal structures protecting the Belgian coastline against wave attack should remain at least equally effective against future storms. Therefore, good knowledge of the coastal processes is required to correctly assess the safety of the existing coastal structures in this new scenario and to improve design guidelines. The main coastal processes involved are, among others, wave overtopping over the crest of the structures, wave run-up on sea dikes and wave forces and pressures exerted by the waves attacking the structures.

Wave overtopping is a key design parameter as it determines the necessary crest level of coastal structures (e.g. breakwaters) that limits the amount of water passing over the structure during wave attack. Traditional research has focused on analysing the average wave overtopping rate establishing its relation with diverse wave parameters of the sea state (wave height, wave period, wave steepness, etc.) and structural parameters (slope angle, crest freeboard, etc.). Furthermore, physical insights show that the damage on infrastructures and people near the coast during a storm is also related to individual volumes from single wave overtopping events.

Despite the large scientific literature available, there are still knowledge gaps to be covered in order to improve the understanding of wave overtopping under different conditions. This knowledge gap consists of overtopping data for steep low-crested structures (coastal structures with steep slopes and vertical walls, with a crest freeboard ranging from small to zero). This type of coastal structures with small freeboards are relevant in a climate change scenario where the sea level is increasing while the crest level of the existing structures is impossible or very expensive to modify. Improving the knowledge on various processes related to wave overtopping will eventually lead to more accurate overtopping prediction formulae with larger ranges of application, and hence to safer coastal defence structures under a climate change scenario.

The EurOtop (2007) manual is the reference manual in Europe about wave overtopping and overtopping assessment of coastal structures. It contains several overtopping prediction formulae for various types of coastal structures, lacking however prediction formulae valid for steep low-crested structures. An updated version, EurOtop (2016), is available with improved overtopping prediction formulae.

To extend the wave overtopping data available in the scientific literature, we performed 2D physical model tests at the large wave flume of Ghent University (Belgium). The experiments consisted in overtopping tests on smooth impermeable coastal structures, both with deep and shallow water conditions. The tests were focused on obtaining average and individual wave overtopping data for a range of slope angles α from steep to vertical walls and for a range of relative crest freeboards R_c/H_{m0} (where R_c is the crest freeboard of the structure and H_{m0} is the significant wave height) from large to zero. Four different datasets resulted from these physical model tests: dataset 'UG10', 'UG13', 'UG14' and 'UG15', all of them focusing on steep low-crested structures.

These new datasets obtained at Ghent University are useful to improve the accuracy of the existing prediction formulae on the range of steep low-crested structures. Recently, van der Meer & Bruce (2014) presented a new overtopping prediction formula fitted through the UG10 dataset and thus being in the range of application of steep low-crested structures. However, this formula underpredicts the results of the UG13, UG14 and UG15 datasets for very steep slopes and vertical walls, and for the zero freeboard limit case. By using the new Ghent University overtopping datasets, improvements on the accuracy of the prediction formulae can be made.

Keywords: coastal engineering; climate change; coastal structures; wave overtopping; steep low-crested structures; physical modelling

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Study of geochemical behaviour of pollutants in the Belgian coastal marine environment

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Belgian coastal area has been polluted for many years due to anthropogenic activities, leading to important changes in local marine ecosystems.

The project of NEWSTHEPS (Belspo project 2015-2019) aims to develop innovative approaches and novel tools and techniques that address the current fundamental scientific and methodological issues related to the implementation of Good Environmental Status (GES) of the Marine Strategy Framework Directive in national and European waters. In this framework, novel and integrated passive sampler techniques for metals (DGT technique) and for organic pollutants are developed for both chemical exposure and biological effect assessment. The novel and integrated approaches aim at (1) better measurement of contamination levels in the marine environment for metallic and organic pollutants, and (2) development of the DGT technique for organic pollutants, while it is now used for inorganic pollutants (BELSPO, 2014).

In total, three stations have been selected for a preliminary campaign in March 2016: OO3 in Oostende harbor, ZB1 in Zeebrugge harbor, and MOW1 about 5km from Zeebrugge coast in the North Sea. By combining the results of particulate metal analysis on suspended matter extracts, total dissolved metal analysis on filtered seawater extracts and labile metal fractions obtained on DGT resin extracts, it seems that OO3 (Oostende harbor station) is more contaminated by metals than ZB1 and MOW1. Particulate and total dissolved metal concentrations are higher at OO3 than at both other stations. Moreover, the labile and hence the bioavailable fraction of Mn, Co, Ni, Cu and Zn is also higher at station OO3. The preliminary results at ZB1 and MOW1 show more comparable and lower values, with the exception that the labile metal fractions of Pb, Cr, Fe and Cd measured at sea station (MOW1) are the highest of the 3 stations. These results, their time-evolution and final interpretation will have to be confirmed by a continuation of the study and the participation in new field campaigns in the coming years.

Preliminary results show that the suspended matter (SPM) amount at station MOW 1 is four times higher than at the two harbor stations, but SPM at ZB1 and OO3 contains higher levels of particulate organic carbon. Particulate nitrogen is only found at the three stations under organic form. Isotopic ratios for carbon show that the particulate carbon measured at OO3 and ZB1 should be from allochthonous sources, while particulate carbon measured at MOW 1 appears to be more from marine origin. Indeed, MOW1 presents less negative values for the isotopic ratio of particulate carbon compared to the isotopic ratios at ZB1 and OO3.

These results will further be used for the development and the validation of an integrated model to quantify the environmental status of the Belgian coastal zone. It will predict the accumulation, trophic transfer and effects of chemicals in this specific local ecosystem. Ultimately, our integrated approach will lead to the development of novel procedures for the monitoring of the contamination levels in the coastal areas.

Keywords: Belgian coastal area; total dissolved metal; labile fraction; DOC; DGT

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Inter Pulse Interval analysis of Sperm Whale (*Physeter macrocephalus*, Linnaeus 1758) "clicks" recorded in Mediterranean Sea between 1996 and 2011: Population structure and distribution of sizes

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The population of Mediterranean sperm whales (*Physeter macrocephalus*) appears to be partially segregated and a genetic divergence with Atlantic populations is evidenced by preliminary genetic studies. To date, in Mediterranean Sea, no animals have been estimated to be longer than 14 meters by using IPI (*Inter Pulse Interval*) analysis; this is in contrast with the length reported in literature for sperm whales in other oceans of the world; the size of adult males in Atlantic and Pacific Ocean is reported to reach 17-18 meters. A Stable *Inter Pulse Interval* can be identified among pulses that compose a single click, and thus the size of the spermaceti can be estimated. Then it is possible to determine the whole size of the animal by using some experimental equations. But the sample of IPI analyzed Mediterranean sperm whales is limited in the literature. With this research, trying to fill a literature gap, we aimed to analyze unexplored recordings made by C.I.B.R.A between 2000 and 2011 in the Mediterranean Sea, provided they had the required quality. Furthermore, we analyzed historical recordings already present in the literature. The acoustic estimate of sperm whale size provided information on the population structure and evidenced the presence of specific size classes. The distribution of size classes could provide useful information on the structure of the population and its trends. Analysis required the selection of suitable recordings, sperm whales sounds with good quality and eligible *Signal to Noise Ratio*, and was based both on manual examination with visual inspection of waveforms and of high resolution spectrograms, and on automatic methods developed in Matlab, mainly based on Cepstrum analysis. Available algorithms have been tested. In optimal cases, automatic analysis provided good data and required minimal time to perform IPI verification. In less than optimal recordings, only manual analysis provided reliable results.

Keywords: acoustics; behaviour; breeding; communication; conservation; distribution; ecology; habitat use; life history; photo-ID; physiology; population dynamics; social interactions; sperm whales; clicks; inter pulse interval; Mediterranean Sea

Detection and degradation of environmental DNA (eDNA) in the marine environment: a lab and field approach using plaice

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Rapid advances in molecular technologies allow the study of biodiversity based on environmental samples (e.g. water) and can significantly improve biodiversity and ecosystem studies. Yet, the development and validation of these methods in the marine environment is limited. In this study, we evaluated the use of environmental DNA (eDNA) for the detection of plaice (*Pleuronectes platessa*), a common marine flatfish, in the laboratory and the field. We conducted a series of 24h experiments to investigate the eDNA degradation and production rate and detection limit for plaice under controlled laboratory conditions and validated our approach in the field. We observed that plaice is almost instantaneously detected immediately after introduction in the experimental aquaria. During the first 8 hours after introduction, eDNA concentrations increased (k_{prod} = 2.794E-8) after which its concentrations reach a plateau or lag phase. After the removal of plaice, we observed an exponential decline in eDNA concentrations (k_{deg} = 0.124) reaching almost non-detectable concentrations after 24 hours. In addition, field samples of eDNA were collected in winter and summer in the Belgian part of the North Sea showed a clear seasonal pattern that corresponds with the known spatio-temporal distribution of this species. Indeed, in summer, when juvenile plaice is migrating from the Scheldt estuary towards the coastal areas, higher eDNA concentrations were detected close to the coast. In contrast, during autumn adult plaice migrate deeper North Sea waters, which is reflected in the lower eDNA concentrations measured near the coast. Our results show that the eDNA technique can be used to identify current presence of common flatfish in marine coastal waters, resulting in an instant snapshot of the environment. Environmental DNA therefore has the potential to significantly improve environmental studies as a high-throughput non-invasive method.

Keywords: environmental DNA (eDNA); degradation; detection; technical and biological detection limits; monitoring

Advanced modelling of wave overtopping for climate resilient coastal defence systems

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Only 67 km long, the Belgian coast represents a significant touristic, socio-economic and cultural value for the densely populated country. Historically, the coastal land-use planning was much less restrictive than that of the neighbouring countries. This caused a very high degree of local coastal development from the end of the 19th century on and increased considerably after the Second World War, with the rise of middle-class mass-tourism and apartment buildings on the sea dike. In addition, since the 1970's, beach nourishment was adopted as a complementary coastal protection scheme: millions of cubic meters of sand were artificially added to the beach and foreshore and pushed the sea further away from the sea dikes. This resulted in the typical Belgian coastal appearance as it is known today: an almost continuous row of high rise apartment buildings fronted by a sea dike with promenade and a mildly sloped beach.

The typical Belgian coastal defence system, in the urbanised coastal areas, is a nourished beach providing a long and (very) shallow foreshore in front of a sloped sea dike with promenade and buildings and/or storm wall(s) close to the dike crest. Climate change is expected to keep causing sea level rise and an increase in storm occurrence and intensity. The Belgian coastal defence system against flooding is therefore being adapted, according to the masterplan for coastal safety [1], by a combination of a beach nourishment and dike crest level increase by a storm wall.

In the functional design of these storm walls, the height of the storm wall is calculated by determining the wave overtopping, which is limited to a specific safety criterion. The wave impact forces need to be resolved for the design of the structural stability of the storm wall. Current state of the art methodologies to calculate the wave overtopping still contain simplifications that are too conservative for this kind of cross-section. For example, they do not take into account some important physical processes resulting from the complex geometry of the typical Belgian coastal profile. In addition, the design criterion for wave overtopping has focussed on the mean overtopping discharge. Individual wave overtopping volumes have not been investigated into much detail yet.

The present research focusses on advanced numerical modelling of wave overtopping over and wave forces on this typical Belgian coastal defence system, resolving the hydrodynamic flow in full 3D (or 2DV, disregarding the alongshore horizontal dimension). This should allow for a much more accurate prediction of the mean overtopping discharge and individual overtopping volumes. Furthermore, hydrodynamic experiments in both a wave flume (2D) and wave basin (3D) in addition to field measurements will be performed to provide data with which the numerical method can be verified. Field tests do not suffer from scale effects nor from model effects and are a crucial source of information to be integrated in the overall validation of the methodology. The field measurements will include an artificial dike constructed on the beach, close to the high water line, to allow measurement of wave overtopping and wave impact on the short term.

Keywords: wave overtopping; wave impact; storm wall; very shallow foreshore; numerical wave modelling; hydrodynamic experiments; field measurements

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Introducing data and model uncertainty into the new 3D voxel model of the Belgian part of the North Sea

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One of the major challenges in creating geological models is to be able to prove how 'realistic' these models are. Since the 3D voxel model developed within TILES (Belspo project on 'Transnational and Integrated Long-term Marine Exploitation Strategies') will form the basis of a decision support system that will 'guide' the sand extraction in the Belgian part of the North Sea for the coming years, it is crucial to integrate estimates of uncertainties into the model. Two major categories of uncertainties are being quantified: database related uncertainties (e.g., core density) and interpolation-related uncertainties. The latter are quantified by calculating entropy (i.e. entropy of a voxel is a single value ranging from 0 to 1 that can be calculated from each of the probabilities of lithological class. An entropy value of 0 means that there is no uncertainty, whereas a value of 1 indicates that all lithological classes have the same probability thus higher uncertainty.) on the lithoclass data (i.e., fine, medium to coarse sand), as well as on the stratigraphical unit the lithoclass belongs to (i.e., the different Quaternary layers). The uncertainty layers can be queried and visualized independently or in comparison with the actual model. In these visualizations areas that have more certainty can be identified, but also areas that need further investigation. This combined information is essential for the effective use of the voxel model by stakeholders.

Keywords: uncertainty; entropy; 3D voxel model; North Sea; sand extraction

DNA barcoding of small mesopelagic fish from the Southern Ocean

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Small mesopelagic fish are an important trophic link in the world's oceans as consumers of zooplankton and prey for large fish, marine mammals, and birds. Myctophidae (lanternfishes) predominate in terms of abundance and diversity, and comprise up to 65 % of the entire pelagic deep-sea fish biomass. Their photophore patterns are particularly useful to distinguish species. However, early life stages with less pronounced patterns and individuals damaged during the hauling process often lead to misidentification. DNA barcoding can resolve specimen identification issues and furthermore pinpoint cryptic species, synonymies, or intraspecific phylogeographic patterns. Here, we present a large-scale cytochrome *c* oxidase I gene (COI) data set of 299 previously unpublished Antarctic mesopelagic fishes, with circum-Antarctic sampling range. This data is combined with Rhodopsin (Rho) sequences and publicly available DNA barcodes yielding > 1000 sequences, the largest dataset of Myctophidae sequences as yet. Morphological identifications and sequence clusters were compared. Phylogenetic trees were created using a Maximum Likelihood approach and compared to recent myctophid phylogenies. Intraspecific genetic distances between geographical areas were evaluated, thus creating a comprehensive picture on Myctophids occurring throughout the Southern Ocean. DNA Barcoding proved useful for Antarctic myctophid specimen identification in most cases. We found few hints of cryptic speciation and shallow intraspecific divergences. In general, the spatial phylogeographic structure is low within the Southern Ocean, possibly due to the high abundance of Myctophids and few barriers to gene flow in the well-connected mesopelagic realm. The phylogeny shows that adaptations to (sub-)Antarctic conditions must have occurred several times, yet only few species manage to thrive in cold waters permanently. Understanding the evolution of this ecologically important group in the Southern Ocean is highly topical given rapid environmental changes and potential exploitation.

Keywords: DNA barcoding; COI; Southern Ocean; Myctophidae

Tracking a marine predator to design flexible boundaries of marine protected areas for an ecosystem approach to fisheries

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African penguin *Spheniscus demersus* populations showed a drastic decline during the past decade, and their abundance is substantially linked with the presence of their prey. However, environmental changes and resource competition with purse-seine fisheries disrupt this prey availability. Therefore, conservation efforts should ensure adequate local food availability. In this perspective, marine protected areas (MPAs) have been identified as a valuable tool in an ecosystem approach to fisheries (EAF). Furthermore, when designed with flexible boundaries, MPAs can be beneficial for both penguins and fisheries. In Algoa Bay, South Africa, an EAF has been attempted through experimental purse-seine fisheries closures from 2009 to present. We investigated the effectiveness of these closures and evaluated the conservation value of a newly proposed, zoned MPA. Using GPS loggers, we studied the foraging behaviour of African penguins to assess their spatial overlap with purse-seine fisheries catches. Furthermore, the GPS tags provided a suite of horizontal and vertical foraging measurements, which were modelled with several explanatory variables. Our results showed a decline in purse-seine fisheries catches since 2012, possibly due to limited fish abundance. Nevertheless, the experimental closures showed a marked reduction in the penguins' foraging effort. The presence of prey is probably the main cue to adapt their foraging strategy, and this is likely to be dependent on environmental conditions and fishing pressure. Also, geospatial mapping revealed a strong overlap between the foraging range of African penguin and purse-seine fisheries in Algoa Bay from 2008 until 2015. Conversely, the no-take zones of the proposed MPA cover less than half of that foraging range. In general, this study indicated that the proposed MPA would provide a legal improvement to the current situation, but is not sufficient to increase numbers of African penguin populations. Larger no-take zones around the penguins' colonies are necessary when prey biomass is low. As an immediate action, ongoing acoustic surveys could provide information on prey availability to design flexible boundaries and increase the benefits for all stakeholders involved. In the future, more quantitative research is necessary to reconcile conservation goals and the socio-economic needs of the coastal community of Algoa Bay.

Keywords: *Spheniscus demersus;* GPS tracking; adaptive fisheries management; no-take zones; foraging behaviour

Connectivity of the skunk clown fish in the Indian Ocean using a combination of microsatellite and mitochondrial genetic markers

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Knowledge of connectivity, the exchange of individuals among populations on coral reefs, is vital for a correct spacing of Marine Protected Areas. For almost all coral reef associated fishes, dispersal between reefs in this patchy environment is limited to a weeks- to months-long pelagic larval stage. This study uses the Skunk Clown Fish (*Amphiprion akallopisos*) as a model species to assess connectivity. It has a disjunct distribution, occurring in the Western Indian Ocean (WIO) and the Eastern Indian Ocean (EIO), separated by more than 4.500 km of open sea. We use a combination of mitochondrial (MtDNA, Control Region) and 15 highly polymorphic microsatellite markers. Fin tissue of 387 individuals from 21 different sites in the WIO (Kenya, Tanzania, Madagascar, and Mozambique) and the EIO (Indonesia) was analysed. Mitochondrial and nuclear markers concurred in detecting strong population structure between the EIO and the WIO populations (AMOVA: φct = 0.63; Fct= 0.10 respectively, P<0.001 both), and panmixia within the EIO. Within the WIO, however, results were different depending on the marker. With MtDNA, a genetic break was detected between the East-Madagascar population of St-Marie and the rest. Microsatellite markers also identified genetic differentiation between Madagascar and the East African coast, but the break was located more to the West of Madagascar. Microsatellites also revealed differentiation between Northern Kenyan populations and the rest, not detected by MtDNA. Both genetic breaks can be explained by dominant sea surface currents that promote larval detention in the Mozambique Channel, creating a barrier to dispersal across the channel, and the influence of the Somali Current which may hamper dispersal towards North-Kenya from populations further south.

Keywords: coral reefs; connectivity; gene flow; clown fish; Indian Ocean

Tri-dimensional modeling of Southern Bight of the North Sea: First results and perspectives

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In the frame of the Belgian research project FaCE-It (Functional biodiversity in a Changing sedimentary Environment: Implications for biogeochemistry and food webs in a managerial setting), the impact of dredging activities and offshore wind farm installation on the spatial distribution of sediment grain size, biodiversity and biogeochemistry will be estimated in the Southern Bight of the North Sea (SBNS) with a focus on the Belgian Coastal Zone (BCZ).

To reach this goal, the tri-dimensional hydrodynamical model ROMS-COAWST is implemented in the SBNS in order to simulate the complex hydrodynamics and sediment transport. Two levels of nesting will be used to reach a resolution of 250 m in the BCZ. The model is forced at the air-sea interface by the 6-hourly ECMWF ERA-interim atmospheric dataset and at the open boundaries by the coarse resolution model results available from CMEMS (Copernicus Marine Environment Monitoring Service), and also considers tides and 4 main rivers (Scheldt, Rhine with Maas, Thames and Seine).

Two types of simulations have been performed: a 10-years climatological simulation and a simulation over 2004-2013 to investigate the interannual dynamics. The model skills are evaluated by comparing its outputs to historical data (e.g. salinity, temperature and currents) from remote sensing and in-situ.

The sediment transport module will then be implemented and its outputs compared to historical and newly collected (in the frame of FaCE-iT) observations on grain size distribution as well as with satellite Suspended Particulate Matter (SPM) images. This will allow assessing the impact of substrate modification due to offshore human activities at local and regional scales.

Keywords: hydrodynamical modeling; ROMS; Southern Bight of North Sea; Belgian Coastal Zone

Sustainability of a tropical shrimp fishery: can genetic research give additional clues?

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Shrimps are one of the most important fishery resources in economic value and constitute around 18% of the world's traded fishery exports. As such, tropical shrimp fisheries provide an important source of food and income for many people, particularly in developing countries. However, due to the wide-spread overexploitation, the destructive way of fishing (bottom-trawling), and the high quantities of bycatch, these fisheries are generally regarded as unsustainable. In order to preserve stocks and the ecosystems they reside in, shrimps have to be exploited in a sustainable way. This insight has led the fishery for the Atlantic seabob shrimp *Xiphopenaeus kroyeri* in Suriname (South America) to take measures to improve the fisheries' sustainability. Thanks to these efforts, the industrial seabob fishery in Suriname has been granted with the Marine Stewardship Council (MSC) label for sustainable fisheries. Nevertheless, to come to an ecosystem-based fisheries management for the seabob fishery, important knowledge on the phylogenetics and population genetic structure of the seabob shrimp in Suriname is lacking.

For instance, genetic studies on the species in Brazil indicated a more complex phylogeny than previously thought, including the presence of cryptic species. In the present study, integrative taxonomy is applied to infer the phylogeny of the seabob shrimp. Therefore, a combination of genetic markers (mitochondrial and nuclear genes) and phenotypic characteristics is applied.

Additionally, the Surinamese are not the only ones targeting the seabob shrimp. Fishermen in the neighbouring countries, Guyana and French Guiana, are also targeting this valuable marine resource. To what extent Suriname shares its seabob stock with its neighbours is unknown. Therefore the population genetic structure and gene flow of the seabob population(s) are analysed by using highly polymorphic microsatellite markers.

Results of this research can guide management decisions to come to an ecosystem approach to fisheries, and will ultimately lead to a more sustainable use of marine resources in Suriname.

Keywords: sustainable fisheries; phylogenetics; population genetics; tropical shrimps

Versatility of marine geological databases in view of MSFD related assessments

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To ensure harmonised seabed mapping over large areas and to facilitate the exchange of information, a common geological knowledge base is being developed for the southern part of the North Sea. Data, sustaining this marine geological database, originate from a variety of sources, including the industry.

Related to the seabed and its subsurface, two main databases are being compiled: one comprising all available lithological descriptions and one with all numerical grain-size information. To enable standardisation of the data and make them easily queryable, non-numerical descriptions are being coded to an international standard (EU FP7 Geo-Seas), of which the Udden-Wentworth scale is the main classifier. Several other parameters were derived, such as percentages mud, sand, gravel, shells and organic material. For the second database, cumulative grain-size distribution curves were compiled, enabling calculations of any desired granulometry parameter, such as percentages of the grain-size fractions (fine, medium, coarse sand) and percentiles that are relevant in seabed-habitat mapping or sediment-transport modelling (D35, D50, D84). To quantify data uncertainty in the derived data products, metadata on sampling and coring techniques, analytical methods, horizontal and vertical positioning accuracy and the exact timing of data acquisition were included. These metadata fields are also crucial in the quantification of habitat changes, a key issue in the assessment of good environmental status within Europe's Marine Strategy Framework Directive.

The newly developed database and its associated data products contribute to the objectives of the projects TILES (Belspo Brain-be), EMODnet-Geology (EU DG MARE), and ZAGRI (private revenues from the marine-aggregate industry).

Keywords: geological resources; data management; seabed mapping; Marine Strategy Framework Directive; North Sea

Poly-extreme adaptation of early life in deep ocean: Minimum amino acid requirement for hyperthermophilic archaea, *Thermococcus eurythermalis, u*nder pH boundaries

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Thermococcus eurythermalis, a hyperthermophilic archaea isolated deep-sea hydrothermal vents at Guaymas basin, has exhibited tolerance to a broad range of different environmental stresses such as pH, temperature, salinity, and pressure. The poly-extreme adaptation of the species can potentially explain its dominance at its initial habitats, which is characterized by ephemeral and drastic gradient change of various chemo-physical factors. While the specific molecular mechanisms of the extreme adaptation remained unclear after intensive studies, physiological studies can provide crucial insight of the regulation of metabolic pathways under different extreme conditions. Here, we firstly examined the minimum amino acid requirement of the archaea under neutral and pH boundary conditions. For the experiment, cells were incubated in a defined medium, TRM basal plus each combination of 19 (out of 20) amino acids, with pH 4.5, pH 7.0, and pH 8.5, respectively. The amino acid was considered essential when the lacking of it could significantly affect the cell growth, and the essentiality was confirmed by incubation that only had selected amino acids added. Amino acids were used as the only carbon and nitrogen source in the medium. The result showed that there are 8 essential amino acids (trp, tyr, phe, his, thr, asp, arg, met) for pH 7, and the 12 essential amino acids (trp, tyr, phe, his, thr, asp, arg, lys, val, ile, leu) for both pH 4.5 and pH 8.5 include all the 8 amino acids for optimal condition plus four extra ones, three branch-chained (val, ile, leu) and one positively charged (lys). All the aromatic amino acids are essential in all the conditions, which might be explained by the easy availability of aromatic substances at the oil-immersed habitats. Comparison between these minimum amino acid requirements with Miller's amino acids provided important insight for the Heterotrophic Hypothesis regarding the origin of life.

Keywords: extremophiles; deep-ocean; hydrothermal vents; pH; adaptation

Unravelling the contribution of halophilic bacteria to the *Artemia* diet

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The stagnation of capture fisheries, combined with the increasing demand of seafood by the world's growing population, urges for the sustained high growth of aquaculture (average annual growth rate of 8.8 % over the last three decades) as a keystone in global food security.

The production of sufficient live food for the larviculture stage is a bottleneck for aquaculture expansion and diversification. As a substitute for the natural food, the larvae ("nauplii") of the brine shrimp *Artemia* are used as a universal live food source thanks to its convenience, flexibility in use, and nutritional value. Natural populations of *Artemia* are found worldwide in hypersaline ecosystems (e.g. inland salt lakes and coastal salt pans). Annually about 3000 tons of dormant eggs ("cysts") are commercialized and hatched into nauplii in larviculture units ("hatcheries") as larval food for most farmed fish/shellfish species.

In addition to cysts from natural salt lakes and to meet the increasing demand for cysts, *Artemia* culture integrated in solar salt production, has been introduced successfully in several (sub)tropical countries where dense brine shrimp populations are maintained through a labour intensive and economically costly stimulation of microalgae blooms and supplementation with inert feeds. Lowering these production costs would therefore contributes to the sustainability of the technique, and hence to the security of the *Artemia* supply.

Recent field studies are trying to optimize such salt pond based *Artemia* production by stimulating the naturally occurring halophilic bacterial flora as additional food source for the *Artemia* nauplii. However, in these xenic and open culture systems there is no way to assess the nutritional contribution of bacterial biomass among a variety of available feeds. The results obtained from these field studies do not allow thus patent conclusions about the contribution of halophilic bacteria to the brine shrimp diet, hindering the widespread application of such techniques.

Hence, in the present research we aimed to use gnotobiotic (animals cultured in axenic conditions or with a known microflora) *Artemia* culture systems to investigate for the first time *Artemia* nauplii's ability to survive and grow on diets consisting of pure halophilic bacteria biomass. We successfully demonstrated that several halophilic bacteria strains have positive effects on survival, body length and swimming speed of *Artemia* nauplii at both marine and hypersaline salinity.

This work is therefore the first step to investigate the relative importance of different halophilic bacterial genera and species for the *Artemia* life cycle and for the surrounding hypersaline food web, shedding light into the promising potential of these microorganisms to maximize *Artemia* production in salt ponds.

Keywords: aquaculture; live feed; hypersaline environments; food web; *Artemia*; halophilic bacteria; gnotobiotic culture systems

Evaluation of the sand extraction impact on the seabed and the water column at short time and space scale in an intensively dredged area of the Belgian part of the North Sea by using multibeam echosounder bathymetric, backscatter strength and water column data

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For several decades the Belgian Continental Shelf (BCS) has been exploited for its natural resources like gravel and sand. As this exploitation in the legal extraction zones intensified over the years, according with the environmental regulation both on national and European levels, evaluating the extraction impact there is an increased importance to monitor these changes in higher resolution and research the possible influences on on the seabed bathymetry and morphology of the seabedas well as on the sediment nature itself is a legal obligation..

Monitoring of sand extraction on the BCS is done by the Continental Shelf Service (CSS) of the Belgian government. For this purpose multiple campaigns a year on board of either RV Belgica or RV Simon Stevin acquire data at the current sand extraction hot spots.By providing simultaneously at high spatial resolution derived bathymetric and backscatter strength (BS) models of the seabed with water column content imagery from raw acoustic data, modern multibeam echosounder (MBES) is certainly the most efficient technology that can be used to provide a full quantitative picture of the impact of extraction on the marine environment.

Since more than 15 years, the Continental Shelf Service from the FPS Economy has collected MBES bathymetric and BS time series on several monitoring areas inside the extraction sectors. This unique dataset demonstrates the direct and non-cumulative impact of the extraction on the bathymetry of the sandbanks and the unsustainable character of the sand resource. However, several questions remain regarding the precise way the extraction impacts directly the seabed morphology and sedimentology. Moreover, dredging by suction (the only technique allowed in Belgium) generates fine sediment plumes which could by transport followed by sedimentation, notably modifying the habitats within a certain radius around the extraction sites. If the sediment plumes generated during the dredging operation have been the subject of numerous publications, few projects have been attempted to visualize these plumes and quantify in real time the volume of fine sediment implies by using MBES water column amplitude data.

These questions are addressed in a master thesis for which specific series of acoustic measurements using the Kongsberg EM2040 MBES installed on the RV Simon Stevin were carried out following operating dredging vessels on the Thorntonbank. In September 2016, three surveys were conducted following three dredging vessels under excellent sea conditions. The resulting high quality dataset allows to evaluate the real time impact of the extraction on the seabed and the water column. By combining this dataset with bathymetric and BS data derived from surveys done with the same acoustic equipment in March, April, November and December 2016, it is possible to clarify with a high spatial resolution the extraction impact on the seabed and on the sediment pattern at different time scale and as well as to evaluate the fading and evolution of the dredging furrows on the seabed after extraction.

Another goal of this research is to evaluate the feasibility to use the MBES water column amplitude data to visualize, characterize and quantify the sediment plumes generated by the dredging operation. 4D visualization of the MBES water column data from the dataset recorded following operating dredging vessels reveals the distinction between the lower plume caused by the dragging of the suction head over the seabed and the upper plume due to the dumping of the water that has been sucked up during the dredging operation.

As this study is still ongoing, different "key correlations" will be analyzed in the near future. It is crucial to assess to what extent the average amplitude level measured in the water column is correlated with the time varying distance and angle between the acoustic system installed on the research vessel and

the dredging head of the dredging vessel. These data will also be used to evaluate the extent to which the density of suspended sediment in the water column as reflected by the water column amplitude can significantly influence the seabed BS as measured by the MBES at the bottom detection window.

Survival in a feast-famine environment: Resource utilization, storage and recycling in cold-water coral reefs

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Cold-water coral reefs play a major role in carbon cycling in the deep-sea, but their high productivity and diversity stands in sharp contrast to infrequently available resources. Organic matter is exported from surface ocean to deep sea in short peaks creating feast feeding conditions intermitted by long famine periods. This natural environmental gradient might be enforced due to global change, likely resulting in lower export production. Further, climate change and ocean acidification potentially increase corals' energetic demands.

In different stable isotope tracer experiments we have investigated the resource cycling within and between key CWC reef organisms: the most abundant framework-forming cold-water coral *Lophelia pertusa*, reef-associated sponges *Geodia baretti* and *Mycale lingua* and bivalve *Acesta excavata*. On the one hand, we have focused on their potential to utilize different resources such as algal detritus, bacteria and dissolved organic matter (DOM), and contrasted their storage capacities (e.g. in fatty acids) with the metabolization of assimilated resources. On the other hand, we have quantified the amount of dissolved and particulate organic matter that those organisms release, which in turn can serve as substrates in recycling pathways of the reef community.

The interconnection between those key reef species, their potential to assimilate DOM and bacteria, i.e. alternative food sources in times of low organic matter export, and their recycling capacity will be discussed as key factors to study resilience towards global climate change.

Keywords: stable isotope tracer experiments; dissolved organic matter (DOM); bacteria; carbon storage; fatty acids

EMODnet: the gateway to marine data in Europe

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Marine data are needed for many purposes: for acquiring a better scientific understanding of the marine environment, but also, increasingly, for decision making as well as supporting economic growth. Data must be of sufficient quality to meet the specific users' needs. It must also be accessible in a timely manner. Both conditions are essential to achieving the crucial balance between making optimal use of our marine resources and protecting them.

And yet, despite being critical, this timely access to high-quality data proves challenging. Europe's marine data have traditionally been collected by a myriad of entities with the result that much of our data are scattered throughout unconnected databases and repositories. Even when data are available, often they are not compatible, making the sharing of the information and data-aggregation impossible. To tackle those problems in 2007 the European Commission through its Directorate General for Maritime Affairs and Fisheries (DG MARE) initiated the development of the European Marine Observation and Data network, EMODnet, in the framework of the EU's Integrated Maritime Policy. Today EMODnet is comprised of more than 160 organisations which gather marine data, metadata and data products and work together to make them more easily accessible for a wider range of users.

We will present how EMODnet has developed, currently consisting of eight sub-portals providing access to marine data from the following themes: bathymetry, geology, physics, chemistry, biology, seabed habitats, human activities, and coastal mapping. In addition, six sea-basin checkpoints have been established to assess the observation capacity in the North Sea, the Mediterranean, the Atlantic, the Baltic, the Artic and the Black Sea. This exciting and innovative approach to assessing the adequacy of our current European data provision system shall also be described.

Keywords: marine data; blue growth; EMODnet; thematic lots; sea-basin checkpoints

Crab community structure as ecological indicator of Matang mangrove forest in Malaysia

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Matang Mangrove Forest Reserve represents the largest contiguous mangrove area in peninsular Malaysia, and has been managed for silvicultural purposes for over a century. The management focuses on charcoal and poles production has ensured the continuous presence of a mangrove ecosystem over an area of 40,000 ha.

This ongoing research aims at investigating mangrove functionality. To accomplish this objective, crab community composition is assessed as an ecological indicator. The crab community is the most abundant macrofauna group in mangrove forests, it plays an important role in the nutrients cycle transfer between the mangrove plants and the soil at the time they are part of the trophic chain. Visual census methodologies for mangrove crabs were applied taking into account differing spatial behaviour of the species. The census and vegetation structure record was carried out in five different functional forest types (i) cleared area, (ii) young plantation (5-7 years), (iii) mid-age plantation (15 years), (iv) mature plantation (30 years) and (v) unmanaged forest (> than 90 years).

A positive correlation between vegetation age and crab community structure is expected. The findings of this ecological study will be fed into a transdisciplinary assessment, including stakeholder perception, to improve conservation and sustainable management of this ecosystem.

Keywords: mangroves; management assessment; crab communities; bioindicator; biodiversity; ecological functionality

MarineTraining.eu – An overview of Europe's Marine and Maritime education landscape

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A few years ago, a question arose:

'How to offer an overview of current Marine and Maritime education, whilst being a supporting framework to foster new training initiatives with exchange of best practice?'

In response, the Belgian node of EMBRC (European Marine Biological Resource Centre - a distributed Research Infrastructure (RI), of which Belgium is an associate partner through Ghent University, University of Hasselt, Royal Belgian institute of natural sciences and the Flanders Marine Institute) has set up a platform that brings together all available marine trainings at a European scale, and provides a series of services towards training organizers and policy makers. This comprehensive database focusses on higher education institutes (both universities and university colleges) and collects existing marine training initiatives for each country, ranging from master and doctoral programmes, to expert trainings and specialist courses. The platform is expanding gradually by including also non-accredited training initiatives (for instance, research institutes and industries) and via involvement in other European projects and networks. Services in the Marine Training Platform include advertising possibilities, practical services to trainees and training organizers (application and registration) and the support of marine dedicated e-learning initiatives.

In other words, the Marine Training Platform is the **one-stop-shop**
- for **trainees** in search of European Marine and Maritime training initiatives,
- for **trainers** in search of assistance for organizing European Marine and Maritime training initiatives,
- for **stakeholders** in search of insights into European Marine and Maritime training initiatives

For more information: www.marinetraining.eu

Keywords: education; training; marine; maritime; platform; EMBRC

EMBRIC – The European blue bio-economy cluster for aquaculture and biotechnology innovations

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EMBRIC - The European Marine Biological Research Infrastructure Cluster - is designed to accelerate the pace of scientific discovery and innovation derived from natural marine Bio-Resources. The novelty of EMBRIC lies in clustering 6 existing European Research Infrastructures (EMBRC, MIRRI, EU-OPENSCREEN, ELIXIR, AQUAEXCEL and RISIS) and 27 interdisciplinary partners from Academia, Research institutes, non-for-profit organizations and industry. Together, these will create new pipelines and novel applications in diverse fields, such as, drug discovery, aquaculture selective breeding, bioremediation, cosmetics and bioenergy.

One of EMBRIC's academic partners, Ghent University, dedicated two research groups to the cluster:
- The marine biology research group is in charge of training and exchange of best practice, by building an integrated training platform for the Blue economy industry and academia.
- The laboratory of protistology and aquatic ecology is in charge of proof of concept that genetically engineered microalgae can produce strains with improved performances in commercial applications.

Moreover, by interconnecting science, industry and policy, EMBRIC will defragment regional research, development and innovation policies. Hence, facilitating technology transfer, knowledge transfer and transnational access, by developing best practices and integrated training programs. Furthermore, the industry will have access to newly available resources, novel techniques and up-to-date industry standards to directly integrate results and protocols in commercial processes, putting forward Europe's Marine Blue Bio-economy innovation in the fields of aquaculture and biotechnology.

www.embric.eu

Keywords: blue growth; biotechnology; aquaculture; innovation; blue bio-economy; EMBRIC

Preliminary study of the water gradient within a Belgian offshore windfarm

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The FaCE-It project aims to examine the effects of sediment fining and hardening on benthic ecosystem functioning caused by anthropogenic impacts. Human activities, e.g. the construction of offshore windfarms, add artificial hard substrate to naturally soft sediments, altering the existing seabed habitats. The input of hard substrates leads to partial or entire replacement of the native benthic communities by fouling communities. The organisms comprising a fouling community act as active "biofilters" consuming organic compounds from the water column and releasing inorganic and organic materials, in the forms of faeces and pseudofaeces, to the surrounding environment – a procedure known as biodeposition. It is known that some fouling organisms, such as the amphipod *Jassa herdmani* and the hydroid *Tubularia indivisa*, build tube-like structures that absorb suspended particulate matter (SPM). These activities result in the alteration of the biogeochemical processes and could also lead to the SPM plumes that have been reported to occur in the Belgian offshore windfarms.

The aim of the present study is to identify the alterations caused by fouling communities to the water characteristics and analyze the water gradient – flow of the water compounds according to the currents - around an offshore windfarm (C-Power) in the Belgian Part of the North Sea. The initial hypothesis is that organic and inorganic materials will flow according to the currents and will appear in different concentrations in front, within and behind the entire windfarm structure. For this purpose, a sampling campaign was organized in order to collect water samples from different areas of the windfarm and analyze them for a variety of water characteristics, such as chlorophyll a, suspended particulate matter, particulate organic carbon and nitrogen and dissolved organic carbon. The results of this study will present the concentrations of the water gradient occurring in the offshore windfarms due to the presence of the fouling community.

Keywords: fouling communities; biodeposition; organic matter; water flow

The ticking time bomb of climate change and sea-level rise

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The 350-year window between 1750 and 2100 may seem like a long time in the context of human lifespans. However, in a seminal paper published in Nature Climate Change, Clark *et al.* (2016) have questioned the overemphasis in the global climate discussion on climate change as a 21st century only phenomenon, and on near term impacts up to 2100. They argue that any increases in carbon dioxide (CO_2) from human activity will remain in the atmosphere and continue to affect Earth's climate for tens to hundreds of thousands of years.

The extent of potential sea-level rise projected by Clark *et al.* for the much longer time frame of 10,000 years is orders of magnitude greater than that projected for the year 2100 in the IPCC 5th Assessment Report. The authors argue that this focus on near-term impacts has given a falsely reassuring impression that the effects beyond 2100 are of lesser importance and that it may be possible to reverse the effects through later emissions reductions.

It is imperative that future global climate policy discussions are underpinned by a clear understanding that we are already committed to significant global warming and sea-level rise that will have profound impacts on future generations and on Earth's ecosystems. Moreover, that continued emissions, even if reduced, will commit us to further warming and sea-level rise. The policy discussions must also recognise and support the critical role of climate and ocean science in ensuring an appropriate, evidence based response to the changes to come. The only option to avoid catastrophic climate change is to make rapid and fundamental changes to our energy, industrial and agricultural systems such that we move towards net-zero or negative carbon emissions within 20-30 years. This may sound dramatic, but compared to the potential human cost, it is basic common sense.

This paper summarizes the key massages of a Science Commentary published in 2017 by the European Marine Board (www.marineboard.eu).

Keywords: climate change; sea-level rise; policy; carbon emissions

Increased sea ice cover disrupts food web structure in coastal Antarctica

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Antarctica currently undergoes strong and contrasted impacts linked with climate change. While the West Antarctic Peninsula is one of the most rapidly warming regions in the world, resulting in sea ice cover decrease, the sea ice cover of East Antarctica unexpectedly tends to increase, possibly in relation with changes in atmospheric circulation. Sea ice is a major environmental driver in Antarctica, and changes in sea ice cover are likely to influence benthic food web structure through several processes (modifications of benthic-pelagic coupling, disruption of benthic production and/or modifications of benthic community structure and therefore resource availability for benthic consumers).

To date, regions where sea ice cover is decreasing have received more attention than regions where it is increasing. Here, on the other hand, we studied shallow (0-20 m) benthic food web structure on the coasts of Petrels Island (Adélie Land, East Antarctica) during an event of unusually high spatial and temporal (two successive austral summers without seasonal break-up) sea ice cover. Using time-tested integrative trophic markers (stable isotope ratios of carbon, nitrogen and sulfur) and state-of-the-art data analysis tools (bayesian ecological models), we studied the structure of the food web associated to benthic macroinvertebrates communities. In total, 28 macroinvertebrate taxa spanning most present animal groups (sponges, sea anemones, nemerteans, nematods, sipunculids, sessile and mobile polychaetes, gastropods, bivalves, pycnogonids, crustaceans, sea stars, sea urchins, brittle stars and sea cucumbers) and functional guilds (grazers, deposit feeders, filter feeders, predators, scavengers) were investigated.

Our results indicate that the absence of seasonal sea ice breakup deeply influences coastal benthic food webs in Antarctica. We recorded marked differences from literature data, both in terms of horizontal (i.e. primary producers and resources supporting animal populations) and vertical (i.e. trophic level of the studied consumers) structure of the food web. Overall, sympagic (sea-ice associated) algae dominated the diet of many important consumers, and the trophic levels of invertebrates were low, suggesting omnivore consumers relied less on predation and/or scavenging than in normal environmental conditions. Surprisingly, few animals seemed to feed on the extremely abundant benthic biofilm, whose exceptional development was also presumably linked with the peculiar sea ice conditions. Interpretation of data was complicated by the peculiar ecophysiological features of Antarctic invertebrates, whose very low metabolic rates could be associated to low tissue turnover. However, comparison of data obtained in the austral summers of 2013-2014 (first year without seasonal breakup) and 2014-2015 (second year without seasonal breakup) clearly showed that the observed trends were linked with actual temporal changes in invertebrate feeding habits rather than with other potential ecological drivers.

Our results provide insights about how Antarctic benthic consumers, which have evolved in an extremely stable environment, might adapt their feeding habits in response to sudden man-driven changes in environmental conditions and trophic resource availability. They also show that local and/or global trends of sea ice increase in Antarctica could cause strong changes in food web structure and therefore impact zoobenthic communities. This reinforces the view that, no matter their overall direction (i.e. increase or decrease), fluctuations in sea ice cover are likely to influence Antarctic benthic ecosystems' structure and functioning.

Studying plankton diversity in the Belgian Continental Zone: Research opportunities

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Planktonic organisms are indispensable for the functioning of marine food webs and are considered as one of the major drivers of global biogeochemical cycles. Because of their short life cycles, these organisms respond rapidly to environmental changes and consequently their abundance and species composition are indicators for the health of the pelagic ecosystem. The planktonic community is composed of numerous organisms from distinct taxonomic groups and their sizes range from about 0.2 μm up to 2 mm. The traditional sampling techniques, such as the WP2 net, filtering, and microscopic identification and counting, are still conducted. These are now complemented by new continuous sampling technologies such as the real-time high resolution underwater camera (VPR) and the Imaging Flow Cytometer which is connected to the continuous water flow system of the RV Simon Stevin. Furthermore, the (semi-) automated data processing software associated with these and other new technological developments (Zooscan and Flowcam) reduce the processing time of samples substantially. Combining these biological sensors, nearly the full size spectrum of phyto- and zooplankton is covered. Additionally, the phytoplankton fluorescence and productivity can be studied using the Fast Repetition Rate Fluorometer (FRRF). In this demonstration we show what the Flanders Marine Institute and LifeWatch infrastructure can offer the researchers to study and quantify the plankton communities in the Belgian part of the North Sea.

Keywords: zooplankton; phytoplankton; pelagic ecosystem; infrastructure; technological developments

Modelling soil erosion potential using remotely sensed data

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Soil erosion is one of the leading forms of soil degradation. Estimating soil erosion from field measurements is expensive hence the extent of soil erosion in many tropical watersheds is unknown. Erosion is a complex process; some of the eroded materials are deposited within the watershed while the rest is transported through waterways into lakes and lowland areas at the coast. The use of remotely sensed data and GIS provide a quick and reliable method to model, estimate, and monitor erosions over large areas even in data deficient tropical areas. In this study, Revised Universal Soil Loss Equation (RUSLE) model is being used to model soil erosion potential in a transboundary catchment of River Umba in Kenya and Tanzania. Data derived from MODIS (land cover), TRMM (rainfall), SRTM (elevation) and FAO (soil map) are processed in GIS environment for use in the model. Preliminary results, including; rainfall and run off erosivity (400 – 1700 MJ mm ha-1 h-1 yr-1), soil erodibility (0.018 – 0.025 t ha h ha-1 MJ-1 mm-1), slope length and steepness (0 – 1338), and vegetation cover factor (0.001 – 0.2) will be overlaid to predict the potential soil lost from the catchment annually. Results of this work will provide information on the extent of soil erosion in the catchment which has adverse effect on critical marine habitats. This information is key for policy makers and scientific community in making sound management decisions.Keywords: soil erosion; RUSLE; remote sensing; transboundary; critical habitats

Angelshark-ID: Photo-identification as a non-invasive monitoring tool

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Angelshark-ID is a research action created for long-term monitoring of angelsharks *Squatina squatina* in the Canary Islands, employing photo-identification. The aim of the project is to generate scientific base knowledge that allows the development of effective species management, while minimizing research impacts to the studied populations. In addition, this initiative responds to the need of creating a baseline for assessment and monitoring of the populations *S. squatina* in the Canarian Archipelago.

For data collection, visual underwater census were conducted in coastal areas from four of the seven Islands since 2006. The use of body marks is evaluated for use in individual identification of specimen and the recorded ecologic data to get insights in different life history aspects of the species. Parameters of interest include activities, population structure, growth and longevity, critical habitats, and population size. In addition, a tissue sample data bank was established for future genetic (ongoing) and isotopic analysis. During the 2016 sampling campaigns in the island of Gran Canaria, it was possible to characterize the individuals in more than 93% of the 173 encounters with angelsharks, resulting in the identification of 157 individuals. Resightings (11%) included individuals that were first observed during 2016 (8%) and two individuals that were first observed in 2007.

The high percentage of characterized individuals and the possibility to detect short, median and long-term resightments indicate that the innovative way of implementing photo-identification for *S. squatina* is a viable method for its monitoring. In addition, it is a technique that causes minimum perturbation to the species while it can provide essential information required for its population assessment and is therefore a potential conservation tool.

Keywords: angelote; squatina; individual identification; genetic assessment; population structure; Canary Islands

Unveilling the artisanal fish trade network in coastal Kenya (with focus on key stakeholders and fish products)

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Artisanal fisheries provide direct employment to several tens of millions and indirect employment to more tens of millions all over the world. Kenyan coastal communities have for long depended on fishing and related activities for their livelihood. Due to its longevity of existence and development, the small scale industry is itself a hive of activities- from fishing to marketing the final fish products. As such, the industry is characterized by a unique organization and challenges between and among the fishermen, buyers, government and other stakeholders such as NGOs. There are also interesting links and processes between the fishermen and small scale traders which shape the fish trade dynamics. Although the trade is described as tradition and artisanal by literature, various sources report that the markets are dominated by dynamism, competition and demand for more and new fish products.

This study will attempt to further understand the organization and challenges of the fish trade, at least among the small scale fishermen and buyers, by setting the following general objectives:
- To identify the challenges and intra-group organization among the fishermen and traders in the context of acquiring the fish and accessing suitable markets / products.
- To identify ways in which traders influence fishermen practices - by selection of fish and provision of capital.
- To get an estimate of fish landing weights and catch composition
- To quantify change in fish landing and fish price in the past years
-
Structured questionnaires, informal interviews, observations and weight measurements were used in collecting data. Primary results to be presented will include a network showing roles of key stakeholders in the fish trade and trends in the trade over the last 10 years. In addition, fish composition at landing and in the market as well as an estimate of price of fish at each trading level will be shown. Challenges reported by the stakeholders will be listed and categorized based on their cause *i.e.* social or ecological.

With this study we aim to enhance the understanding of the artisanal fish trade, which is paramount in supporting livelihoods. We also seek to identify interventions at the different trade levels that can make the trade more viable and sustainable.

Keywords: artisanal fisheries; stakeholders

Salt-making in mangroves, an economic activity that influences genetic diversity and structure of the fiddler crab *Uca annulipes* (H. Milne Edwards 1837)

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The fiddler crab *Uca annulipes* is a dominant species in mangrove forests along the East African coast. It enhances soil aeration and, through its engineering activities, makes otherwise-inaccessible food available for other marine organisms. Despite its importance, the habitat of *Uca annulipes* is threatened by human activities. Clearing themangroves for salt farming and selective logging of mangroves trees continue to jeopardise mangrove ecosystems in the Western Indian Ocean. This study aims to use partial mitochondrial COI gene sequences and nuclear microsatellites to determine whether salt farming activities in mangroves have impacts on the genetic diversity and structure of *Uca annulipes* collected along the Tanzania coast. The level of genetic diversities for both mitochondrial DNA and nuclear microsatellites are relatively lower in samples from salt ponds compared to natural mangrove sites. Analysis of molecular variance (AMOVA) among all populations showed low but significant differentiation (CO1: F_{st} = 0.022, P < 0.05; microsatellites: F_{st} = 0.022, P < 0.001). An hierarchical AMOVA indicates genetic differentiations between populations from salt ponds and natural mangroves sites (COI: F_{ct} = 0.033, P <0.05; microsatellites: F_{ct} = 0.018, P = < 0.01). These results indicate that salt farming has a significant impact on the genetic population structure of *U. annulipes*. Since higher genetic diversity and genetic connectivity contribute to a stable population, restoring the cleared habitats may be the most effective measures for the conservation of genetic diversity and hence adaptive potential to environmental change in this species.

Keywords: solar salt production; mangal forest; fiddler crabs: genetic variations; West Indian Ocean

Patterns of regional- and local- scale genetic connectivity in Eastern Atlantic mangroves

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Gene flow in mangroves is influenced by both intrinsic and extrinsic factors, of which the interaction controls the genetic connectivity between populations both close and remote. Identifying these factors and understanding the way they interact in shaping connectivity among populations at variable geographic scales allows to better understand how populations established, how coastlines have been colonized, how they might respond to climate change, and how conservation efforts can be strategized. We studied the genetic diversity of mangroves along the Cameroonian coastline to investigate the role of oceanic barriers and surface currents, tidal regime, estuarine geomorphology, and forest landscape structure in shaping the spatial distribution of genetic diversity at different spatial scales. We used a combined approach of genetic markers, mathematical modeling, and release-recapture experiments.

Our results indicate that contemporary ocean currents limit inter-regional gene flow, resulting in a clear genetic discontinuity between populations on either side of an oceanic convergence zone. Dispersal corridors created by the submergence of lowlands by historical sea levels rise have favored genetic connectivity between areas that were isolated during glacial times. Additionally, we found that the interplay of river currents and tidal patterns allow for asymmetric bidirectional dispersal along a major river, while winds and local hydrodynamics, forest structure, as well as hidden founders might contribute to among and within catchment connectivity in the most anthropized Cameroonian mangrove area - the Cameroon Estuary complex (CEC). Observed fine-scale genetic structure in these populations suggests that the vast extent of mangroves of the CEC persists due to recent re-expansion and re-colonization from micro-refugia. Genetic diversity hotspots were identified in the Rio del Rey and CEC, while populations in the southerly estuaries were less diverse.

Our findings highlight the importance of considering ocean surface current patterns and local hydrodynamics, and hence the importance of considering hydrological connectivity in formulating management practices and restoration measures for anthropized mangrove areas. Finally, our results also indicate that (climate-driven) changes in coastal geomorphology shape the distribution of genetic variability at different geographical scales.

Keywords: African mangroves; sea level rise; dispersal modelling; genetic connectivity; hydrochory

Interactive effects of next-century ocean acidification and warming on the common cockle *Cerastoderma edule*

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Estuarine habitats have high ecological and economical importance but are worldwide impacted by multiple stressors. To understand the impacts of future ocean scenario's, we assessed the combined effects of ocean acidification and warming on *Cerastoderma edule*, a species that play a key role in the estuarine ecosystem. Our experimental setting comprised of two factors in a fully orthogonal experiment; temperature (ambient or elevated (+3°C)) and pH (current or lowered (-0.3 pH units)). Condition (i.e. condition index) and other physiological responses (e.g. respiration, clearance and calcification) were measured after three and six weeks of incubation. Survival of the examined adult cockles persisted high and was not affected by elevated temperature (+3°C) nor lowered pH (-0.3 units). However, the morphometric condition index of the cockles incubated under high pCO_2 conditions (i.e. combined warming and acidification) was significantly reduced after six weeks of incubation. Respiration rates increased significantly under low pH, with highest rates measured under combined warm and low pH conditions. Calcification decreased significantly under low pH while clearance rates increased significantly under warm conditions and were generally lower in low pH treatments. The observed physiological responses suggest that the reduced food intake under hypercapnia is insufficient to support the higher energy requirements to compensate for the higher costs for basal maintenance and growth in future high pCO_2 ocean.

Keywords: future ocean; ocean acidification; ocean warming; *Cerastoderma edule*; ecophysiology

Relationship between water and sediment dynamics at Mariakerke beach

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Parts of the highly anthropized Belgian coast are vulnerable to storms which simultaneously combine high water levels and large waves. These storms can result in breaching the coastal defense causing the flooding of densely inhabited areas. To reinforce the weak coastal sections in order to meet the required safety levels in case of a catastrophic storm-event (with a return period of 1000 years), the Flemish government adopted the Master Plan for Coastal Protection in 2011. The Masterplan aims to find new sustainable coastal protection measures, alternative to the traditional ones, such as shoreface sand nourishments.

One of the weak links of the coast, Mariakerke beach, lies west of Ostend and it has been selected for a full-scale pilot experiment on shoreface nourishment. In order to evaluate the efficiency, safety and sustainability of the nourishment at Mariakerke beach is intensively monitored. Better insight into the relationship between hydrodynamics and sediment dynamics in this area is one of the objectives of the monitoring plan.

The waves, the marine currents, the suspended sediment concentrations (SSC) and the water level variations are monitored at four locations: two at section 100, Raversijde area, depths -3.5m and -6.5m TAW and two at section 104, Mariakerke area, at the same depths. At each location a metallic frame is deployed twice a year for approximately 6 weeks carrying instruments which measure the hydrodynamics for the entire water column (AWAC and Aquadopp ©Nortek) and in one fixed point (Vector ©Nortek) and sediment concentrations (Optical Backscatter Sensors - OBS). The OBS are mounted at three different levels within the first 1 m above the sea floor to capture the variation of sediment concentrations under various hydrodynamic conditions.

The objective of this work is to study the relationship between turbulent kinetic energy (TKE) and the contribution of waves and currents to the observed suspended sediment concentration distribution patterns. Consequently, the understanding of the sediment transport dynamics, both during normal weather conditions and during storm conditions and its variations with the depth are also set as objectives.

As expected, preliminary results indicate strong decrease of the sediment concentrations with the distance from the sea floor, mostly during normal hydrodynamic conditions. However, the largest sediment concentrations do not occur always during maximum velocity of the currents and maximum turbulent kinetic energy. The closer insight into these relationships which will be provided at the end of the study will significantly increase the knowledge on sediment dynamics at the Belgian coast.

Keywords: Belgian coast; hydrodynamics; sediment concentration; in situ measurements; shoreface nourishment

Hammerhead Shark Research: Knowledge from the populations in the Canary Islands

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Hammerhead Shark Research is a project that studies hammerhead sharks *Sphyrna spp.* in the Canary Islands with the aim to generate scientific base knowledge that contributes to the development of effective species management strategies.

This pioneering initiative, responds to the total lack of scientific information about these sharks in the Canarian Archipelago. Hammerheads are prone to bycatch in a wide range of fishery gears and interesting for shark fin trade, which make them vulnerable to overexploitation considering their life history characteristics. However, species knowledge in the Canary Islands is limited to the presence of the scalloped hammerhead shark *S. lewini* and the smooth hammerhead shark *S. zygaena*, isolated records about behaviour and scant sighting reports by citizens in social media.

To get insights into the population dynamics, tagging and tissue sampling is combined with biological and fisheries data collection in four of the seven islands of the Canary archipelago. In addition, the professional and recreational fishing communities are actively involved in the study and are, together with citizens, encouraged to report sightings of tagged and untagged individuals. Results of the initial study revealed the identification of two potential critical habitats for *S. zygaena* in the islands of Gran Canaria and Lanzarote. In addition, tissue samples were collected, post-release behaviour monitored and relationship with ectoparasites established. Future data collection and subsequent analysis is required to document the first local knowledge that can help in effective decision making of the species.

Keywords: cornudas; sphyrna lewini; sphyrna zygaena; tagging; population dynamics; citizen collaboration

Micropredator and parasites: an indicator for the status of angelsharks?

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Although parasitism represents the most frequent consumer strategy, they are usually not considered as conservation targets. There is, however, increasing evidence that parasites are extremely diverse and play key roles in ecological and evolutionary processes. In addition, they have been used as indicators for ecosystem change and to illustrate the ecology of their hosts. It is yet to be explored whether host-parasite interactions can be used as an indicator for the vulnerability of angelshark *Squatina squatina* populations.

Recently, novel associations were documented from parasites and a micropredator with elasmobranchs in the Canary Islands based on long-term (10-year) observations in their natural habitat. The marine leech *Stibarobdella macrothela* and the isopod *Aegapheles deshaysiana* were regularly observed taking blood meals from *S. squatina*, while the marine leech *Branchellion torpedinis* was rarely encountered preying on *S. squatina* and the marbled electric ray *Torpedo marmorata*. No noteworthy aspects were observed in the attachment area, the behaviour, and the apparent fitness of the hosts and preys.
These findings provided useful information about biologic diversity and the relationship of marine leeches and an isopod with *S. squatina* and *T. marmorata* in the Canarian Archipelago, and are relevant for the future conservation management of these species. Future research challenges include the assessment of:
- The prevalence and intensity of parasites and micropredators in the angelshark population.
- Parasites and micropredators selectivity (single-host or multihost), and their relation with intrinsic and current vulnerability of angelsharks.
- Evidence of host-parasite co-extinction.

Keywords: *squatina squatina*; *aegapheles deshaysiana*; *branchellion torpedinis*; *stibarobdella macrothela*; isopod; marine leech

A new version of the Maritime Boundaries Geodatabase

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The United Nations (UN) Convention on the Law of the Sea (UNCLOS), which was signed in 1982 and came into force in 1994, defines a series of maritime zones (internal waters, archipelagic waters, territorial sea, contiguous zone, exclusive economic zone and continental shelf) and establishes the degree of rights and obligations of a country in each of those areas.

The Exclusive Economic Zone (EEZ) is the basic geo-unit to be considered with regards to a country's management of marine natural resources. This includes sectors such as mineral exploration and exploitation, energy, fisheries, biodiversity and species conservation, etc. Despite the strategic significance of EEZs, a standard georeferenced product with maritime boundaries was not available at the global level (Claus et. al, 2014), until it was developed and made available by the Flanders Marine Institute (VLIZ) in 2006 (Deckers and Vanden Berghe, 2006).

The product developed at VLIZ consisted of two GIS layers providing both the maritime boundaries (lines) and the EEZs (polygons). The layers were regularly updated with 8 consecutive versions published between 2006 and 2014. Version 9 was launched by the VLIZ-hosted portal Marineregions.org in October 2016 and implied major new developments and features. Straight and archipelagic baselines were included, together with remaining areas defined by UNCLOS: internal waters, archipelagic waters, territorial seas and contiguous zones.

To create the new Maritime Boundaries Geodatabase (Flanders Marine Institute, 2016), more than 22000 point data were collected and converted to lines to delineate baselines and treaties. The main source for these coordinates was the UN repository of all the claims from UNCLOS's signatories. When there are no treaties available for two territories a median line is calculated following the equidistance principle. This is achieved by drawing Thiessen polygons rooted on points along both the straight and normal baselines. The seaward outermost boundary for the EEZ, the contiguous zone and the territorial sea were drawn by calculating a buffer distance of 200, 24 and 12 nautical miles respectively, measured from the baselines.

The Maritime Boundaries is the most popular product available at Marineregions.org. In all its different versions, the product has been downloaded a total of 36674 times (as of end 2016), representing an average 64% of the total downloads. These boundaries and other derived products are used in many projects related to bio-geographic research and conservation, such as the World Register of Marine Species (WoRMS), The Sea Around Us, Global Fishing Watch or The Ocean Health Index.

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- Keywords:

Diversity and natural distribution of electricity-generating cable bacteria in the seafloor

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Very recently, long filamentous so-called "cable bacteria" have been discovered in marine sediments that are able to generate and mediate the transport of electrons across centimeter-scale distances. These electrogenic bacteria make that the ocean floor operates like a natural battery, and thus cable bacteria have an enormous potential for novel bio-electric applications. Cable bacteria belong to the family Desulfobulbaceae and have been identified in sulphide-rich coastal environments. Because cable bacteria have only been recently discovered, they remain enigmatic in many aspects. The goal of this Master thesis project is to gain a better understanding of their distribution in the natural environment as well as their phylogenetic diversity.

Enumeration of cable bacteria is evaluated using fluorescent in situ hybridisation with a probe specific for the family Desulfobulbaceae in combination with the filamentous growth form of the cable bacteria. Cable bacteria abundance will be quantified at different depths to yield distribution profiles in the sediment.

The phylogenetic diversity of cable bacteria will be studied by extending the currently small number of available full 16S rRNA sequences. Sediment samples in which cable bacteria have been detected from a wide variety of locations worldwide will be used to amplify the 16S rRNA gene region using primers applicable for the Desulfobulbaceae family. The extended dataset will be used to investigate the phylogenetic affiliation and evolutionary relationships of cable bacteria.

Keywords: cable bacteria; long-distance electron transport; natural abundance; fluorescent in situ hybridisation; phylogenetic diversity; 16S rRNA gene

Ionic liquids: Defending ships' hulls against corrosion and biofouling

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Ships' hulls are the target of two ongoing threats. On the one hand, there is corrosion, attacking the structure of the metal and shortening the active life of the ship; on the other hand, organisms attach themselves to the outer layers and form large organic conglomerates of matter (called fouing), leading to a loss of hydrodynamic shape and to a substantial increase of fuel consumption during the voyage. In recent years, an innovative class of chemical compounds, the ionic liquids, has arisen, which may act against both corrosion and fouling. These chemicals, liquid at ambient temperatures (< 100°C), consist completely of ions: usually an organic cation, such as (a derivative of) imidazolium, and an inorganic anion such as a chloride, bromide, tetrafluoroborate or hexafluorophosphate, although organic anions have been used as well. Several different types of ionic liquids (such as imidazolium derivates) have been found useful to combat metal corrosion (see e.g. Arenas and Reddy 2003, Likhanova et al. 2010) or bacterial growth (e.g. Pernak et al. 2003, Fitzpatrick et al. 2005, Ganske & Bornscheuer 2006); however, the exact mechanism is still largely unknown.

To test the anticorrosive action of different imidazole-based compounds under maritime conditions, grade A steel coupons of 2 cm by 8 cm by 6 mm were etched for 15 min in 37% HCl containing 20 g/L Sb_2O_3 and 60 g/L $SnCl_2$ to remove existing layers of rust. These coupons were then cleaned in running tap water and submerged overnight (20-24 h) in 1 M H_2SO_4 containing one of the ionic liquids at one of two concentrations: 100 mg/L or 10 mg/L $C_{16}C_1$-imidazolium bromide, 400 mg/L or 40 mg/L 1.3-dioctyl imidazolium bromide, 100 mg/L or 10 mg/L $C_{12}C_1$-imidazolium chloride, or 100 mg/L or 10 mg/L 1,6-imidazole ionene. The coupons were attached to small frames, labelled, and put in the sea for the specified amount of time. They were suspended in sea water in the Ostend harbor at a platform, designed for these experiments (Swain and Schultz 1996, Casse and Swain 2006). After one month, a series of coupons was taken out, etched again to remove all corrosion and fouling and weighed again. Significant corrosion inhibition was observed on the coupons treated with the higher concentrations of the ionic liquids. Further tests are needed, however, to elucidate whether these ionic liquids are suitable for maritime use, to protect ships.

Additionally, some coupons, having been exposed between one day and one month in the seawater, were also collected for microbiological analysis. To this end, the coupons were taken out of the sea, fixed in 3% formaldehyde, washed in tap water, dehydrated in three steps in ethanol (50% - 75% - 100%) and dried before further microscopic analysis. Coupons were stained with DAPI (visualizing the cells' DNA) to assess the spread of the microfouling, which is currently ongoing.

Lastly, toxicological testing using *Artemia* sp. as a test organism showed that the toxicity of the imidazolium derivates depends upon its substituents. The low overall toxicity of the ionic liquids supports the possible use of these components.

Keywords: corrosion; fouling; paint; ionic liquids; toxicity

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The acoustic receiver network: a sea of opportunities

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Technology does not stand still and it helps researchers to perform high-quality scientific research. Improving technology allows us to gather continuous information on ocean processes, animal behaviour and environmental variables using automated devices. The acoustic receiver network for instance, allows flexible and cost-efficient spatio-temporal tracking of migratory fish species. This network uses acoustic telemetry to gather the data. In the framework of LifeWatch (http://www.lifewatch.be), which was established as part of the European Strategy Forum on Research Infrastructures, the Flanders Marine Institute (VLIZ) and the Research Institute for Nature and Forest (INBO) created this receiver network to support biodiversity research and environmental impact studies. The network currently consists of 177 receivers, covering both the Belgian part of the North Sea, the Western Scheldt Estuary and different rivers in Belgium. Detailed observations of animal movements and behaviour in relation to the aquatic environment can significantly improve our understanding of ecosystem functioning and dynamics (e.g. migration routes, spatio-temporal habitat use and migratory behaviour). In addition, it provides the scientific basis for fisheries management, species protection, marine spatial planning and environmental impact assessments.

But having a network of devices, each able to capture up to 1.6 million detections, leads to big data. Big data easily goes beyond the limits of normal spreadsheets, requires a lot of RAM memory and requires (too) much calculating power from your personal computer when performing the analyses. Thus, we have to look for alternative solutions to work with this kind of data.

VLIZ hosts a PostgreSQL database able to store all the information. The database is linked to a collaborative environment (R-server) where all analyses of the data can be performed and results can be visualized using the R-shiny application. To facilitate the data-entry for researchers, a web interface was developed in PHP using symphony framework. There are several entry forms to manage metadata on receivers, tags and animals and upload the detections into the database in an automated way.

This database is a first step towards a European animal tracking network (ETN). Such a network will: 1) improve the capacity to monitor large scale movements of marine animals, 2) foster cooperation between scientists across borders and 3) foster efficient use of resources and compete for large scale European funding opportunities.

Do you want to know more? Visit us at the LifeWatch demo booth!

Keywords: telemetry; acoustic receiver; network; ETN

Dimethylsulfonopropionate as a reactive oxygen species scavenger for phytoplankton cell

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The goal of this project is to improve knowledge about the role of dimethylsulphonopropionate (DMSP) and dimethylsulfoxide (DMSO) as antioxidant for phytoplankton and the impact of light intensity on their production and degradation by phytoplankton. In addition to their antioxidant role for the phytoplankton, DMS(P,O) are the precursors of the dimethylsulfide (DMS), a gas affecting the climate through the production of atmospheric aerosols. These lasts could have an effect on terrestrial albedo by creating clouds.

The production of DMS(P,O) by phytoplankton is specific and varies with environmental conditions in particular light. The research methodology will combine laboratory experiments, field measurements and mathematical modelling. DMS(P,O) production processes and related enzymatic pathways will be studied in laboratory experiments based on monospecific cultures of key phytoplankton species in different light conditions. We have chosen for laboratory experiments four key species: *Phaeocystis globosa, Skeletonema costatum, Nitzschia closterium* and *Thalassiasira pseudonana.*

Process level studies will be carried out using two complementary techniques:
(1) The expression of several candidate genes implied in DMSP production will be examined using molecular techniques (qPCR). This expression will be done in parallel to
(2) The production/degradation of DMSP and DMSO measured by gas chromatography for the phytoplankton species of the Southern North Sea (SNS) in different environmental conditions of light.

Reactive oxygen species (ROS; OH-, O2-, H2O2) relative production will also be measured to estimate their variability in different light conditions. Monthly field measurements will allow to quantify the importance of DMS(P,O) production and DMS emission in the SNS area related to phytoplankton diversity and environmental factors. It will also be measured the relation between the presence of phytoplankton species and bacteria diversity thanks to genomic data with in mind that DMS(P,O) could be the key molecule between them. A temporal series could be made to have a better understanding in time scale.

Data from laboratory experiments and field measurements will then be used to calibrate and validate the DMS(P,O) dynamics in the biogeochemical MIRO-DMS model applied to the SNS. Model simulations will allow to (i) determine the relative importance of biotic and abiotic factors in the production of DMS(P,O) by aquatic ecosystems and (ii) improve the prediction of current and future emissions of DMS.

Keywords: DMS; oxidative stress; DMSP; phytoplankton

The influence of pile driving noise on harbour porpoises

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The harbour porpoise (*Phocoena phocoena*) is the most common marine mammal in the Belgian part of the North Sea and is protected by both national and EU law. In the North Sea, the harbour porpoise is considered under threat because of high bycatch levels and increasing noise pollution. Impulsive pile driving noise originating from the construction of offshore wind farms has been shown to affect porpoises up to distances of 20 km from the noise source. Driven by high porpoise densities in Belgian waters, a pile driving ban is in force from the start of January up to the end of April. However, The Netherlands have the Borssele offshore wind farm at only one kilometre away from the Belgian offshore wind farm zone, and do not enforce such an embargo. Considering the high mobility of harbour porpoises, there is a need for improved insights into the impact of pile driving noise on porpoises which can serve as a basis for an objective evaluation of the respective legal regimes.

From May to September 2016 pile driving was taking place at the Nobelwind wind farm located on the Bligh Bank in Belgium. In this period, porpoise activity was recorded using passive acoustic monitoring (C-PODs), at various distances from the construction site (1 – >15 km). In this study we compare porpoise detections before, during and after pile driving with the focus on the influence of repeated pile driving events. In addition to porpoise monitoring data, noise measurements and noise levels are extrapolated for the different locations. The data for this study were obtained from the RBINS wind farm monitoring programme and the VLIZ Lifewatch observatory.

Keywords: harbour porpoise; underwater noise; pile driving; passive acoustic monitoring

Lipidomics of harpacticoid copepods in a changing ocean

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Long-chain polyunsaturated fatty acids (LC-PUFA) like omega-3 are highly concentrated in marine fish and receive a lot of attention in view of their considerable health benefits for humans. In the marine environment, basically all long-chain fatty acids are made *de novo* by microalgae but can be modified by bioconversions as they pass up the food chain. However, this fatty acid supply may be threatened in the future since LC-PUFA production is predicted to be reduced in phytoplankton as a result of climate warming (Hixson et al. 2016).

Since these omega-3 fatty acids are essential for zooplankton growth and consequently for the development of higher trophic levels (fish) feeding on them, knowledge on the assimilation and modification of fatty acids in first-level consumers/grazers like copepods is crucial to anticipate the effects of global change. However, the factors triggering fatty acid bioconversion in marine grazers remain unknown.

Therefore, this PhD research aims (1) to determine the efficiency of transfer of fatty acids from primary producers (diatoms) to consumers (harpacticoid copepods) in a global change scenario and (2) to study the molecular pathways of fatty acid biosynthesis and metabolism by means of transcriptomics.

A transcriptome will be assembled which we will use to identify desaturase/elongase genes in a harpacticoid copepod. Subsequently, mRNA expression of these genes under different experimental conditions of temperature and pH can be analyzed. The results can be used to translate the fatty acid pathways at the individual and species level to the community level by means of food web modeling and should improve our knowledge on potential effects of global change on the production of fatty acids.

Keywords: transcriptomics; fatty acid biosynthesis; global change; omega-3; copepods

Reference
- Hixson, S. M., Arts, M. T. (2016). Climate warming is predicted to reduce omega-3, long-chain, polyunsaturated fatty acid production in phytoplankton. *Global Change Biology*, *22*(8), 2744–2755. https://doi.org/10.1111/gcb.13295

The "Landgull": how important is terrestrial food for Lesser Black-backed gulls *Larus fuscus* in Belgium, and are there consequences to this diet?

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Human action has benefited a range of opportunistic species through the mass production of food waste. Urban and industrial garbage, together with fishery discards, have provided the Lesser Black-backed Gull (*Larus fuscus*) with an opportunity to thrive along the coasts of the North Sea, and increasingly inland. Fishery discards are one of the main food sources exploited by this species in Belgium during the breeding season (May to July), but these are bound to gradually disappear due to changes in fishery practice induced by European legislation. Lesser Black-backed gulls breeding in the region will soon be left with mostly terrestrial options to feed their chicks. Given this scenario, our research addresses the following questions: how abundant is terrestrial food in the current diet of local *L. fuscus* chicks? Does raising a Lesser Black-backed Gull chick on a fully terrestrial diet have consequences on its growth and condition? Is it plausible that the Lesser Black-backed Gull population breeding in Belgium will be fully dependent on terrestrial food in the near future?

Results from aviary experiments evidence that it is possible to fully raise a Lesser Black-backed Gull chick on terrestrial food. This, however, comes at the cost of delayed growth curves that may affect winter survival rates. The proportion of terrestrial to marine food currently comprising the diet of *L. fuscus* chicks raised in the Port of Zeebrugge was assessed at different ages by means of stable isotope analysis. These proportions in chick diet are compared to foraging patterns of their parents, obtained by GPS tracking of adult birds, to assess time and energy costs implied in obtaining each type of food. The high adaptability of both chicks and breeding adults to exploit varying sources of food shows that gulls may dispense with fishery discards and still thrive locally, as long as terrestrial food remains largely available at low foraging costs.

Keywords: gulls; fishery discards; garbage; breeding; aviary experiments; stable isotope analysis

We want YOU to join the EMODnet/IMEC Open Data Competition!

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EMODnet[a] is teaming up with IMEC[b] to launch an Open Data competition within the context of the Colombus[c] project.

The main objective is to investigate how the accessibility and uptake of data (and data products) from EMODnet by end-users (policy, industry, science and wider society) can be improved. EMODnet and IMEC will collaborate to explore the potential of EMODnet-data and to demonstrate and valorise this potential in an open data contest, including an innovation bootcamp with guidance for participants.

During five consecutive days/evenings, the selected teams are coached and given the chance to advance their ideas and concepts based on the EMODnet open datasets. The following courses and interactive workshops are included in these five days:
· Open Data workshop with open data experts
· LLAVA matrix introduction and application with business model experts
· User Innovation toolbox and 'get-out-of-the-building' with user research experts
· Hackathon/prototyping the open data solution with prototyping experts

EMODnet together with IMEC will then select the most promising solutions. Results will be widely disseminated across the EMODnet consortium.

http://www.emodnet.eu/
http://www2.imec.be/be_en/home.html
http://www.columbusproject.eu/

GPS tracking network of Lesser Black-backed Gulls and Herring Gulls breeding at the southern North Sea coast

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As part of our terrestrial and marine observatory for LifeWatch (http://lifewatch.inbo.be and http://www.lifewatch.be/en/gps-tracking-network-large-birds), the Research Institute for Nature and Forest (INBO), the Flanders Marine Institute (VLIZ), Ghent University (UGent), and University of Antwerp (UA) are tracking Lesser Black-backed and Herring gulls with lightweight, solar powered GPS trackers. The project builds upon the extensive knowledge the INBO has acquired over the last 15 years when studying in particular postnuptial migration, as well as mate and site fidelity of large gulls, by means of sightings of colour-marked individuals ringed in Belgium and via individual-based life-history studies by UGent and UA. The data collected through this bird tracking network allows to study the migration patterns and habitat use of the gulls in more detail. Furthermore, they are no longer biased towards locations where observers can see colour-ringed birds. To allow greater use of the data beyond our research questions, all data are published as open data. The recorded data are periodically released in bulk as open data (http://dataset.inbo.be/bird-tracking-gull-occurrences), and are also accessible through CartoDB and the Global Biodiversity Information Facility (GBIF).

During the afternoon demo session Eric Stienen will present some persuasive results of the tracking study showing interesting aspects of gull migration, habitat use and individual specialization and he will elaborate on unexpected gull behaviour during the breeding season.

The importance of the little things: Bacteria-diatom interactions in intertidal mudflats

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Estuaries are one of the most valuable ecosystems on earth; their productivity at times even matching those of coral reefs and tropical forests. This, largely due to their microscopic component. The estuarine mudflat biofilms, in particular, are hotspots of primary production, nutrient cycling and microscopic diversity. Diatoms, eukaryotic microalgae, are one of the main primary producers in the biofilms. They share the biofilm with large numbers of bacteria (10^9 cells/ml of sediment). We have shown that these bacteria and diatoms interact in numerous ways, ranging from antagonistic to synergistic. The bacteria-diatom interactions are complex as they are highly specific and condition-dependent. They can have important ecological implications for the composition and functioning of the biofilm community. We can conclude, that whilst occurring on a microscopic scale, these interactions might be powering the mudflat ecosystem and might thus be one of the main reasons why these systems are so exceptionally productive and valuable.

Keywords: microbial interactions; bacteria; diatoms; mudflat; biofilm; interkingdom signalling

Automatic monitoring of birds in marine video content

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Technology makes it possible for scientists to study animals in their natural environment, without being present at that location. For example a camera at a nesting area allows scientists to observe from a distance. This reduces time spent on visiting the targeted area and limits the number of nest disturbances. The resulting video content gives scientists a unique insight in the life and behavior of the animals filmed. However, this leads to many hours of raw video content, which still have to be processed.

In the framework of LifeWatch, the Flanders Marine Institute (VLIZ) installed several cameras in a marine environment. Next to behavioral aspects, scientists are interested in the number of birds present during a certain time frame. This could be done by continuously watching and counting birds throughout the video content. But this is very time consuming, so an automatic processing is researched. The biggest problem encountered is the typical background in marine video content, namely the sea. Water is very dynamic, and next to waves and water ripples, also light reflections and shadows are visible on the surface, and can easily be interpreted as birds.

Most research done on the image processing of marine video content is for maritime purposes (harbor security and control). In this research, the techniques for maritime processing are tested for the monitoring of birds. An automatic detection of ships is deemed less difficult, due to the larger, and above all, static form of ships.

To identify the foreground items (i.e. birds) in video content, the background is subtracted first. A very dynamic background (i.e. water) is hard to distinguish from the foreground and a common problem in dynamic background subtraction (DBGS). In this research it was found impossible to remove all waves from marine video content without also removing some birds.

To counter this problem a two-step detection of birds was created. At first, without removing any birds, as many waves and reflections on the water as possible are removed. In a second step, the resulting foreground candidates are classified as water or bird by an offline trained classifier. Finally all valid detections are tracked throughout the video content (individual bird tracks).
- Step 1: For the removal of the dynamic background, several pixel based and block based techniques were tested. Pixel based techniques have a higher detail (per pixel) and are less complicated, and thus faster than block based techniques. But neighboring foreground pixels must be merged into a foreground candidate in each frame.
- Step 2: Birds can be distinguished from water by an image texture analysis. Namely the pattern of image gradients for birds differ sufficiently from water gradients to train a bird-water classifier on. A very rudimentary explanation is to envisage a bird as the Japanese flag (centered object), and water as mostly horizontal bands, like the flag of the Netherlands.
- Step 3: Tracking throughout the video is done by linking overlapping detections in subsequent frames. Once two detections are linked, the trajectory of the object is used. New detections are checked with the location predictions of all active objects. This works well for most cases, but is insufficient for birds that fly by near the camera (no overlapping).

The software created in this research allows for an automatic processing of marine video content. The output consists of a new video fragment in which detected birds are marked (identifier, bounding box and track) and the number of birds for each frame is indicated. Next to the new video fragment, a metadata (json) file is written. This file can be queried to know when (frame number) and where (position) birds were seen throughout the given video.

This research allows for scientists to focus on the statistical analysis of birds in a video without having to look at the entire video fragment.

Keywords: dynamic background subtraction; texture analysis; image classification; object detection; tracking; seabirds; marine environment

Impact of macrofaunal activities on the biogeochemical cycling in anthropogenically disturbed sediments

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Marine coastal areas are important as they provide a wealth of goods and services to humanity. These goods and services result from many ecological processes, many of them occurring in the sediment. Therefore, it is a major challenge to understand and predict how human activities will affect the link between benthic communities and benthic mineralization processes. In this study, we investigate how activities of macrofaunal organisms (bio-irrigation and bioturbation) contribute to benthic biogeochemical cycling in a range of natural sediments, and sediments affected by human activities.

We conducted closed-core incubations of sediments sampled along a gradient from fine to coarse sediments, including sediments subjected to anthropogenic fining. Sediment-water exchange of nutrients and oxygen were followed by sampling the overlying water every 2 hours. Bio- and physical irrigation was quantified by following the decreasing concentration of a bromide tracer in the overlying water. At the end of the incubation, the sediment was sieved and the organisms retrieved to calculate the bioturbation potential of the community (BPc). With those data, we will estimate the degradation rate and transformation of organic carbon deposited at the sediment surface, and the nitrification/denitrification rates. Furthermore, we will assess whether the effect of macrofaunal activity on benthic ecosystem functioning varies with natural and human induced changes in sediment properties.

The research plan and the preliminary results will be reported.

Keywords: ecosystem functioning; nutrients fluxes; bio-irrigation; bioturbation; oxygen consumption; macrobenthos

Determining nutritional quality and chemical changes during storage of fish silage produced using different combinations of undersized quota species

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Fishery byproducts are often underutilized, even though they form an excellent source of nutritional proteins and fats. A problem concerning fishery byproducts is that these materials are very unstable and processing methods such as freezing or drying are often expensive, requiring large volumes before they become feasible. Producing fish silage could be an alternative for countries with smaller fishing communities, such as Belgium.

Fish silage implies the liquefaction and stabilization of minced whole fish or fish offal by the addition of mineral and/or organic acids. It is considered a low-investment, low-cost and easy-to-produce fish meal substitute. Fish silage quality is strongly influenced by the initial composition and quality of the raw materials. Therefore, according to the industry, a steady supply of fresh, low-variety raw materials is required to ensure product uniformity and quality. However, many of the European demersal fisheries are mixed fisheries, thus supplying a large variation of raw materials.

During this research the nutritional value was determined and chemical changes were monitored in four types of fish silage, each produced using a different combination of undersized Belgian quota species. The following nutritional parameters were investigated: degree of hydrolysis (DoH), dry matter (DM), crude protein (CP), crude lipid (CL), ash, amino acids (AA), fatty acids (FA), total volatile basic nitrogen (TVB-N), trimethylamine (TMA), and thiobarbituric acid reactive substances (TBARS). The goal of this research was to determine the stability of the fish silages over a 3-month time period and to ascertain the effect of raw material combination (RMC) on fish silage quality.

There was no difference in initial CP (74%DM) and in all fish silages CP decreased over time. However, CP of the less complex RMCs levelled off at approximately 64%DM after 3 weeks, whereas the more complex RMCs exhibited a stronger decrease in CP over time and dropped to levels below 60%DM after 3 months. Accordingly, these complex RMCs also displayed a slower but longer DoH compared to less complex RMCs, 73.5% and >85%, respectively. Extended hydrolysis leads to a loss of protein-N in the form of NH_3 and a decrease in nutritional value.

Initially there were slight differences in CL between the fish silages, ranging from 5.0 - 9.3%DM. After 3 months all silages had a similar CL of approximately 5.8%DM. Ash decreased slightly (2.8%DM) and DM increased slightly (1.2%) in all fish silages during the 3-month storage period. The low DM of approximately 24.6% negatively influences the economic value of the silage.

All silages seemed to be a good source of essential and non-essential amino acids. The fatty acid profile showed a large amount of polyunsaturated fatty acid (PUFA). However, the relatively low CL values signify that the silages are not an ample source of PUFA.

TVBN – an indicator for the freshness of the raw materials – is the product of NH_3 and TMA, and increased significantly during storage. The relatively stable TMA values in all the fish silages indicate that the increase in TVBN is mainly due to the production of NH_3, which corresponds with the CP decrease. Initial TVBN and TMA values were slightly above the limits of 35 mg/100g and 8 mg/100g, respectively. TBARS remained below the limit of 8 mg/100g in all fish silages indicating that there was limited lipid oxidation.

There seems to be an effect of RMC on the nutritional value of fish silage and a decrease in quality during storage. The decrease in quality is mainly based on the changes in the protein fraction since this was the most dominant change and fish silage is often used as protein source in animal feed. Product pasteurization could limit hydrolysis, thus minimizing protein losses. Fresher raw materials should be used in the future to minimize TVBN and TMA. Also, a concentration step could be added to increase the DM content.

Keywords: fish silage; nutrition; fishery byproducts; valorization; fishmeal substitute

Phycotoxins in sea spray aerosols: friend or foe?

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Throughout our history, coastal environments have been linked to the promotion of human health. While the first written references date back to ancient Romans, there is evidence that ocean-therapy was already used to treat health conditions in Ancient Egypt. Today, epidemiological studies continue to observe that coastal populations are healthier and have longer life-expectancies than urban populations. The food and air quality (physiological hypothesis), as well as the recreational possibilities oceans and coastal areas offer (blue gym hypothesis), are suggested as main contributing factors. At the same time, these aquatic systems are under pressure by global change, the introduction of invasive species, overfishing and eutrophication. The relative recent increase of harmful algal blooms (HABs), previously referred to as red tides, is believed to be a consequence of these anthropogenic disturbances.

About 100 HAB-species are classified as toxin producing algae, able to cause human intoxications via several exposure pathways. The **consumption of poisoned fish and shellfish** is the main documented pathway for human exposure to these phycotoxins. Much less is, however, known about the exposure via the **inhalation of sea spray aerosols** (SSAs). Phycotoxins that are actively excreted or released after cell rupture may aerosolize by bursting air bubbles at the sea surface. So far, exposure to brevetoxins and ovatoxins via SSAs has caused severe respiratory irritation in vulnerable groups living around the Gulf of Mexico and the Mediterranean. These events show that the inhalation of phycotoxins, via SSAs, is an important and underappreciated exposure pathway. However, such an exposure does not have to be strictly negative, i.e. causing adverse health effects. It was recently hypothesized that the improved health in coastal areas could be related to the regular exposure to low levels of airborne biogenic compounds (**biogenics hypothesis**). Products from bacteria, algae, fungi and plants are thought to inhibit specific kinase pathways (PI3K/Akt/mTORC1) whose activity has been linked to pathological conditions such as tissue inflammation, cancer, diabetes, immunosuppression, and neurodegenerative diseases.

To test this biogenics hypothesis we conducted a series of proof of principle experiments. We examined the effects of this respiratory exposure by exposing human lung cell lines for two days to several major phycotoxins (brevetoxin, okadaic acid, domoic acid & yessotoxin). Using MTT assays on alveolar (A549) and bronchial (BEAS-2B) epithelial cells, the cell metabolic activity was measured as a proxy of cell viability. In these experiments, compared to the other tested phycotoxins, yessotoxin (YTX) exhibited the highest toxicity. This is somewhat surprising since YTX is generally accepted to be one of the least potent marine toxins. Extrapolating these effect concentrations of YTX to environmental concentrations in the air, based on the surface area of the exposed cell tissue, the levels causing an effect were of the same magnitude as the measured brevetoxin concentrations during a bloom of *Karenia brevis*. This illustrates the possible human health importance and environmental relevance of the tested concentrations range. Analysing the effect of YTX on the activity of the PI3K/Akt/mTORC1 pathway, using SDS-page and western blotting, a possible hormesis effect (i.e. a positive effect at (very) low exposure concentrations) was found at concentrations approximately ten thousand lower than the discussed viability effect concentrations. Algae producing YTX, however, do not have the same blooming capabilities as *K. brevis*. It can thus be suggested that in ambient coastal air (cf. sea spray) only YTX concentrations producing a hormesis effect – i.e. a health promoting effect - can be reached in natural circumstances. If confirmed by subsequent testing, these experimental results are the first supporting the biogenics hypothesis.

Keywords: harmful algal blooms; biogenics hypothesis; phycotoxins; sea spray aerosols

Role of mangroves and salt marshes for nature-based flood risk mitigation in major deltas of the world

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Coastal areas are facing several threats, on the one hand due to their population that is expected to increase considerably over the coming decades and on the other hand due to climate change, resulting in rising sea level and increasing cyclone intensity generating flooding risks by storm surges and coastal erosion by waves.

Over the past years and in some regions, it has been proposed to use preserved or restored coastal habitats, such as tidal wetlands, for protection of populations and economic assets from coastal hazards, often in addition to hard engineering structures as dikes or embankments. This approach defined as nature-based management, relies on the ability of the wetland vegetation to mitigate the impact of storm waves and surges, and to adapt by sedimentation to sea level rise, in addition to other valuable ecosystem services.

Our study investigates a specific aspect of the nature-based management approach, namely, the benefits in terms of storm surge mitigation of salt marshes and mangrove forests for low-lying areas and populations by comparing the consequences of their presence and absence during storm surge events in the world's most populated deltas. We investigate these consequences using a GIS model based on globally available data and relatively simple assumptions that assesses the relative magnitude of the nature-based flood risk mitigation by salt marshes or mangrove forests in large delta areas.

We applied our model to 11 deltas with a worldwide distribution. For every delta, the presence of tidal wetlands influenced the flood pathway, the storm surge passed through the tidal wetlands during the flooding, and then resulted in a benefit in terms of storm surge mitigation. Nevertheless, the results present a large diversity among the deltas in terms of the relative magnitude of storm surge mitigation. This diversity highlights the influence of the local characteristics of the deltas, such as effects of the channels structure, the location of the tidal wetlands in the delta or the topography of the low-lying areas.

Two main conclusions could be drawn from the results. There is no correlation between the total surface area of tidal wetlands in the delta and the land area benefiting from flood mitigation, however, the relative magnitude of flood risk mitigation is higher when tidal wetlands occupy a larger area of the delta.

A second concern in the study was the influence of the tidal wetlands on the mitigation of flood risks for the delta population. Results show some dissimilarity with the land area benefiting from flood risks mitigation. Those differences are reflecting the impact of the population distribution and density as well as the historic settlement and development of the population.

Despite the large variations in the magnitude of flood risk mitigation by tidal wetlands over our 11 deltas in terms of land area and delta population, we can affirm that at a delta scale tidal wetlands are providing flood risk mitigation.

Keywords: tidal wetlands; flood risk mitigation: delta: storm surge

The impact of dredging on the geochemical cycling in coastal ecosystems

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The intensity of anthropogenic activity in the coastal ocean is rapidly increasing, and this puts more and more pressure on local marine ecosystems (Halpern et al., 2008). Industrial activities, such as bottom trawling and dredging, have a profound impact on the seafloor via the relocation and homogenization of the surface layer sediment (IADC/CEDA, 1997). Dredging is connected to the maintenance of man-made waterways and access channels to harbours, the construction of wind farms and coastal defence, as well as land reclamation. In the European Union alone, dredging results in the excavation of 200-250 million tons of sediment per year, of which ~80 % is redeposited in marine environment (EuDA annual report 2005), thus illustrating the large scale on which this operates in coastal waters.

The coastal seafloor receives high amounts of organic carbon, which limits the oxygen penetration depth, and allows for the preservation and burial of organic carbon. This makes these sediments an essential carbon sink that couples the long- and short-term carbon cycle (Middelburg and Meysman, 2007). Frequent dredging in coastal waters directly disturbs this carbon sink and thus could leave an anthropogenic fingerprint on the global carbon cycle. To date, the magnitude of this impact has however not been quantified.

We investigated the sediment biogeochemistry at a field site in the shallow Southern North Sea, ~5 km offshore the Belgian coast (BCZ130, N 51°16.3', E 2°54.3') during 12 consecutive monthly campaigns over 2014. Unexpectedly, a major disturbance event took place between the May and June 2014 sampling campaigns, which affected the upper ~15 cm of the sediment and had a major impact on the geochemical cycling. Evidence from both *in situ* data and model simulations suggest that this disturbance was caused by the deposition of a 15 cm thick layer of dredged harbour sediment. Our results show that this deposition event induced a reset of the geochemical cycling within the upper seafloor, which leads to an alternating sequence of electron acceptors, following the thermodynamic energy gain. It takes > 1 year for the mineralisation rates to return to the steady state. This result implies that 1 - 2 depositions per year keeps the seafloor in a permanent transient state. The anthropogenic influence on the biogeochemistry of these coastal systems is likely larger than previously acknowledged.

Keywords: dredging; carbon cycle; mineralisation; transient diagenesis

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Modeling mangrove propagule dispersal trajectories using high-resolution estimates of ocean surface winds and currents

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Mangrove forests are systems that provide ecosystem services and rely on floating propagules of which the dispersal trajectories are determined by ocean currents and winds. Quantifying connectivity of mangrove patches is an important conservation concern. Current estimates of connectivity, however, fail to integrate the link between ocean currents at different spatial scales and dispersal trajectories. Here, we use high-resolution estimates of ocean currents and surface winds from meteorological and oceanographic analyses, in conjunction with experimental data on propagule traits (*e.g.*, density, size and shape) and dispersal vector properties (*e.g.*, strength and direction of water and wind currents). We incorporate these data in a dispersal model to illustrate the potential effect of wind on dispersal trajectories of hydrochorous propagules from different mangrove species. We focus on the Western Indian Ocean, including the Mozambique Channel, which has received much attention because of its reported oceanic complexity, to illustrate the effect of oceanic features such as eddy currents and tides. In spite of the complex pattern of ocean surface currents and winds, some propagules are able to cross the Mozambique Channel. Eddy currents and tides may delay arrival at a suitable site. Experimentally demonstrated differences in wind sensitivity among propagule types were shown to affect the probability of departure and the shape of dispersal trajectories. The model could be used to reconstruct current fluxes of mangrove propagules that may help explain past and current distributions of mangrove forests and assess the potential for natural expansion of these forests.

Keywords: connectivity; eddy currents; long distance dispersal; Mozambique Channel; tidal motion; Western Indian Ocean

First steps to rear *Crangon crangon* larvae successfully in captivity

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In Europe, *Crangon crangon* or the European brown shrimp is economically a very important species because of its high commercial value. The past few years, the demand for big live shrimp (> 7 cm) has increased and has a higher sales value than its cooked counterpart. Unfortunately, fisherman cannot meet this demand. But due to this rising demand and decreasing availability from wild shrimp stocks, this species could be a suitable candidate for aquaculture.

Despite its high commercial value, little attention has been paid to improve the rearing techniques for this species and people have not yet succeeded to rear *C. crangon* in captivity. Attempts to "catch and hold" wild adults in land-based rearing systems have been proven to be very difficult, due to high mortality and slow growth.

In order to establish a commercially viable culture of the European brown shrimp, this research focused on the development of a rearing system for larvae. Larviculture forms a crucial step for the establishment of a viable aquaculture for crustaceans. One of the major constraints is the availability of qualitative seed and larvae, but also mechanical damage, cannibalism, adequate food intake and efficient metamorphosis during the hatchery phase are important bottlenecks that have to be overcome by good aquaculture practices.

During the first trials one-day old larvae were reared in Erlenmeyer flasks with 500 mL of sterile seawater (300 larvae.L^{-1}). Three different diets were tested in triplicate namely *Artemia* nauplii, *Artemia* nauplii in combination with micro-algae and one-day old enriched *Artemia* nauplii. Survival of each treatment was determined weekly. After 21 days the survival rate for each of the treatments was 67.61 % (*Artemia*), 75.65 % (combination) and 61.30 % (enriched *Artemia*). Although the combination of *Artemia* and micro-algae resulted in a higher survival rate in comparison to the other two treatments, the difference was not significant (P>0.05). In case a high survival is needed to get a large batch of juveniles for further grow-out settings, we advise to feed the larvae with a combination of *Artemia* nauplii and micro-algae. Also the feeding rate of the larvae was determined with individual trials. Three feeding regimes were tested namely, 10, 30 or 50 *Artemia* nauplii per day per larvae. Survival and feeding rate were determined daily. At the end of this experiment a feeding regime was suggested based on the major moulting periods during the larval development. This means that the amount of nauplii per larvae (per day) is increased with 5 to 10 nauplii per larvae after each major moulting event (± each 7 days). This way the larvae have enough feed to develop and reach the next zoeal stage.

Based on these results we took the first steps to rear *C. crangon* larvae successfully in captivity. This research will enable us to provide an effective larval rearing system to maximize production of healthy post-larvae for further potential grow-out settings.

Keywords: *Crangon crangon*; European brown shrimp; larviculture; larval rearing

Influence of anthropogenic measures on large-scale estuarine morphodynamics

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We investigate the influence of the embankment of tidal basins (anthropogenic measures) on large-scale estuarine morphodynamics. To this end, we employ a depth-averaged numerical morphodynamic model to describe the hydrodynamics and resulting morphodynamic evolution. We find that the embankment of a tidal basin can profoundly impact the location and orientation of the main channel the estuary and the location and orientation of the elongated tidal banks (tidal delta) in the mouth region.

Our model findings are compared with historical bathymetrical data of the Scheldt estuary, which is a tidally dominated estuary is located at the border between The Netherlands and Belgium. The estuary has a profound economic value, providing e.g. maritime access to several ports in Belgium and The Netherlands. Over the past two centuries, the geometry and bathymetry of the Scheldt estuary have profoundly evolved, a.o. due to anthropogenic measures such as land reclamation. Comparing modelling results with historical bathymetrical data appears to support our findings.

Keywords: morphodynamics; estuary; tidal delta; anthropogenic measures

Historical agricultural land use leads to reduced groundwater dynamics in a restored freshwater tidal marsh

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In the past centuries, many tidal marshes along estuaries and coasts have been embanked to gain land for industry and agriculture, leading to severe problems such as an increased flooding risk, loss of biodiversity and a deteriorating water quality. Nowadays, numerous of these former marshes are being restored to recover their ecosystem functions. There are, however, more and more indications that restored tidal marshes do not deliver these ecosystem services to the same extent as natural tidal marshes. In particular, we hypothesize that a reduced groundwater flow, caused by historical agricultural soil compaction, implies a reduced biogeochemical nutrient cycling and decreased contribution of restored marshes to water quality improvement.

Conducted research

We measured groundwater flow and soil properties in both a natural freshwater tidal marsh (De Plaat) and a restored freshwater tidal marsh (Lippenbroek) along the Scheldt estuary in Hamme, Belgium. The soil in the restored marsh consists of a layer of freshly accreted sediment, underlain by compact relict agricultural soil. Our results indicate that groundwater level fluctuations are occurring over a deeper soil profile in the natural marsh (median groundwater level 34 cm below the surface) compared to the restored marsh, where groundwater level fluctuations are restricted to the layer of freshly accreted sediment (median groundwater level 8 cm below the surface). Using X-ray CT-scans of soil cores, we found that the soil in the natural marsh and the upper layer of the restored marsh is intersected by macropores that increase the hydraulic conductivity of the sediment, whereas these macropores are absent in the compacted agricultural soil. As a result, the compact agricultural soil forms a barrier for groundwater flow and puts constraints on the amount of pore water that is exported to the creeks in between tidal events. By consequence, nutrient cycling in restored tidal marshes is expected to be negatively affected.

The next step

We now aim to link the groundwater dynamics to the biogeochemical functioning of freshwater tidal marshes. Furthermore, we will determine the optimal soil structure and creek density a restored tidal marsh should have to maximize its nutrient cycling function. To accomplish this goal, we will set up a mesocosm experiment in which different soil treatments (e.g. adding organic matter or creating artificial macropores) will be applied to the compact agricultural soil. Additional field measurements will be performed to set up a numerical groundwater and solute transport model. Based on this model and in consultation with the hydraulic engineering sector, the ultimate goal of the research is to formulate viable design criteria for future tidal marsh restoration projects.

Keywords: tidal marsh restoration; Scheldt estuary; groundwater dynamics; soil structure; nutrient cycling

Towards a climate resilient coast: Numerical modeling of wave overtopping and wave loads considering the influence of sediment transport

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The Belgian coast is only 67 kilometer long, but has nevertheless a high touristic, socio-economic and cultural value. Throughout history, the shoreline has been significantly modified. From the end of the 19th century until after the Second World War a series of high apartment buildings were constructed on the sea dike. Furthermore since 1970, millions of cubes of sand were artificially added to the beach as coastal defence measures, pushing the sea further away from the dikes. This resulted in the typical Belgian coast as it is known today: a mildly sloped beach in front of a dike with a promenade and an almost continuous line of high rise buildings.

Coastal safety against flooding is provided by a hard dike structure, fronted by a nourished beach and a storm wall on the crest of the dike. However, due to the climate change, the sea level is expected to rise and the severity and frequency of extreme storm events will increase. The Belgian coastal defence system is therefore adapted according to the Master Plan Coastal Safety by a combination of beach nourishment and dike crest increase by a storm wall, providing as such enough safety for the entire Flemish coast until the year 2050.

Within the design of these coastal defence measures, data such as wave loads and wave overtopping volumes are taken into account. Though, an accurate prediction of wave overtopping and wave loads on structures at the Belgian coast is not straightforward.

At present, traditional EurOtop prediction formulae are used to provide a quantitative description of the overtopping processes according to the original Master Plan methodology. However, these empirical formulas are based on assumptions (e.g. concerning geometry, wave boundary conditions...) that are too conservative for this type of cross-section.

New research actions are organized to improve understanding of processes and development of more accurate design tools within CREST (Climate Resilient coaST, www.crestproject.be). Within the CREST project, an integrated modeling of wave, flow and sediment in the swash zone will be carried out. A numerical (2DV) model will be developed employing the OpenFOAM code, solving the Reynolds-averaged Navier-Stokes (RANS) equations. Navier-Stokes type models resolve the flow over the complete water depth and allow modeling of the complex overtopped flow.

Sediment transport is known to have a significant effect on wave propagation during a storm, and thus also on the wave overtopping and wave loading processes. Therefore, our research will first focus on the incorporation of a sediment transport module within the OpenFOAM source code to capture all relevant sediment transport processes (i.e. bed load and suspended load transport). With this model, we aim to reduce empiricism in the present bed load formulations under combined current-wave action. On the other hand, we aim to study the loss of energy by mobilized sediment and the changing morphodynamics of the beach before waves reach the dike. Wave flume experiments including sediment transport documented in literature will be used to validate the numerical model.

Keywords: openFOAM; sediment transport; wave overtopping; wave loads; Belgian coastal safety; CREST

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Sole in the Irish Sea: Do fishermen and fisheries-scientists see things from a different perspective?

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Fishing opportunities for sole (*Solea solea*) in the Irish Sea have declined severely in recent years. Due to consecutive TAC reductions Belgian fishermen fear the loss of a historically important fishing ground and claim that the low numbers observed in the British beam trawl survey (UK-BTS) do not reflect what they observe at sea: a healthy stock consisting of a broad age distribution. To determine whether this is a true mismatch between science and industry a Fisheries Science Partnership (FSP) was set up in order to gather additional information on sole in the Irish Sea.

From 2013 onwards, the data collection on commercial fishing trips to the Irish Sea was intensified. This fisheries-dependent data showed that for some parts of the Irish Sea catches (LPUE) have remained stable or even increased. However, since 2016 directed fisheries for sole in the Irish Sea is no longer allowed, making it difficult to follow up. Consequently, the scientific survey remains in the sole assessment as the most important data source. Fishermen are sceptical about scientific surveys, especially when livelihoods are at stake. More specifically, the reliability of the scientific method for data collection is questioned.

As part of the Irish Sea project (IRIS), funded by the European Maritime and Fisheries Fund (EMFF), ILVO organized an industry survey. This type of scientific survey involves close collaboration with the fishing industry. A commercial beam trawler was chartered to deliver information on areas (and species) that are not covered by the scientific survey. Moreover, an industry survey aims to improve communication between scientists and fishermen and aspires to gain industry support in scientific methods for fisheries data collection. Three main research questions were focussed on: 1) What is the distribution of sole in the Irish Sea, investigating also areas not covered by the UK-BTS? 2) Is there a difference in catch composition between the research vessel and commercial vessel? 3) What is the geographical origin of sole in the Irish Sea?

The industry survey was organized from 15/09/16 – 02/10/16, where the commercial beam trawler Jasmine (Z.483) performed parallel fishing with RV Endeavour (performing the scientific beam trawl survey UK(E&W)-BTS-Q3) in 28 locations. Another 42 locations, systematically spread over 11 ICES statistical rectangles in the Irish Sea, were sampled only by the commercial vessel to gather information on sole and other demersal fish species. To study the geographical origin and reveal important nursery grounds of sole, tissue and otoliths were collected (spring and autumn) from the Irish Sea and the adjacent areas of the Celtic Sea and Bristol Channel. Samples were analysed using three different stock identification techniques conducted at KU Leuven (next-generation sequencing, otolith shape analysis and otolith elemental analysis).

First results show that in almost all statistical rectangles more than half of the catch was above minimum landing size (24 cm). However, proportionally more juvenile sole (< 24 cm) were caught in the eastern part of the Irish Sea (ICES statistical rectangles 36E6, 37E6 and 38E6), implying the presence of the nursery grounds. In that area also proportionally more sole (in numbers and weight) were caught. No other areas with exceptionally high numbers of (juvenile) sole were found and the catch composition (*i.e.* size distribution) of the conventional scientific survey agrees with the industry survey. Moreover, no other commercial species were caught by the commercial vessel compared to the scientific survey. Finally, during the survey a close and positive collaboration between scientists and fishermen was established.

These findings do not yet solve the problem of the opposing perceptions on the abundance of sole. Perhaps there is no mismatch between science and industry, but the fish observed by the fishermen were born elsewhere and therefore not detected by the scientific survey on the main nurseries in the eastern Irish Sea. Further research will provide more insight on the geographical boundaries of the Irish Sea sole stock, with potential implications for future management of this stock.

Keywords: *Solea solea*; Irish Sea; Fisheries Science Partnership (FSP); industry survey; stock identification

A near-shore phytoplankton bloom in Belgian waters observed from space

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With the launch of the first Sentinel-2 (S2) satellite on June 23, 2015, the capabilities of space-borne remote sensing of coastal and inland water quality improved considerably. A single S2 satellite has a revisit time of 10 days globally, but can observe the Belgian coast twice in every 10 day period. A second S2 unit will be launched in 2017, effectively doubling this revisit frequency. The main imager on board of S2, the MultiSpectral Imager (MSI), has a spatial resolution of 10-60 m and is hence able to resolve small features in surface turbidity of aquatic ecosystems. Moreover, thanks to the inclusion of a spectral band at the red spectral edge, the chlorophyll a absorption in the red can be quantified. In the past year, the mapping of chlorophyll a concentration at 20 m spatial resolution with S2 imagery has been demonstrated by various teams.

Here we present the discovery of a near-shore phytoplankton bloom that occurred early May 2016 in the very near-shore part of the Belgian coastal zone, in front of the port of Oostende. The satellite image reveals a bloom with an extremely high chlorophyll a concentration. This kind of bloom would be very difficult to detect with ship-borne measurements, due to the bloom location and extent, and the shallowness of the water. Traditional ocean colour satellites lack the required spatial resolution to resolve these near-shore events. With a five or ten day revisit time, and these novel chlorophyll and suspended sediment mapping capabilities, the S2 mission will contribute significantly to the understanding of near-shore phytoplankton dynamics and sediment transport not only in Belgian waters, but also globally. S2 derived data will be of significant importance for the near-shore monitoring required by the European Water Framework Directive.

Keywords: coastal zone; remote sensing; phytoplankton bloom; chlorophylla

Coupling methodology for modelling the near field and far field effects of an array of wave energy converters

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Wave Energy Converters (WECs) are devices used to capture ocean energy into useable electricity. In order to produce a large amount of electricity at a competitive cost, arrays composed of large numbers of WECs will need to be deployed in the ocean. Due to hydrodynamic interaction between the WECs (near field effects), the geometric layout of the array is a key parameter in maximizing the overall array power production and minimizing far field array effects on the surrounding area and wave field. Consequently, it is essential to model both the near field and far field effects of a WEC array. Modeling both effects by employing a single numerical model that offers the desired precision at a reasonable computational cost, is, however, still a great challenge.

Two types of models are mainly used to model WEC array effects: wave interaction solvers and wave propagation models. Wave interaction solvers can accurately model the physical processes of wave energy absorption by solving the body motion. They are accurate but their computational cost increases exponentially for large numbers of WECs in arrays and large domains and therefore when modelling far field effects. Wave propagation models can model the WEC array far field effects in large domains at a reasonable cost, however the simplifications when representing the WEC near field effects and absorption can lead to errors. The objective of this research is to present a coupling methodology that will combine the strengths of both types of models to model the entire spectrum of WEC array interaction.

The proposed coupling methodology is based on a one-way coupling between the wave interaction Boundary Element Method (BEM) solver, NEMOH, and the depth-averaged mild-slope wave propagation model, MILDwave. In a one-way coupling the wave field for each numerical model is calculated independently. In the presented cases, NEMOH is used to resolve the near field effects whilst MILDwave is used to resolve the far field effects.

The coupling methodology consists of the superposition of two different wave field simulations: an incident wave field and a radiated/diffracted wave field. The incident wave field is calculated intrinsically in MILDwave. The diffracted/radiated wave field is calculated around the WEC array in NEMOH and then propagated in MILDwave by imposing it on an internal wave generation boundary along a circle. The WEC type presented in this study is a heaving flat disk buoy, similar to the WECs currently planned for commercial array projects in the UK, Australia and Sweden.

Results are presented for different sets of conditions for regular waves with varying wave periods. In the immediate domain around the WECs (the near field), the resulting wave field is compared to the wave field provided by NEMOH, used to assess the accuracy of this coupling methodology. A good agreement is found between the NEMOH wave filed and the coupled wave field in MILDwave. The effects of varying the number of bodies, the incident wave period and the coupling radius are detailed. The advantages and disadvantages of the coupling methodology are also discussed when modelling WECs arrays.

Keywords: WEC arrays; hydrodynamic interaction; wave propagation; coupling; MILDwave; boundary elements method

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Waves: an ocean of modelling options

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Half of the world's population lives within 60 km of the coastline, and 8 of the 10 most populated cities are located at the coast. All of these cities have major harbour infrastructure, which has to withstand the power of the ocean waves. Additionally, the ocean is a huge alternative energy source; ocean waves have a higher energy density than wind, and the first commercial tidal turbines are already operational. As coastal engineers, our goal is to design coastal and offshore structures, able to survive storm conditions; even those occurring only once in 100 or even 1000 years.

An optimal design of coastal and offshore structures is only achievable by having an as good as possible understanding of the physical properties of ocean waves, such as surface elevation, orbital velocities, dynamic pressure, wave energy. This is done by attempting to describe the real-life behaviour with mathematical models. There are several possibilities to model ocean waves, each one with its advantages and disadvantages. The goal of this research is to highlight the ones applied by coastal engineers.

Within the coastal engineering field, models are applied for several types of research: hydrodynamics, morphodynamics, wave-structure interaction, wave propagation, wave statistics…. Here, focus is put specifically on wave propagation models such as:
- Analytical models based on Airy wave theory
- Spectral wave models
- Phase-resolving wave models:
- Mild-slope equation models
- Boussinesq models
- Computation fluid dynamics (CFD) models
- Smoothed particle hydrodynamic (SPH) models
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The models listed above will be discussed based on their simplicity, computational cost, accuracy, applications but also limitations.

Keywords: ocean waves; numerical modelling; coastal engineering

Visualization of small lesions in skin of fish using fluorescein dye

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Assessing the effects of anthropogenic activities on the health of fish is important to safeguard the marine ecosystem. The impact of fisheries is mostly evaluated by monitoring the fish's reflexes or skin lesions. The latter is done with the naked eye. However, in this way, possibly present non-visible, microscopic lesions may be missed. Although these may seem small, they still may have a non-negligible impact on the health of fish. Indeed, they may be colonized by pathogenic bacteria such as *Vibrio tapetis* and *Aeromonas salmonicida* leading to the development of skin ulcerations. To allow the implementation of this risk factor in the impact-assessment of anthropogenic activities, these lesions need to be visualized.

The present study aimed at pinpointing a protocol to map microscopic skin lesions in salt water fish by means of fluorescein sodium salt. Fluorescein sodium salt is a yellow, nontoxic, water-soluble dye that is commonly used in ophthalmic medicine. Due to its high degree of ionization this dye is not able to penetrate or bind to intact epidermis. When the epithelial barrier is damaged, the fluorescein can penetrate the tissue and fill up intracellular spaces. Although fluorescein already proved its use in detecting skin lesions in fresh water fish it is not widely used in salt water species. In first instance, both macroscopic and microscopic skin lesions were inflicted on freshly euthanized marine fish by means of a scalpel, needle or rubbing with a net. Various immersion times (2min-4min-6min) in a fluorescein supplemented bath (0.2 mg/ml) and consequent rinsing protocols (2x 1min or 3x 1min) were tested. Fish were subsequently photographed under long wave-length ultraviolet (UV) light to visualize the lesions.

All induced lesions were clearly visible under ultraviolet light even if they were not discernible with the naked eye as they were stained with the fluorescein dye.

Generally, the shorter the immersion time, the weaker the staining. Therefore, the best staining was found when fish were placed in the fluorescein supplemented bath for a minimum of 6 minutes. With regard to the rinsing protocol, it was noted that three rinsing steps with clean sea water for one minute, were necessary to eliminate nonspecific staining of the fish which hampers a clear evaluation of the lesions.

To conclude, using the fluorescein dye allows to plot skin lesions in marine fish that are not macroscopically discernible. This rapid and cost-efficient method may prove valuable in various applications (e.g. fish disease monitoring; impact of fisheries, aquaculture), where an assessment of the impact on the fish's skin is needed, both macroscopic and microscopic.

Keywords: skin lesions; visualization; fluorescein dye; marine fish

Distribution of zooplankton in the coastal area of the Belgian part of the North Sea, with focus on the harbors of Oostende, Zeebrugge and Nieuwpoort

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The mesozooplankton community structure and its spatial variability in the Belgian part of the North Sea (BPNS) was studied in relation to the interaction between physicochemical stressors, such as temperature, salinity, nutrients and pollutants. This is the first study that integrates the main Belgian harbors of Nieuwpoort, Oostende and Zeebrugge. The sampling campaign at 13 stations during June 2015 yielded a total of 67 different mesozooplankton taxa (19 classes, 21 taxa identified at order level, 15 at genera level and 22 at species level). From this, 44 taxa were considered holoplanktonic, 18 meroplanktonic and five tychoplanktonic. The order Maxillopoda dominated the mesozooplankton (79%), especially with high densities of calanoid (57%) and harpacticoid (15%) copepods, followed by appendicularians represented by *Oikopleura dioica* (9%) and ophiurid larvae (7%). The predominance of small copepods such as *Acartia (Acartiura) clausi, Temora longicornis, Paracalanus parvus, Pseudocalanus elongatus, Centropages hamatus* and *Euterpina acutifrons* characterized the mesozooplankton community of the BPNS. Based on the mesozooplankton diversity and the ubiquitous spatial distribution of the dominant species, the mesozooplankton in the BPNS could be described as one neritic community with the occasional appearance of oceanic species. In terms of density, the mesozooplankton showed a density peak in the midshore stations. The mesozooplankton was distributed in small-scale assemblages with dominance of certain species in different zones and the communities were mainly influenced by temperature, salinity, ammonia, phosphate and acenaphthene. Hence, environmental stressors may influence the densities of the mesozooplankton in the BPNS. The effect of seasonality between spring (March) and summer (June) was marked by differences in density, with highest densities noted in June. The copepod communities also exhibited a succession of species. However, this effect was more marked in the harbors with an increase in density from cyclopoids during spring compared to summer. The demographic composition of the species *Acartia (Acartiura) clausi* and *Temora longicornis* was dominated by copepodite stages I-III with variations in adult sex ratio with females being more abundant than males. This study is the first effort to identify mesozooplankton communities in a high spatial resolution taking the influence of diverse environmental parameters and pollutants into account. It provides a detailed insight on the current status of zooplankton in the BPNS and contributes to the development of predictive models.

Keywords: mesozooplankton; copepods; marine biodiversity; community

Is the sea cucumber *Holothuria polii* a potential indicator species for organic enrichment in the sediments of coastal lagoons?

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Anthropogenic organic enrichment is a major threat to coastal lagoons. To quantify the impact of this pollution, organisms can serve as ecological indicators if their behavior is directly correlated with changes in pollutant levels.

Aspidochirote holothurians or deposit feeding sea cucumbers are sensitive to changes in the sea-floor composition because they ingest sediment to obtain nutrition. However, limited research has been done on their potential as an indicator. Their preference for enriched sediment suggests a correlation between holothurian density and sediment organic matter content.

This study investigates the suitability of *Holothuria polii* as an indicator species for coastal lagoons. Holothurian abundance and sediment enrichment level were measured during June 2015 and May 2016 in control and enriched sites in the Mar Menor lagoon in Spain.

A significantly higher density of holothurians was observed in the enriched sites. However, more organic matter content was found within the control sites, which is possibly linked with seagrass abundance. The lower holothurian densities associated with this natural enrichment suggest that *H. polii* prefers anthropogenic organic matter which is easier to digest. These preliminary results encourage more research into the feeding habits of *H. polii* in order to further evaluate their indicator potential.

Keywords: indicator species; organic enrichment; coastal lagoons; holothurians; *Holothuria polii*

Impact of seasonal environmental stress in sea ice on the production and emission of dimethylsulfide by microbial communities

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Sea ice plays a significant role in the exchanges of climate active gases between the ocean and the atmosphere. One of them, dimethysulfide (DMS), is a precursor of sulfate aerosols which affect the Earth radiation balance. DMS is produced by the degradation of two algal metabolites: dimethylsulfoniopropionate (DMSP) and dimethylsulfoxide (DMSO). The dynamic of these sulfur compounds in the sea ice ecosystem is poorly understood. In particular, little is known about the factors driving the production of DMS and of its biological precursors in the stressing environmental conditions (temperature, salinity) of the brine channels in which sea ice algae grows. In this context, the objective of this research project is to quantify the impact of seasonal environmental stress in brine on sea ice algae, focusing on the production of DMSP and DMSO. We will study the production of sulfur compounds by axenic cultures of two characteristic species of the polar ocean: a diatom, *Fragilariopsis cylindrus* and a prymnesiophyte, *Phaeocystis antarctica.* On one hand, as a reference point, we will determine the sulfur compounds production by both algae under typical ocean conditions in the polar area (temperature of 4°C and salinity around 34-35). On the other hand, we will focus on the DMSP and DMSO production under the seasonal environmental conditions encountered by both algae when their living environment is the sea ice: the incorporation into the sea ice matrix during the autumn, the survival in the brine channels during the winter and then the return to more favourable conditions in spring. For comparison, we will also perform the same experiments with natural sea ice algae samples collected in the Arctic and Antarctic. Globally, the outcomes of this research project should drastically improve our understanding and modelling capabilities of the sea ice sulfur cycle.

Keywords: sea ice; brines; DMS; DMSP; DMSO; microalgae; stress

From measured 2D short wave spectrum to infragravity wave surface modulation in deep water

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Infragravity waves (IGs), often referred to as gravity waves with periods from 0.4 to 4 min (Herbers 1995), are generated by the interaction of primary wind waves offshore. When the whole wave set travels across the ocean as far as the near coastal region, short waves steepen and eventually break. Due to the long wavelength of IGs, they never reach the steepness required for breaking and are able to travel all the way to the coast. Under stormy conditions, IG contain much more energy when travelling close to the coast due to shoaling process. They are also believed to play an important role in harbor oscillations and nearshore sediment transport (Herbers 1995).

Within the framework of the CREST project, from the short wave spectrum which has been measured during a storm in January 2016 at location Westhinder, the corresponding water surface variations caused by IGs have been computed. The calculation procedure has been verified by comparing to the analytical solution (assuming a constant water depth). Sensitivity tests show that the total long wave energy varies quadratically with the short wave energy, and that the long wave energy increases dramatically at lower water depths.

The following step is to incorporate the computed long wave water level variation in the water level variation in the coastal region. The objective is to take into account the complex processes that occur nearshore: the release of the bound IG into free IG during the short wave breaking, and the generation of additional IG energy due to the time varying breaking point. We have identified two main limitations to this methodology, with heavy computational time being the main difficulty.

Keywords: infragravity wave; offshore; water surface modulation

Reference
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VLIZ Master Thesis Awards

Een 'Search Group Algoritme' voor de optimalisatie van offshore windmolenparken

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In onze tijd wint hernieuwbare energie meer en meer aan belang en in vele landen is het al een belangrijke pijler van de energievoorziening. Windenergie is een betrouwbaar en betaalbaar alternatief voor de klassieke bronnen en los van het duurzame karakter wordt de groei ervan mede gestimuleerd door de dalende productiekosten. In het Belgische klimaatplan is offshore windenergie onmisbaar. De bestaande en geplande windmolenparken op de Noordzee staan garant voor 50% van de te behalen hernieuwbare energieproductie tegen 2020 volgens de Europese doelstellingen.

De productie van een windmolenpark hangt af van allerlei factoren zoals het aantal en type windturbines. Een evenzeer cruciale factor voor het rendement is de lay-out van het windmolenpark i.e. de positionering van de verschillende turbines. De totale productie van een windmolenpark ligt lager dan de som van de capaciteit van de individuele turbines. Dit is te wijten aan het zog die zich achter een turbine vormt, de zogenaamde zogeffecten. Lay-out optimalisatie van een windmolenpark streeft ernaar deze effecten zoveel mogelijk te reduceren en voor elke windturbine de beste positie te bepalen voor een maximale energieproductie. Vermits de lay-out van windmolenparken aan land vaak bepaald wordt door externe landschapskenmerken is deze methode vooral praktisch voor offshore parken waar er doorgaans meer speling is op de positionering.

Dit onderzoek focust op het toepassen van het 'Search Group Algoritme' op dergelijk optimalisatieprobleem. Hiervoor werd een mathematisch model van de zogeffecten en van de energieproductie geïmplementeerd. Tevens werd ook de invloed van de instellingen eigen aan het algoritme, zoals bijvoorbeeld het aantal iteraties, onderzocht. Na berekening verkrijgt men de optimale lay-out en de ermee gepaard gaande productiecapaciteit. Om de prestaties van het algoritme te evalueren werden de resultaten vergeleken met deze van twee referentieproblemen. Het gaat om het 'Evolutionary Algoritme' en het 'Imperialist Competitive Algoritme', twee courante algoritmes uit de literatuur die als maatstaf gelden. Daar de resultaten van het 'Search Group Algoritme' variëren met de willekeurig gegenereerde initiële populatie werd elk probleem telkens 100 maal opgelost. Zo konden de verschillende berekende ontwerpen van de windmolenparken met elkaar vergeleken worden en werd er eveneens een statistische basis voor verdere vergelijking voorzien.

Na berekening bleek dat het 'Search Group Algoritme' aangaande de energieproductie over het algemeen efficiëntere lay-outs ontwierp in vergelijking met de twee algoritmes uit de literatuur. Uit het onderzoek naar de invloed van de instellingen van het algoritme op de resultaten werden richtlijnen voor het hanteren van het algoritme afgeleid. Uit de statistieken van de verschillende gegenereerde oplossingen kwam een zeer lage variatiecoëfficiënt naar voor. Er is dus slechts een zeer geringe dispersie tussen de verschillende oplossingen. Dit houdt in dat een relatief laag aantal iteraties volstaat om een optimale layout te berekenen wat bijgevolg leidt tot een aanzienlijk kortere berekeningstijd. Het betreft kortom een zeer robuust algoritme dat zeer geschikt is voor het oplossen van dergelijke complexe optimalisatieproblemen.

De combinatie van een mathematisch energieproductiemodel met het optimalisatiealgoritme kan ook voor tal van andere toepassingen aangewend worden. Mits ontwikkeling van een aangepast model kan dit bijvoorbeeld gebruikt worden voor de positionering van golfenergieconvertoren in een zogenaamd golfenergieconvertorenpark. Verder onderzoek kan ook de combinatie van zowel windturbines als golfenergieconvertoren optimaliseren. Er dient dan te worden nagegaan hoe de convertoren en de turbines elkaar en de golfhoogte beïnvloeden om zo de operationaliteit en de energieproductie te verhogen en de onderhoudskosten te verlagen.

Momenteel ben ik werkzaam als onderzoeker aan de Universiteit Gent in de onderzoeksgroep van professor Troch waar ik verder werk aan mijn thesisonderzoek. Er wordt een kostenfunctie geïmplementeerd om zo niet enkel de energieproductie te maximaliseren maar ook rekening te houden met de kosten van bijvoorbeeld de bekabeling. Desgelijks wordt naar een optimale balans tussen de kosten over de levensduur van het windmolenpark en een maximale energieproductie gestreefd. Daarnaast wordt er getracht een mathematisch energieproductiemodel voor golfenergieconvertoren te ontwikkelen om hiermee de optimale posities van golfenergieconvertoren

binnen een park te bepalen. Daar golfenergievectoren ook constructieve effecten op elkaar kunnen hebben kan een park van convertoren meer energie dan enkel de som van de individuele toestellen opleveren. Het is dus bijzonder belangrijk om de juiste posities te bepalen om een maximum aan constructieve effecten te verwezenlijken.

The use of echosounder tools for fish detection in the North Sea

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Geluid is een belangrijke factor in het aquatisch milieu. Veel diersoorten gebruiken geluid voor communicatie, predator-prooi interacties en het waarnemen van de omgeving. Wanneer bijvoorbeeld een dolfijn een geluidsgolf produceert zal deze zich directioneel voortbewegen in het water. Hierbij kan deze golf op een object in het water botsen waarbij een deel van deze golf zal reflecteren als een echo die opnieuw gedetecteerd kan worden door de dolfijn. Dit is echolocatie, een techniek die de mens heeft overgenomen van de natuur om objecten in de waterkolom waar te nemen die niet zichtbaar zijn met het bote oog. Alle toestellen die een detectie kunnen uitvoeren van objecten in het water door gebruik te maken van voortbeweging van geluid, worden gegroepeerd onder de term SONAR (Sound Navigation And Ranging). Een passieve sonar is een toestel dat luistert. Het detecteert geluiden die geproduceerd zijn door bepaalde objecten in de waterkolom. Een actieve sonar, daarentegen, produceert zelf geluidsgolven en detecteert de echo's die onstaan na een interactie met een object in het water. Wanneer een actieve sonar gelijktijdig akoestische geluidsgolven kan verzenden en ontvangen in een directionele richting, is dit een echosounder. Van oorsprong worden deze echosounders voornamelijk gebruikt voor bodemdetectie (bathymetrie), waarbij de echo's afkomstig van de waterkolom weggefilterd worden als ruis om zo de waardevolle bodemreflectie te behouden. Met deze data kan vervolgens veel informatie verkregen worden over de structuur van de zeebodem maar ook over objecten op de zeebodem zoals pijpleidingen, vliegtuig- en scheepswrakken. Een andere toepassing van deze technologie situeert zich binnen het meer recente bio-akoestische onderzoek dat zich meestal richt tot de visserij.

Wereldwijd staan de zeeën en oceanen onder druk door menselijke overexploitatie. Hierdoor zijn effectieve management strategiën essentieel om de biodiversiteit en abundanties van de vispopulaties te beschermen. Onderzoek naar de huidige visbestanden moet als gevolg gebaseerd zijn op accurate metingen en schattingen. Sinds het begin van het Belgisch visserijonderzoek zijn traditionele vangsmethoden, zoals het gebruik van een boomkor en pelagische trawls de voornaamste staalnamemethoden. Deze vangsmethoden zijn echter invasief en houden enkele technische beperkingen in. Zo is trawlvisserij verboden in bepaalde gebieden die kwetsbare structuren herbergen, denk maar aan windmolenparken en scheepswrakken. Vaak zijn juist deze gebieden zeer interessant voor biologisch onderzoek. Daarnaast heeft onderzoek reeds uitgewezen dat vissen een gedragswijziging (verticale migratie, herkenning en ontwijken van visnetten) vertonen als reactie op trawls. Omwille van deze tekortkomingen om accurate totale biomassa en abundantie schattingen te maken, bieden echosounders een oplossing.
Door gebruik te maken van geluidsgolven kunnen verschillende targets gedecteerd worden. Niet enkel vissen en plankton worden hiertoe gerekend maar ook koralen, onderwatervegetatie en zeebodemstructuren. Echter in deze thesis is de focus gezet op de targets die van groot belang zijn voor het marien biologisch onderzoek in het Belgische deel van de Noordzee (BPNS). Deze prioritaire groep is de vissen. Iedere structuur in de waterkolom met een bepaalde densiteit kan een reflectie van de uitgezonden geluidsgolf veroorzaken. Veel vissoorten hebben een zwemblaas om de drijfcapaciteiten te reguleren gedurende het zwemmen. Dit is een met gas gevulde blaas die voor een sterke terugkaatsing van een geluidsgolf zorgt omwille van het grote contrast in densiteit tussen het gas en het vloeibaar medium. Iedere vissoort heef een specifiek vorm van zwemblaas, waardoor iedere soort eveneens voor een verschillende reflectiesterkte zorgt.

De drie verschillende echosounder toestellen die voornamelijk gebruikt worden in het visserijonderzoek zijn single beam, split-beam en multibeam echosounders. Een single beam echosounder is een standaard instrument voor zeebodemdetectie binnen hydrografisch onderzoek. De zender verzendt een geluidsgolf in een enkele straal die directioneel gericht is. Daaropvolgend kan de ontvanger een reflectie waarnemen, die op de aanwezigheid van een bepaald object kan wijzen. Hierbij is het echter niet mogelijk om de exacte positie en richting binnen de straal te bepalen. Enkel de diepte van het gedetecteerde object kan bepaald worden. Een split-beam echosounder daarentegen heeft een ontvanger die opgedeeld is in vier kwadranten. Dit creëert de mogelijkheid om een specifieke locatie binnen de straal te bepalen. Met andere woorden laat een split-beam echosounder toe om een driedimensionele locatie van een object te bepalen, terwijl dit voor een single beam echosounder beperkt is tot een dimensie, de diepte. Een multibeam echosounder is verschillend van de voorgaande types omwille van het gelijktijdig gebruikmaken van verschillende stralen met geluidsgolven. De meeste echosounders vertonen bij een toename in meetdiepte een afname in ruimtelijke resolutie. Een

multibeam echosounder biedt een oplossing voor deze beperking omwille van het vergroten van het meetoppervlak, door gebruik te maken van meerdere stralen. Het grotere detectievolume van een multibeam echosounder leidt, met als gevolg, tot een tijdsbesparing in wetenschappelijke staalname duur, wat eveneens belangrijke economische voordelen biedt.

Multibeam echosounders worden traditioneel gebruikt in bathymetrisch onderzoek, waarvoor deze toestellen geijkt zijn. Een ijking voor waterkolomdetecties is echter amper tien jaar geleden ontwikkeld, waardoor het gebruik van multibeam echosounder voor waterkolomdetectie nog in zijn kinderschoenen staat. Deze ijking laat toe om niet enkel kwalitatieve metingen uit te voeren, maar ook kwantitatieve metingen die bijdragen tot meer inzichten omtrent de wereldwijde visbestanden.

Het gebruik van echosounder toestellen is niet enkel beperkt tot wetenschappelijke doeleinden. Wereldwijd worden echosounder toestellen gebruikt in de commerciële en recreationele visserij. In de recreationele visserij wordt meestal gebruik gemaakt van een viszoeker (fishfinders). Deze viszoekers hebben een eenvoudige opbouw en genereren een niet-kwantiatieve uitvoer, omwille van het ontbreken van een ijking. Het ontbreken van een ijking betekent dat de standaard prestaties niet kunnen gecontroleerd worden, met als gevolg dat exacte abundantie en biomassa schattingen niet kunnen uitgevoerd worden. Een ijking gebeurt meestal door een standaard target met een gekende akoestische target sterkte te gebruiken en te detecteren, waarna de verkregen detectie waarden vergeleken worden met de standaard waarden. Wetenschappelijke echosounder toestellen zijn dus steeds geijkt en bieden ook de mogelijkheid tot het opslaan van grote volumes aan verkregen data. Deze data kunnen dan nadien verwerkt worden met specifieke naverwerking software.

Niet alle zones in de waterkolom zijn detecteerbaar. Er zijn twee belangrijke zones die voor een beperking in detectie zorgen, namelijk de blinde en de dode zone. De theoretische blinde zone ligt dicht tegen het wateroppervlak. Omwille van de eigenschappen van geluidsgolven, ontstaat er steeds een 'near-field' waarin er geen lineair verband is tussen de sterkte van de geluidsgolf en de afstand tot de bron van het geluid. De dode zone, anderzijds, bevindt zich lager in de waterkolom, net boven de zeebodem. Omwille van de sferische vorm van de geluidsstraal, wordt een vlakke bodem eerst gedetecteerd in het midden van de straal. Hierbij bevinden de beide zijden van de geluidsstraal zich nog op een bepaalde hoogte boven de zeebodem. De interpretatie van objecten tussen de effectieve zeebodem en deze bepaalde hoogte is niet mogelijk en creëert vervolgens de dode zone. Echter in de praktijk is de detectie niet enkele beperkt door de theoretische blinde en dode zone. Verstoring van de waterstroom door golfwerking en bewegingen van het schip zorgen voor luchtbubbels in het water, die een sterke reflectie veroorzaken van het geluidssignaal. Deze interactie in de turbulentiezone zorgt dus voor een grotere beperking in detectiecapaciteiten. De totale blinde zone bestaat uit de theoretische blinde zone en de morgelijke turbulentiezones. In extreme condities kan deze akoestische blinde zone een diepte van 15 m bereiken. Omwille van de kleine schaal van de theoretische blinde zone (twee golflengtes) wordt de blinde zone, in het geval van turbulentie, hoofdzakelijk bepaald door de invloed van deze turbulentie. Een multibeam echosounder toestel zorgt voor een extra uitbreiding van de dode zone, omwille van de meerdere geluidsstralen. Iedere straal afzonderlijk beschrijft een theoretische dode zone. Daarbovenop hebben de meerdere stralen samen eveneens een sferische vorm, waardoor een volume ontstaat dat zich buiten het detectiegebied bevindt van de geluidsgolven. Hierdoor wordt de totale dode zone de som van de individuele theoretische dode zones en het gebied dat zich buiten het detectiegebied bevindt van de multibeam echosounder.

Voor deze studie zijn waterkolom detecties gebruikt die verkregen zijn met behulp van een Kongsberg EM2040 multibeam echosounder, gemonteerd op de RV Simon Stevin. Sinds 2012 zijn in totaal 598 waterkolom opnames verzameld gedurende 15 staalnames, goed voor 662 GB aan waterkolom data. Deze bestanden zijn vervolgens verwerkt door gebruik te maken van verschillende nawerking softwarepakketten (BioSonics Visual Acquisition, QPS Fledermaus en Echoview). Hierbij is besloten dat Echoview het meest flexibel en uitgebreide softwarepakket is om een naverwerking uit te voeren van waterkolomopnames afkomstig van verschillende echosounder toestelllen.

De waterkolom databestanden hebben een grote verscheidenheid aan detectieaantallen aangetoond, variërend van 165 tot 13555 gedetecteerde targets per 1000 m³ watervolume. Beide staalnames vertonen echter een groot verschil in gemeten watervolume, waarbij respectievelijk 507230 en 1219 m³ water gemeten zijn. In de total dataset vertonen ook de andere variabelen, zoals de totale afgelegde afstand en diepte van de waterkolom, tevens een grote variabiliteit. Het feit dat alle staalnames, met uitzondering van een, geen naverwerking voor visdetectie als doel hadden, kan hiervoor als verklaring dienen. De vergelijking in detectiecapaciteit tussen de twee verschillende echosounder toestellen (BioSonics DT-X split-beam echosounder en Kongsberg EM2040 multibeam echosounder) heeft aangetoond dat er een verschil is in de hoeveelheid targets die gedetecteerd zijn per volume eenheid.

De Kongsberg EM2040 multibeam echosounder detecteert met een gemiddelde van 1052 targets per 1000 m³ watervolume minder targets dan de BioSonics DT-X split-beam echosounder die 2464 targets per 1000 m³ watervolume detecteert. Na een correctie voor bijkomende residuele error in de data, is dit verschil echter niet significant toe te schrijven aan het verschil in echosounder toestel.

Turbulentie is geacht een grote invloed te hebben op de detectiecapaciteiten in het BPNS. Dit is bevestigd door deze studie, waarbij in 9 van de 15 staalnames meer dan 50 % van de totale hoeveelheid gedecteerde targets zich in de turbulentiezone bevinden. Hierdoor is het niet mogelijk te bepalen of deze targets verwijzen naar een echt object in de waterkolom of een storing door luchtbubbels. Er is geen significant verschil aangetoond tussen de hoeveelheid gedecteerde targets per 1000 m³ watervolume binnen en buiten de turbulentiezone. Dit betekent dat turbulentie, veroorzaakt door bewegingen van het schip en golfwerking, luchtbubbels in de waterkolom brengt die voor een vergelijkbare hoeveelheid gedecteerde targets zorgen als de gewenste objecten (vissen) zelf. In de kustwateren van het BPNS is turbulentie een zeer belangrijke bron van ruis, die de akoestische blinde en dode zone van de waterkolom sterk uitbreiden. In extreme omstandigheden kan, omwille van de geringe diepte op sommige plaatsen, de gehele waterkolom verstoord zijn door turbulentie. Hierdoor is het gebruik van akoestiek beperkt in zijn toepasbaarheid. Desondanks kunnen in goede weersomstandigheden, zuivere waterkolom opnames bekomen worden waarbij de detectie van vis targets mogelijk is.

Deze studie heeft tevens een theoretische tekortkoming blootgelegd in het gebruik van target signaalsterkte voor het berekenen van target lengte. Hierbij wordt de assumptie gemaakt dat de gemeten akoestische signaalsterkte de volledige akoestische reflectie van het individu beschrijft. Vissen, bijvoorbeeld, zwemmen vrij in alle richtingen in de waterkolom. Een positieve of negatieve tilt van het individu zorgt voor een wijziging in oppervlak dat beschikbaar is voor volledige terugkaatsing van de inkomende geluidsgolf. Het is aangetoond dat schepen een verticale migratie veroorzaken van vissen, waarbij hun reflectief oppervlak gewijzigd kan zijn gedurende de meting vergeleken met normale omstandigheden, in afwezigheid van verstorende elementen.

Tot slot heeft de vergelijkende studie tussen drie potentiële echosounder toestellen voor visserijonderzoek in het BPNS, de SeaPix 3D sonar als meestbelovende echosounder toestel bevonden. De combinatie van meerdere stabiliteitssystemen die de resolutie positief beïnvloeden, de vereenvoudigde in situ ijking en de mogelijkheid om gelijktijdig meerdere rijen van geluidsstralen uit te zenden die een 3D visualisatie mogelijk maken, geven voordelen die niet kunnen geëvenaard worden door de andere echosounder toestellen (Kongsberg EM 2040 multibeam echosounder en BioSonics DT-X split-beam echosounder). Deze beslissing is echter op een theoretische basis, gezien een in situ demonstratie en daaropvolgende naverwerking van de waterkolomdata niet binnen deze studie zijn opgenomen. De Kongsberg EM2040 multibeam echosounder, die momenteel gemonteerd is op de RV Simon Stevin en gebruikt wordt voor het mariene biologische onderzoek in het BPNS, heeft ondanks het ontbreken van een ijking voor waterkolom opnames, potentieel getoond in het vergaren van kwaliteitsvolle waterkolomdata. Deze data zijn voor interpretatie vatbaar met naverwerkings-software pakketten Echoview en Fledermaus (FMMidwater).

Een finale conclusie stelt dat het gebruik van de huidige beschikbare geijkte multibeam echosounder toestellen, ontwikkeld voor visserijonderzoek, de beste keuze is voor visserijonderzoek in het BPNS. Geringe abundanties van verscheidene vissoorten en een turbulent watervolume vereisen een toename in het te detecteren watervolume, met een behoud in ruimtelijke resolutie zodat op een accurate manier targets van verschillende grootteklassen gedetecteerd en onderscheiden kunnen worden.

Tracking a marine predator to design flexible boundaries of marine protected areas for an ecosystem approach to fisheries

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Werelwijd neemt het aantal zeevogels af (Paleczny et al. 2015), doordat de combinatie klimaatopwarming/overbevissing een temporele en ruimtelijke mismatch veroorzaakt tussen zeevogels en hun prooi (Brander 2007; Grémillet & Boulinier 2009). Zeevogels vervullen als mariene predator een belangrijke rol in het voedselweb van de oceaan. Hierdoor kan deze globale afname verreikende gevolgen hebben in de structuur en dynamiek van volledige marine ecosystemen (Estes et al. 2011). Het is daarom van cruciaal belang om beschermende maatregelen voor zeevogels uit te denken en in de praktijk om te zetten. Het voorgaande is zeker van toepassing op Afrikaanse pinguïns *Spheniscus demersus*, een zeevogelsoort waarvan de aantallen drastisch afgenomen zijn het afgelopen decennium (Crawford et al. 2011). Om deze reden verkregen zij in 2010 de status 'bedreigd' van de *International Union Of Conservation*. Afrikaanse pinguïns hebben vele bedreigingen. De belangrijkste zijn de klimaatverandering en competitie met de ringzegen-visserij, een visserij die de belangrijkste prooien van Afrikaanse pinguïns als doelgroep heeft, namelijk ansjovis *Engraulis encrasicolus* en sardine *Sardinops sagax* (Crawford et al. 2006). Afrikaanse pinguïns hebben een economische waarde via ecotoerisme (Lewis et al. 2012), maar de ringzegen-visserij heeft ook een belangrijke lokale economische waarde (Food and Agriculture Organization of the United Nations or FAO 2016), waardoor deze niet zomaar kan stopgezet worden.

Afrikaanse pinguïns komen alleen voor in zuidelijk Afrika, en deze studie bekijkt hun meest oostelijke broedlocatie, Algoa Bay. Deze baai is omringd door een drukke stad met een actieve haven. Er zijn twee eilandgroepen in de baai: diegene waartoe het eiland St. Croix behoort, dat 50% van de globale Afrikaanse pinguïn populatie telt (Crawford et al. 2011), en diegene rond het eiland Bird. Om de populaties terug te doen toenemen is een ecosysteembenadering tot visserijen (EAF) nodig. EAF omvat immers de behoeften van het volledige ecosysteem, wat impliceert dat het ook rekening houdt met de effecten van de visserij op soorten die geen doelgroep zijn (Cochrane 2004; Garcia & Cochrane 2005). Daarbovenop wordt er in deze systematiek ook rekening gehouden met de socio- economische belangen van de kustgemeenschap. In deze optiek werd een experiment gestart in 2009, waarbij de activiteiten van de ringzegen-visserij verboden werden in een radius van 20 km, alternered om de drie jaar rond elk eiland. Dit bleek echter onvoldoende om de pinguïn populaties terug te laten toenemen. Om die reden heeft de Zuid-Afrikaanse regering nu een voorstel klaar om het huidige zeereservaat in Algoa Bay te vergroten. Zeereservaten (MPAs) werden in het verleden reeds erkend als handige hulpmiddelen in een EAF (Roberts et al. 2001; Pauly et al. 2002). Wanneer ze ontworpen worden met flexibele grenzen kunnen ze een voordeel opleveren voor zowel pinguïns als visserijen. In tijden van klimaatverandering is dergelijk adaptief management een vereiste om ecosystemen efficiënt te beschermen (Hyrenbach et al. 2000). Bovendien is constante monitoring noodzakelijk om dynamisch te kunnen inspelen op veranderingen (Maxwell et al. 2015). Het MPA-voorstel van de Zuid-Afrikaanse regering heeft verschillende zones maar zonder flexibele grenzen. Er zijn bovendien geen maatregelen voorzien om in te spelen op veranderende omstandigheden. De zones waar de ringzegen-visserij niet mag vissen zijn beperkt tot twee kleine rechthoekige gebieden rond de St. Croix en Bird eilanden, die kleiner zijn dan de reeds aanwezige experimentele sluitingen. Deze masterproef brengt het effect van de voorbije experimentele sluitingen in kaart en onderzoekt het nut van de geplande MPA. Voor de uitvoering ervan kon er gebruik gemaakt worden van bestaande GPS-data van 2008 tot 2015.

In de ruimtelijke analyse werden de GPS-locaties van de forageertrips van de pinguïns gesommeerd per jaar en per kolonie. Vervolgens werden deze geconverteerd in minimum convex polygonen (MCP100%), die een schatting geven van het totale jaarlijkse forageergebied (Jennrich & Turner 1969). MCP100% kan gebruikt worden om de grenzen van een MPA te bepalen, omdat het het totale ruimtelijke gebruik van de pinguïns toont. Als laatste werd dit alles overlapt met de jaarlijkse visvangst per rastercel (10 zeemijlen x 10 zeemijlen). Deze ruimtelijke analyse toonde een sterke overlap tussen de ringzegen-visserij en het forageergebied van de pinguïns. Een afname in visvangst resulteerde echter niet in kleinere forageergebieden. Daar staat tegenover dat de voor visvangst gesloten zones van de nieuwe MPA nog niet de helft van het forageergebied van de pinguïns omvatten, en bovendien gelegen zijn in gebieden waar er sowieso minder gevist wordt.

In het tweede deel van deze studie werd de GPS-data gebruikt om informatie over foerageergedrag aan het wateroppervlak te bestuderen. Door te kijken naar de bezochte GPS-locaties kon de duur van de trip, de horizontale afgelegde weg, en de maximale afstand tot de kolonie bepaald worden. Voor een aantal jaren was er ook data van tijd-diepte registreerapparatuur beschikbaar. Deze gaven informatie over duikgedrag zoals de totale verticale afgelegde afstand, de gemiddelde duiktijd en – diepte, en de duikfrequentie. Verschillende verklarende variabelen waren gekend die deze foerageerkenmerken konden beïnvloeden, onder andere kolonie, jaar, visserij sluitingen, en visvangst. Het geslacht van de ouders en de totale massa van de kuikens werden aan deze lijst toegevoegd wanneer ze een wezenlijk verschil uitmaakten. Alle foerageerkenmerken werden gemodelleerd met alle mogelijke combinaties van deze verklarende variabelen, maar enkel relevante biologische interacties werden meegenomen, met name tussen kolonie, jaar en visserij sluitingen. Hiervoor werden lineaire modellen gebruikt, of wanneer er heterogeniteit aanwezig was, de gegeneraliseerde kleinste kwadraten (Zuur et al. 2009). De statistische resultaten toonden dat pinguïns aanzienlijk minder inspanningen leverden wanneer de ringzegen-visserij gesloten was rond hun kolonie. Tijdens deze periodes foerageerden de pinguïns minder lang en minder ver, en vertoonden ze een lagere duik frequentie en minder verticale afgelegde afstand. Pinguïns van de St. Croix kolonie hadden bovendien hogere waarden voor alle foerageerkenmerken. Dit laatste kan verklaard worden door het feit dat er meer gevist wordt rond dit eiland en doordat de kolonie dichter bij het vasteland ligt, en dus minder pelagische ruimte heeft rondom. Echter, de belangrijkste reden is waarschijnlijk het feit dat het de grootste kolonie is en hierdoor meer intraspecifieke competitie vertoont. Dit laatste is zichtbaar in Ashmole's halo, een zone van voedseldepletie die grote zeevogelkolonies omringt (Ainley et al. 2006; Gaston et al. 2007).

Eerdere conclusies over de invloed van de grootte van de kolonie op dat foerageergedrag werden hiermee bevestigd (Lewis et al. 2001). De gedetecteerde jaarlijkse fluctuaties vertoonden geen correlatie met andere onderzochte variabelen, en werden waarschijnlijk beïnvloed door omgevingsvariabelen. Van dergelijke omgevingsvariabelen werd in het verleden reeds aangetoond dat ze het foerageergedrag van pinguïns kunnen beïnvloeden (van Eeden et al. 2016). Visvangst toonde geen algemeen effect op het foerageergedrag. Het feit dat de totale visvangst per jaar aanzienlijk is afgenomen in deze regio, tot zelfs geen visvangst in 2015, verklaart de ontkoppeling tussen foerageergedrag en visvangst. Waarschijnlijk is de afname in visvangst een logisch gevolg van de achteruitgang van de aantallen kleine pelagische vis. Dit laatste werd gedetecteerd met behulp van dieetstalen uit de magen van pinguïns die terugkeerden van foerageertrips, en van het in kaart brengen van de aanwezige biomassa van de vis met behulp van recreationele viszoekers (RFFs) (McInnes et al. 2015). Dit verklaart tevens waarom de pinguïn populaties tot hiertoe nog niet zijn toegenomen. Het toont ook aan dat in toekomstige studies, biomassa een betere variabele is dan visvangst.

Met behulp van de resultaten uit deze studie werd een voorstel uitgewerkt dat de noodzakelijke bescherming van de Afrikaanse pinguïn combineert met de socio-economische noden van de kustgemeenschap in Algoa Bay. Om onmiddellijk te kunnen starten met adaptief management kan de regel van Cury et al. (2011) toegepast worden, die stelt dat de biomassa van de prooi minstens één derde van de totale biomassa moet bedragen om de productiviteit van de zeevogels veilig te stellen. Wanneer dit niet het geval is, dienen de afgesloten zones minstens de grootte te hebben van de experimentele sluitingen. Wanneer er niet voldoende vis aanwezig is, is het sowieso economisch niet interessant om in deze gebieden te investeren. Alle visserij activiteiten stopzetten voor een bepaalde periode bevordert bovendien de heropleving van het visbestand, wat een voordeel oplevert voor vissers in de toekomst. Wanneer de biomassa opnieuw voldoende hoog is, kunnen deze gebieden verkleind worden, eventueel op verschillende schaal rond beide kolonies. Met deze maatregel worden de grenzen van de MPA flexibel, wat tevens optimaal is voor de kustgemeenschap. Daarbovenop is extra flexibiliteit mogelijk door te kijken naar het type van visserij. Aangezien Afrikaanse pinguïns alleen in competitie zijn met ringzegen-visserijen, kunnen andere visserijen wel toegelaten worden in een bufferzone rond de gesloten zones. Dynamisch management is momenteel mogelijk door het in kaart brengen van de biomassa met behulp van de lopende maandelijkse RFF-inspecties. Wanneer de biomassa beoordeelt wordt aan de hand van deze metingen, kunnen de grenzen van de MPA aangepast worden aan de actuele situatie. In de toekomst zijn echter meer kwantitatieve beoordelingen en evaluaties noodzakelijk om de criteria te verfijnen, aanpassingen die kaderen in adaptief management (Agardy et al. 2003).

Samenvattend kunnen we stellen dat het MPA-voorstel van de Zuid-Afrikaanse regering niet voldoende is om de afname van Afrikaanse pinguïn populaties tegen te gaan. De MPA dient ontworpen te worden met grotere, maar flexibele grenzen voor de afgesloten zones. Wanneer bovendien de biomassa continu in kaart gebracht wordt, kan er dynamisch ingespeeld worden op de omgevingsfactoren en tegemoet gekomen worden aan de socio-economische noden van de kustgemeenschap in Algoa Bay.

Habitat suitability modelling for the harbour porpoise (*Phocoena phocoena*) in the Belgian part of the North Sea

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De bruinvis (*Phocoena phocoena*) is de meest algemene walvisachtige in de Noordzee en in het bijzonder in het Belgische deel van de Noordzee (BDNZ). Recent is een zuidwaartse verschuiving waargenomen in het verspreidingsgebied van bruinvissen, waarschijnlijk door een verandering in de voedselbeschikbaarheid. Op dit moment ondergaan zeezoogdieren vele menselijke bedreigingen: bijvangst, overbevissing, vervuiling, geluidsoverlast... Daardoor is beheer noodzakelijk voor het behoud van de bruinvis. Voor een grondig beleid is voldoende kennis van de ecologie van deze soort nodig. Er is echter weinig geweten over de habitatsvoorkeuren van bruinvissen. Het doel van de huidige studie was het identificeren van geschikte habitats voor bruinvissen en bepalen welke omgevingsvariabelen het meest relevant zijn voor de verspreiding van deze dieren. Om deze doelstellingen te bereiken, werden waarnemingen van bruinvissen verkregen via luchtsurveys in het BDNZ tijdens de periode 2008-2014. Zeven factoren werden geselecteerd om de habitatgeschiktheid voor bruinvissen te bepalen, gebaseerd op eerdere studies: diepte, Bathymetric Position Index (BPI), macrobenthische gemeenschappen, afstand tot scheepvaartsroutes, Suspended Particulate Matter (SPM), afstand tot offshore windmolens en oceaanstromingen (twee snelheidsvariabelen in de x en y richting). Om eventuele distributiepatronen aan te tonen, werden eendimensionale relaties tussen de veranderlijke variabele, i.e. de observaties van bruinvissen, en elke omgevingsvariabele afzonderlijk geëvalueerd. Verder werd de software MaxEnt (Maximum Entropy Species Distribution Modelling) gebruikt om de belangrijke factoren te identificeren. MaxEnt levert een efficiënt instrument op in verspreidingsmodellering van soorten, omdat enkel aanwezigheidsdata nodig zijn. De drie belangrijkste omgevingsvariabelen waren oceaanstromingen, afstand tot offshore windmolens en diepte. Volgens de modellering met MaxEnt waren kalme en diepe wateren op grote afstand van offshore windmolens de meest geschikte habitats voor bruinvissen in het BDNZ. Hoewel de andere factoren minder bijdroegen aan het model, werd er toch een duidelijk verspreidingspatroon gevonden. Echter, dit onderzoek toonde enkel correlaties tussen het voorkomen van bruinvissen en verschillende omgevingsvariabelen; oorzakelijke verbanden konden niet worden afgeleid. Toekomstige studies zijn noodzakelijk om de oorzaken van het verspreidingspatroon van bruinvissen te ontrafelen.

Groundwater flow in freshwater tidal marshes: A comparison of a natural and a restored marsh

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Numerous tidal marsh areas are being restored for flood protection and ecological development along estuaries. Recently, however, questions arise about the extent to which restored marshes deliver ecosystem services. Due to the historical land use, soil properties and associated subsurface water flow might be altered in restored marshes, hereby affecting important ecosystem functions. In this study we mapped the physical movement of water in the soil of a natural and a restored freshwater tidal marsh for the first time. Furthermore, this research served as a pilot study to evaluate different methods to assess estuarine subsurface hydrology.

A newly developed method for *in situ* groundwater flux measurements (integrated passive flux sampler) was used in combination with measurements of soil characteristics and groundwater level time series along a transect. Special attention was paid to the presence of organic matter and macro pores (using CT-scanning) in the soil, and their effect on subsurface groundwater flow.

Our results indicate that the soil in the restored marsh consists of a layer of freshly accreted sediment, characterized by a large organic matter content and macroporosity, and an underlying layer of compacted agricultural polder soil with a low organic matter content and few macropores. Both layers differ significantly from each other and from the soil in the natural marsh. Furthermore, the underlying polder soil in the restored marsh was found to put constraints on the amount of pore-water that is exported to the creek in between tidal events.

As a result, fluxes of nutrients, the source-sink function and the development of vegetation in the restored marsh might be negatively affected. For new marsh restoration projects, we therefore suggest to explore the possibility to mix the agricultural polder soil with organic matter to induce the formation of macropores, which will ultimately lead to an improved ecosystem service delivery.

Keywords: Scheldt estuary; marsh restoration; pore-water; groundwater flux; macroporosity

Reconstructie van de aardbevingsgeschiedenis van Aysén fjord (Zuid-Chili) aan de hand van seismische stratigrafie en de analyse van een sedimentkern

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Chili is een van de seismisch meest actieve zones ter wereld en wordt op regelmatige basis getroffen door zeer zware aardbevingen. Deze zijn niet alleen het gevolg van de nabij gelegen subductiezone, maar worden ook gegenereerd door de Liquiñe-Ofqui Fault Zone (LOFZ). Deze laatste is een actieve, dextrale horizontaalverschuivingsbeuk ('strike-slip') die het noordelijk gedeelte van het Chileense fjordland doorkruist (Cembrano et al., 1996). In 2007 werd deze breukzone geteisterd door een aardbevingszwerm, met het epicentrum van de zwaarste schok (21 april, Mw 6.2) gelegen in Aysén fjord (Legrand et al., 2011). De intensiteit van de seismische schokken was zo hoog dat tientallen landverschuivingen en andere massabewegingen plaatsvonden en uitmondden in het fjordbekken (Naranjo et al., 2009). Deze vervormden de aanwezige sedimenten op de fjordbodem en lieten op deze manier een afdruk na die duizenden jaren later bewaard blijft in de vorm verormde bodemsedimenten, al dan niet geassocieerd met (mega)turbidieten. Dergelijke afzettingen (algemeen Mass Transport Deposits genoemd, MTDs) kunnen op eender welk ogenblik in de geologische tijd worden gegenereerd en kunnen worden teruggevonden met behulp van onder andere seismische profielen en sedimentkernen. Op deze manier zijn ze ideaal voor paleoseismische reconstructies.

De aardbevingsgeschiedenis van Aysén fjord werd gereconstrueerd aan de hand van een dicht netwerk van hoge resolutie seismische profielen van zowel TOPAS, sparker als airgun seismische bronnen. Deze werden bekomen tijdens twee campagnes: een eerste in 2009, uitgevoerd door het Renard Centre of Marine Geology (RCMG, Universiteit Gent) en een tweede tijdens de DETSUFA cruise in 2013 onder leiding van de Universitat de Barcelona. Aan de hand van deze profielen werden verschillende MTDs teruggevonden in de sedimentaire opvulling van de fjord en kon worden geëvalueerd of deze al dan niet veroorzaakt werden door seismische activiteit. Bijkomstig werd ook gebruik gemaakt van een beschikbare sedimentkern van ongeveer 20 meter lang, genomen in 2007 door R/V Marion Dufresne. Deze kern bevat verschillende tefras en turbidieten die correlatie met de seismische data toelieten. Het organisch materiaal dat aanwezig is in de sedimenten kon worden gedateerd met behulp van koolstofdatering en op die manier bijdragen tot het opstellen van een ouderdomsmodel voor de kern. De link tussen de seismische profielen en de sedimentkern kon dan uiteindelijk leiden tot een aantal gedateerde aardbevingen die Aysén fjord gedurende de geologische tijd getroffen hebben. Om meer informatie te krijgen over de locatie én sterkte van deze aardbevingen werden grondbewegingen gemodelleerd. Hierbij werd rekening gehouden met het feit dat een bepaalde intensiteit van grondbewegingen vereist is om een massabeweging te kunnen veroorzaken.

Na analyse van de seismische profielen zijn elf stratigrafische niveaus ontdekt waarop MTDs terug te vinden zijn (Figuur 1). Deze zijn te herkennen door hun chaotisch of transparant facies en een geometrie die veel verschillende vormen kan aannemen. Ze zijn meestal ook geconcentreerd langsheen de randen van het bekken waarin ze zijn afgezet (Moernaut et al., 2007). Eén van deze stratigrafische niveaus waarop afzettingen van massabewegingen te vinden zijn, is het oppervlakteniveau. Dit is het gevolg van de vervorming die de hoofschok van de aardbevingszwerm uit 2007 veroorzaakt heeft. Dieper in de invulling van de fjord worden gelijkaardige structuren teruggevonden op locaties verspreid doorheen de hele fjord. Het feit dat op hetzelfde stratigrafische niveau op verschillende locaties in de fjord dergelijke structuren kunnen worden teruggevonden, wijst erop dat deze vermoedelijk veroorzaakt geweest zijn door aardbevingen.
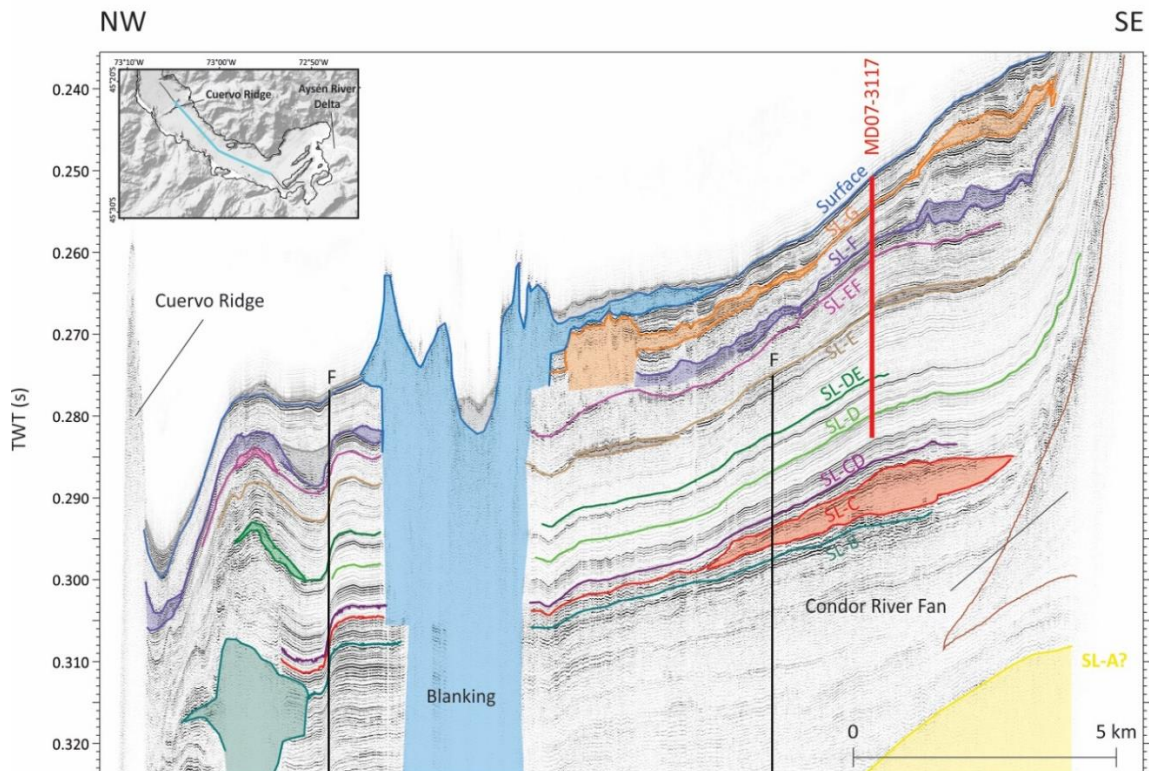
Figuur 1: Overzicht van de verschillende stratigrafische niveaus waarop MTDs werden teruggevonden, aangeduid op één van de TOPAS seismische profielen (uitgedrukt in two-way-travel time, TWT). De MTDs zelf zijn aangeduid met gekleurde vlakken. Wanneer de basis niet geobserveerd werd ('blanking') is de afzetting niet afgebakend. De locatie van de sedimentkern is weergegeven (rood, MD07-3117), alsook enkele breukstructuren (zwart, F).

Aangezien de seismische profielen aantonen dat er verschillende niveaus zijn in de fjord waarin MTDs kunnen worden teruggevonden, moeten deze ook kunnen worden teruggevonden in de sedimentkern. De meest voorkomende expressie van dergelijke afzettingen is door de aanwezigheid van een turbidiet, gekenmerkt door een piek in densiteit en magnetische susceptibiliteit (St-Onge et al., 2012), alsook door een visueel zichtbare verstoring van de gewone sedimentatie in de fjord door bijvoorbeeld een abrupte kleur en/of textuurverandering. De kern bevat zeer veel van dergelijke laagjes en verder onderzoek met behulp van smear slides kon uitwijzen of het om turbidieten of tefra-laagjes ging. Het voorkomen van tefra in de fjord is niet geheel onverwacht aangezien de omgeving van Aysén fjord gekenmerkt wordt door de aanwezigheid van verschillende vulkanen. Vulkanisch materiaal kan dan op twee manieren in de fjord terecht komen. Enerzijds kan tefra terechtkomen in het drainagegebied van één van de voedende rivieren van de fjord, en wordt het op die manier getransporteerd naar het bekken. Anderzijds is voor sommige historische erupties geweten dat Aysén fjord in het verspreidingsgebied van tefraneerslag gelegen is. Zo kan tefra dus ook rechtstreeks in de fjord worden afgezet (Naranjo and Stern, 1998, 2004).

Een van de belangrijkste elementen bij een paleoseismische reconstructie is het bepalen van ouderdommen. Voor dit onderzoek werd hiervoor gebruik gemaakt van de sedimentkern waarin voldoende organisch materiaal aanwezig was voor koolstofdatering. Om het opgestelde ouderdomsmodel te kunnen interpreteren in termen van aardbevingsgeschiedenis was het cruciaal om de kern te correleren met de seismische profielen. Dit kon worden bekomen rekening houdend met het feit dat de zichtbare reflectoren op de profielen een gevolg zijn van veranderingen in akoestische impedantie en dus van densiteit en/of de geluidssnelheid. Dit leidde uiteindelijk tot een robuuste correlatie en een duidelijke link tussen de aanwezige turbidietlaagjes in de kern en de stratigrafische niveaus met MTDs terug te vinden in de seismische profielen (Figuur 2).
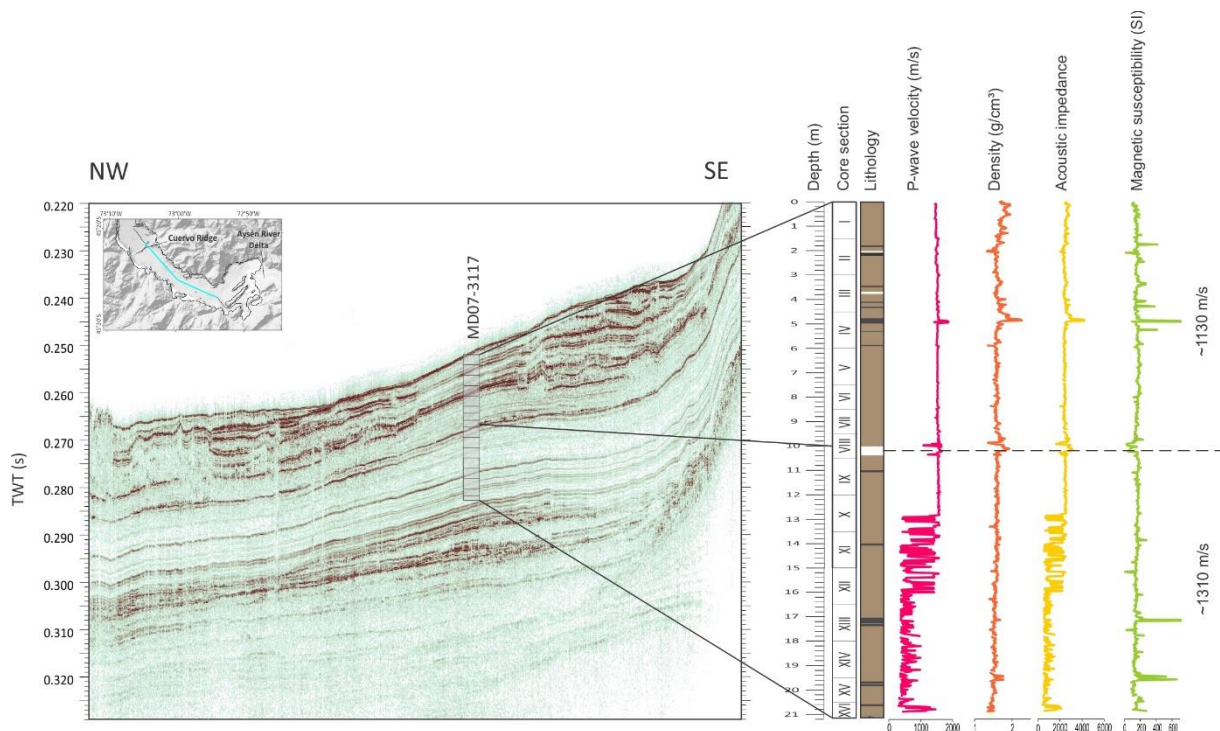
Figuur 2: Correlatie tussen één van de seismische profielen en de sedimentkern. Lithologie van de kern is schematisch weergegeven samen met de curves voor de snelheid van P-golven (roze), dichtheid (oranje), akoestische impedantie (geel) en magnetische susceptibiliteit (groen).

Dankzij de correlatie tussen de kern en de seismiek kon elk van de stratigrafische niveaus gerelateerd worden aan een *event horizon* uit de kern. Bijkomend konden de verschillende tefra-laagjes in de kern met behulp van geochemische analyse gelinkt worden aan gekende prehistorische erupties, wat leidde tot een extra controle op het ouderdomsmodel opgesteld met behulp van de koolstofdateringen (Figuur 3). Uit de correlatie bleek dat één van de seismische niveaus met MTDs gerelateerd was aan een vulkanische eruptie eerder dan aan een aardbeving. Anderzijds bleek één van de turbidietlagen uit de kern niet overeen te komen met één van de stratigrafische niveaus, maar werd op basis van de sterke gelijkenissen tussen dit laagje en de overige turbidietlagen verondersteld dat ook deze turbidiet seismisch getriggerd werd. Na applicatie van het ouderdomsmodel op de kern kon worden afgeleid dat Aysén fjord gedurende het Holoceen zeker acht keer getroffen werd door een zware aardbeving en vermoedelijk nog minstens driemaal in de tijdsperiode tussen deglaciatie van de fjord en het begin van het Holoceen.

Om een beter idee te krijgen over de mogelijke sterkte en locatie van deze MTD-genererende aardbevingen werd gebruik gemaakt van grondbewegingsmodellering (o.a. Bakun en Wentworth, 1997 en Barrientos, 1980). Dit toonde aan dat de epicentra van dergelijke aardbevingen nooit verder dan ongeveer 100 km van de fjord verwijderd kunnen zijn aangezien ze dan niet meer in staat zijn om grondbewegingen te genereren die voldoende intensiteit hebben om een massabeweging te veroorzaken. De uitzondering hierop zijn subductie-aardbevingen; deze vinden plaats op meer dan 100 km van de fjord maar vereisen oppervlaktemagnitudes van 8 of hoger om massabewegingen in het bekken te veroorzaken. Zulke magnitudes worden echter slechts zelden bereikt. Dit liet toe om zones af te bakenen rondom de fjord waarin de epicentra mogelijk gesitueerd waren, rekening houdend met de verdeling van de MTDs in het fjordbekken. Hierbij geldt dat hoe verder het epicentrum verwijderd is van de fjord, hoe hoger de magnitude moet zijn om nog een gelijkaardig patroon aan MTDs te genereren als een aardbeving van lagere magnitude die dichter bij de fjord gelokaliseerd is. Binnen deze zones is de locatie van de LOFZ en zijn vertakkingen bekend. Aan de hand van deze locaties werden dan veronderstellingen gemaakt over welke breuken verantwoordelijk konden zijn voor welke aardbevingen en dus de geobserveerde MTDs in de fjord veroorzaakt hebben. Hieruit kon worden geconcludeerd dat slechts één van de prehistorische events mogelijks gerelateerd was aan subductie, terwijl de overige negen gerelateerd zijn aan de LOFZ (Figuur 4).
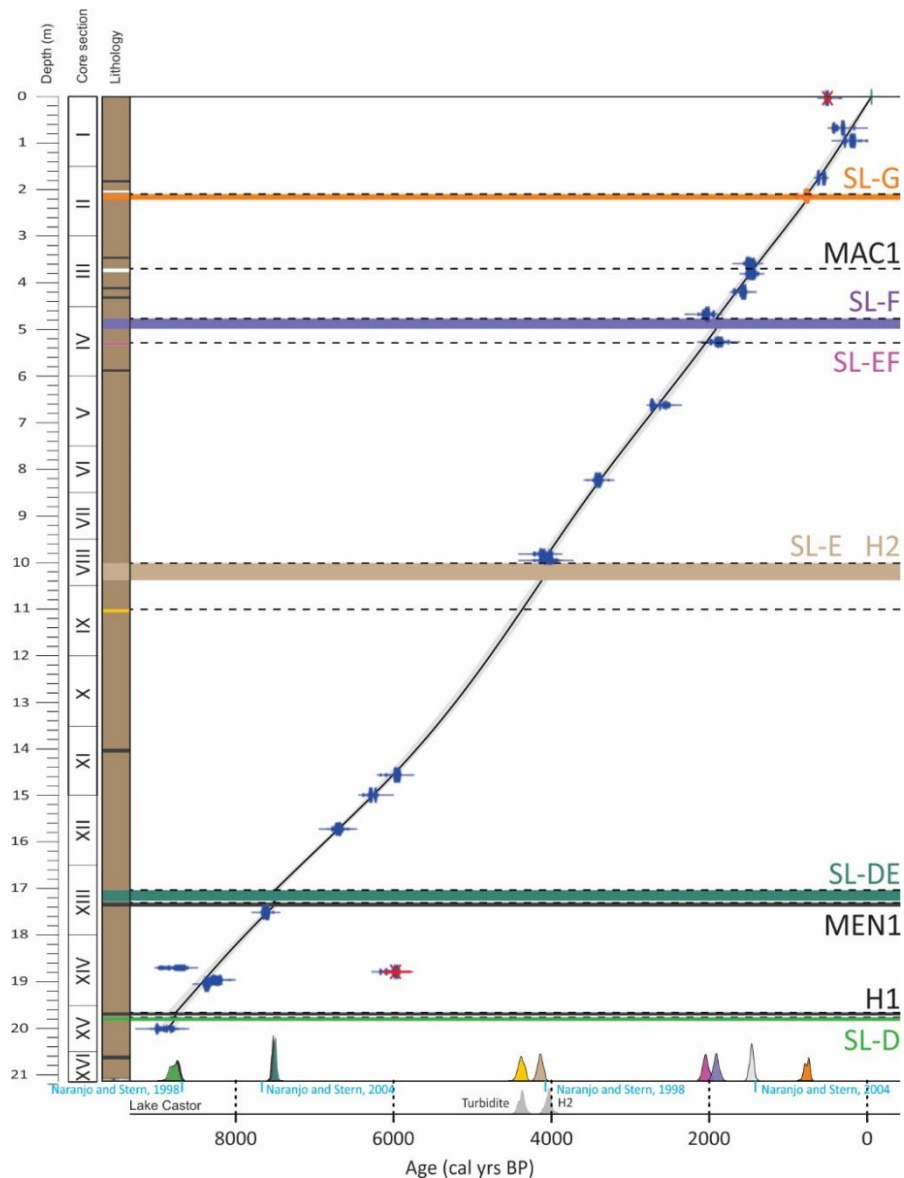
Figuur 3: De verschillende secties in de kern, hun lithologie en het ouderdomsmodel met daarop de verschillende stratigrafische niveaus aangeduid die werden teruggevonden in de seismische profielen (zie Figuur 1) en de tefra-laagjes die konden worden gelinkt aan een historische eruptie. Ouderdomsdistributies voor al deze niveaus zijn onderaan weergegeven, in combinatie met de gekende ouderdom uit de literatuur van enkele historische vulkanische erupties en de turbidiet teruggevonden in een nabij gelegen meer (S. Bertrand, persoonlijke communicatie).

Algemeen gesteld werd het gebied rondom Aysén fjord tijdens het Holoceen ongeveer één keer per 2000 jaar getroffen door een zware aardbeving gerelateerd aan de LOFZ, hoewel zeer grote afwijkingen van dit gemiddelde interval optreden. Dit is echter een relatief lage waarde gezien de grote spanning die opgebouwd wordt in de regio door subductie. Dit kan worden verklaard door de beperkte regio waarin aardbevingen in staat zijn om voldoende grondbewegingen te veroorzaken om een spoor na te laten in de fjordsedimenten. Aardbevingen die plaatsvinden op meer dan ongeveer 100 km afstand van de fjord zullen namelijk nooit kunnen worden teruggevonden in de sedimenten.

Dit onderzoek heeft nogmaals aangetoond dat fjorden een ideale setting vormen om paleoseismisch onderzoek te verrichten en waarom het belangrijk is om (hoge resolutie) seismische data te combineren met een sedimentkern om optimale resultaten te bekomen. Zonder sedimentkern is het namelijk onmogelijk om na te gaan wat de oorsprong van een MTD is of wat zijn ouderdom is. Anderzijds, een sedimentkern alleen kan de distributie van de verschillende MTDs doorheen een bekken niet visualiseren en er kan dus niet worden nagegaan of deze MTDs al dan niet seismisch getriggerd werden. De combinatie van beide methoden in dit onderzoek heeft ertoe geleid dat een gedetailleerde seismische historiek van Aysén fjord succesvol kon worden opgesteld.
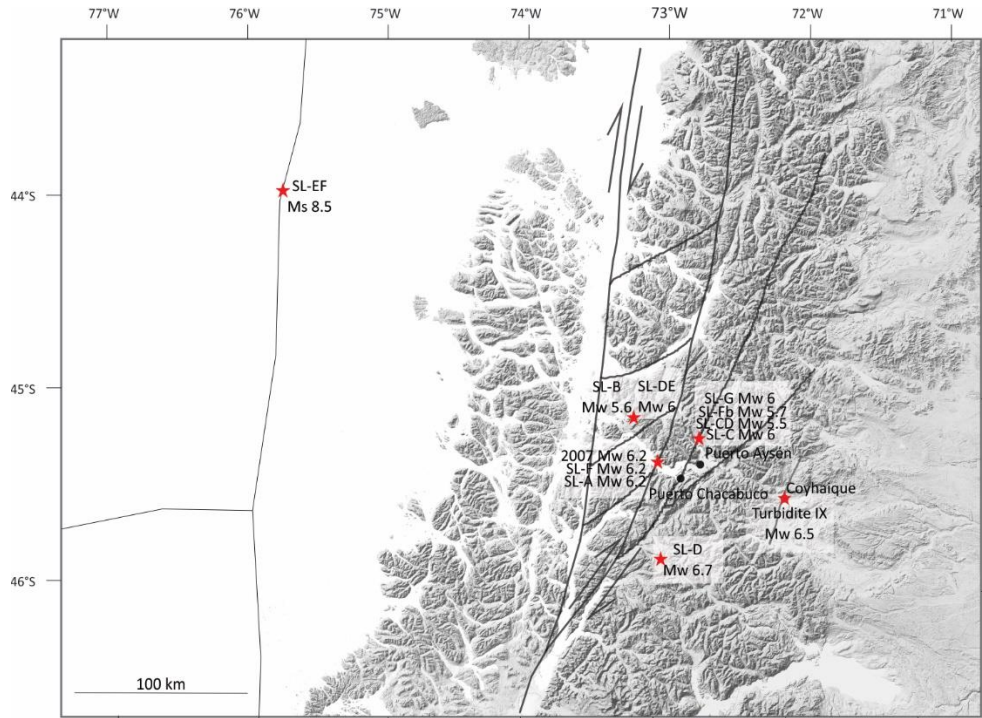
Figuur 4: Mogelijke epicentrumlocaties (rode sterren) van de verschillende aardbevingen die werden teruggevonden in de fjord met aanduidingen van de geschatte magnitude (Mw voor momentmagnitude, Ms voor oppervlaktemagnitude).

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VLIZ NORTH SEA AWARD

On the effects of high intensity impulsive sound on young European sea bass *Dicentrarchus labrax,* with special attention to pile driving during offshore wind farm construction

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More than 25 years ago, the link was established between anthropogenic sound and negative effects on marine mammals. Since then, marine mammals have dominated the bioacoustics research, but recently the focus has widened to fish, and to a lesser extent, also invertebrates. The frequency range of anthropogenic sound often overlaps with the hearing range of the fish. Consequently, underwater sound has the potential to cause auditory injuries, physiological stress, and behavioural disturbance and to mask biological relevant sounds. In addition, sound pressure can influence the swim bladder volume which can result in (mortal) internal injuries. So, depending on the characteristics of the sound and the fish species, the impact of anthropogenic sound on fish can range from immediate death to no impact whatsoever. Since fish are a vital component in most ecosystem food webs, and since a lot of fish species have a high economic value, it is necessary to document sound effects and to define thresholds for different combinations of sound sources and fish species.

In this study, we addressed sound effects as a result of pile driving during offshore wind farm (OWF) construction, an increasingly important human activity throughout the North Sea. Pile driving effects were assessed for young individuals of European sea bass *Dicentrarchus labrax*, which is a fish species with a closed swim bladder (physoclists). The PhD study started from the assumption that there is a 100% mortality in fish eggs and larvae up to 1 km around a pile driving source, as formulated by Prins et al. (2009).

The multidisciplinary study aimed to disentangle the effects of impulsive sound (produced by pile driving) on young fish, focusing on the following research questions:
- Are young fish (larvae and juveniles) affected by impulsive sound, what are the effects, and at what level do they manifest, *e.g.* mortality, (sub)lethal injuries, stress responses or behavioural responses?
- Can the effects on young fish be linked to a specific sound-related metric or biological parameter? Can sound thresholds at which underwater sound negatively affects young fish be identified?
- What is the ecological significance of the observed effects?
- How will the results from this PhD add to management and policy regulations in Belgium (and Europe), *i.e.* in order to minimise the environmental impact of pile driving activities in future offshore wind farms, and to achieve Good Environmental Status (GES) for Marine Strategy Framework Directive (MSFD) descriptor 11?

The context of this multidisciplinary study on the impact of pile driving on European sea bass is outlined in Chapter 1. A general introduction to the underwater world of sound is given. In the North Sea, shipping, seismic surveys, underwater explosions and pile driving are identified as the main contributors of the anthropogenic sound energy. Sound plays an essential role in conveying environmental information to marine fauna (e.g. marine mammals, fish and invertebrates). Particularly in marine mammals, sound has a key role in social and foraging behaviour. But of all vertebrates, fish exhibit the greatest diversity of hearing sensitivity and structures. The frequency range of most anthropogenic sound overlaps with the hearing range of fish and in addition, the sound can cause (mortal) physical damage to the marine mammal and fish. The exact impact, the underlying mechanisms and the ecological consequences of anthropogenic sound on marine life are not yet understood, especially for fish. In Europe, anthropogenic underwater noise was labelled as a pollutant within the MSFD of the European Commission. Consequently, the impact of the underwater sound generated by various anthropogenic sound sources, need to be evaluated on marine life in order to take appropriate measures. In Belgium, a new anthropogenic sound source, pile driving, was recently introduced. It is the main method to install OWFs and will regularly be used in the next couple of year. Therefore, this PhD study took pile driving as the impulsive sound source to study its impact on marine fish.

The following four chapters (Chapter 2-5) present the assessment of the impact of high intensity pile driving sound on acute and delayed mortality, acute and chronic physiological stress responses and at low intensity impulsive sound the impact on behaviour of young European sea bass. Furthermore, the critical sound parameters of the physiological stress responses were studied in detail.

Chapter 2 presents the results of an *in situ* experiment on board of a pile driving vessel addressing acute and delayed mortality of juvenile (68 and 115 days old) European sea bass. It was the first field study to assess fish mortality as close as 45 m from the offshore pile driving source for a complete pile driving session. Fish were exposed to 1739 up to 3067 pile driving strikes with a single strike sound exposure level (SEL$_{ss}$) between 181 and 188 dB re 1 µPa².s, and a cumulative sound exposure level (SEL$_{cum}$) between 215 and 222 dB re 1 µPa².s. No increased acute mortality was observed when we compared European sea bass (of 68 and 115 days post hatching) exposed to pile driving with a control group exposed to ambient background sound levels in between the pile driving sessions. This study validates the results provided by previous laboratory studies inside acoustically controlled chambers. The surviving fish were transported back to the lab and their survival was further monitored for two weeks. At least under optimal laboratory conditions, we observed no delayed mortality caused by pile driving. This study rejected the 100 % mortality hypothesis, and if internal injuries were present, they were shown not to be mortal.

In Chapter 3 the physiological stress response of juvenile sea bass (68 and 115 days old) was investigated under pile driving. So far, this has never been studied. During the same *in situ* experiment as described in chapter 1, primary, secondary and tertiary stress responses were investigated during or after exposure to a complete pile driving session. As a primary stress response, whole-body cortisol seemed to be too sensitive to 'handling' bias. However, a strong secondary stress response to pile driving was detected as significant reductions in oxygen consumption rate (49 – 55%) and low whole-body lactate concentrations. Contrary to fish used on the first day of the experiment, the fish used on the second day were already indirectly exposed to pile driving. Fish in the control group of that experiment reduced their respiration by 34 – 40% compared to the control group on the first day. This may be indicative of a prolonged stress response or increased sensitivity towards new stressors. A tertiary stress response only manifests when homeostasis cannot be re-established. After 30 days in the laboratory, specific growth rate and fitness of the exposed fish were not affected compared to unhandled fish, so a tertiary stress response was absent. Still, a short term reduction in fitness was demonstrated. Hence, we can assume that repeated exposure to impulsive sound in the field will inevitably lead to less fit fish in the wild.

Chapter 4 explores the critical sound parameters responsible for the acoustic physiological stress response observed in the field experiment. The primary and secondary stress responses of larval and juvenile European sea bass to strong impulsive sound were compared between two lab experiments using different sound sources (SIG Sparker and Larvaebrator) and associated frequency spectra, and with the stress responses measured in a recently conducted field study (*in situ* pile driving). Both lab sound sources produced similar levels for the standard sound pressure metrics as the *in situ* pile driving, being zero-to-peak sound pressure level (L$_{z-p}$) of 208 dB re 1 µPa), SEL$_{ss}$ of 181 dB re 1 µPa²·s and SEL$_{cum}$ of 214 dB re 1 µPa²·s. However, the three sources differed in their sound frequency spectra. The whole-body cortisol results (a proxy for primary stress responses) confirmed the susceptibility of both juvenile and larval fish to handling stress. Still, based on the increased (or altered) whole-body cortisol levels, high intensity impulsive sound evoked an acoustic primary stress response. Common ground was found at the high energy levels (SEL$_{ss}$) produced between the 63 and 630 Hz 1/3 octave bands. This frequency range covers the hearing range of European sea bass, relating primary stress response in juvenile fish to hearing discomfort. Reduced oxygen consumption rates of ~50% were observed in the juveniles in the field experiment and larvae in the sparker experiment, and to a lesser extent in the juveniles of the sparker experiment. Consequently, the secondary stress response was most likely linked to high intensity sound produced at higher frequencies (>800 Hz). This secondary stress response may be related to pressure induced swim bladder discomfort.

Still, high intensity impulsive sound covering a broad frequency range (like a real *in situ* pile driving) is needed to evoke strong secondary stress responses (e.g. reduced oxygen consumption rate and whole-body lactate levels) in juvenile sea bass. This implies that lab results may not directly be translated to the real world, as some known or unknown parameters (like frequency content) may differ. Based on the experiments, the sound pressure level at which stress responses were evoked, seemed to be located between SEL$_{ss}$ 170 and 180 dB re 1 µPa²·s. More studies on different life stages and the role of non-standard sound parameters such as particle motion are needed to confirm these values as real stress thresholds for fish.

Underwater sound has the potential to disturb the behaviour of fish even at lower sound pressure levels, resulting in a much wider impact range around the pile driving source than high sound pressure levels. Since functionally important behaviour, such as social interactions and foraging, can contribute significantly to the survival and reproduction of fish, any impact on functional traits can directly be translated into fitness consequences. However, so far only a couple of studies have tested the acute impact of anthropogenic sound exposure on fish behavior. Consequently, in Chapter 5, fish behaviour was studied in response to impulsive sound on three consecutive days in a laboratory set-up. In this indoor laboratory study, we tested the influence of pile driving sound on the swimming activity and

aggression of young juvenile European sea bass *Dicentrarchus labrax* before, during and immediately after the 25 min sound exposure (1000 strikes, SEL_{ss} =156 dB re 1 μPa²·s, L_{z-p} = 175 dB re 1 μPa; SEL_{cum} = 186 dB re 1 μPa²·s). We also tested the impact on feeding tendency and efficiency when fish were already 15 min exposed to the impulsive sound. The sea bass interrupted their swimming activities and ceased any aggressive actions to conspecifics at the onset of the impulsive sound exposure. The behavioural effects of sound exposure returned to the pre-exposure base line within the 25 minute exposure period. On the first day, the sound exposure caused an attention shift. Resulting in a delayed reappearance of the aggressive attacks to just after the sound exposure and a reduction in the number of food intake events during and immediately after the sound exposure compared to pre-exposure food intake events on the first day. This attention shift was no longer observed during day 2 or 3 of the experimental trials. These findings indicate that fish can habituate to the impulsive sound over a relatively short period of time, which may moderate the acute sound impacts on behaviour. It remains to be tested whether this also applies to wild-ranging fish and whether such habituation effects are also to be found in other species.

In Chapter 6, the technical and practical challenges to field and lab experiments in bioacoustics are discussed. Results of this PhD study are discussed in a broader perspective through comparison with literature on pile driving and other anthropogenic sources of impulsive sound. The results obtained during this study allowed describing the impact of pile driving sound on European sea bass. Exposure to a complete pile driving session as close as 45 m from the pile driving activity did not result in acute or delayed mortality of juvenile European sea bass. It did lead to a strong physiological stress response limited to a relative short period of time, which can be extended by multiple sound exposures. Based on the field and lab results, the physiological stress response found in larvae and juveniles could be related to the standard sound metrics (SEL_{ss}, SEL_{cum} and L_{z-p}) and the frequency range of the highest energy. Furthermore, hearing discomfort and swim bladder discomfort could explain respectively the primary and secondary stress response. The above mentioned studies involve high intensity underwater sound found at close range from the pile driving source. At larger distance from the pile driving source, the impulsive sounds contain less energy but can still induce a behavioural response in juvenile European sea bass at the onset of the sound exposure. During the sound exposure, European sea bass were able to recover from their initial response and repeated exposure had no effect on feeding. Combining these results with other data from literature reveals the interspecific variability of in their behavioural response to the same type of stressor. Therefore, more species varying in life history strategies need to be studied before the results can truthfully be generalised.

Integration of the study results with current knowledge allows us to make suggestions regarding sound thresholds for mortality, physiological stress and behavioural changes. Since mortality was absent in our field study, the mortality threshold must lie above the measure sound parameters (SEL_{ss} > 188 dB re 1 μPa².s; SEL_{cum} > 222 dB re 1 μPa².s.; L_{z-p} = 210 dB re 1 μPa). This study was the first to propose a sound threshold range in which physiological stress could occur in fish which lied between SEL_{ss} of 170 - 181 dB re 1 μPa²·s at frequencies higher than 315 Hz. A threshold for behavioural disturbance linked to pile driving cannot yet be determined.

Additionally, consequences on an ecological level need to be evaluated. In other words, effects on an individual level need to be modelled into population effects since individual effects in fish are subordinate to population effects from an ecological point of view. In order to do so, data on the presence of sound sources, sound propagation, important impact on the individual, population size, distribution and affected (sub)population need to be estimated before the individual effect can be modelled into a population effect. This is not yet possible for fish, but given the results about the effects found on individual fish, we can expect that the ecological consequences of pile driving sound on fish health are subtle.

In Europe, the MSFD defined a GES in which underwater sound needs to be at levels that do not adversely affect the ecosystem. A Technical Subgroup Noise (TSG Noise) was commission to further develop the descriptor on underwater noise. They advised to set up a sound register, to log all sound producing human activities. In addition, they appointed 'considerable displacement' of marine organisms as the most relevant impact of impulsive sound. Finally, an inventory of the pulse-block days in the EU's regional seas can be modelled, based on the presence of anthropogenic sound sources in ¼ ICES rectangles that are producing sound levels above the threshold linked to the 'considerable displacement' impact. A GES contains all marine organisms while the TSG Noise mainly based its advice on marine mammals. 'Considerable displacement', may not be the most relevant impact of fish. Fish are also neglected in the national legislation of the Member States of Europe. Based on this PhD, the effects of pile driving sound on fish are more subtle than anticipated and no stringent measures are needed ad hoc in Belgium or other member states. Still, more research is needed to support or reject the decision to exclude fish from management in order to ensure a GES for all marine fauna.

Finally, future research targets are identified. These are needed to progress towards an acoustically sound approach. The lack of particle motion data remains the big gap and needs to be addressed by

future studies. The underlying critical sound parameters need to be further unravelled that induce the physiological stress and behavioural responses. Furthermore, data is needed on the long-term impact of the acoustic stressor in order to model the ecological consequences of pile driving at population level. Studying the fish in its natural environment with new technologies is the way to go. And finally, the impact of the continuous sound produced by the next 20 years of the operational OWF on fish health need to be addressed.

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Bioavailability and mixture effects of metals in different European mussel populations

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More than 100 million tons of chemicals that have the potential to pose a risk to the environment are produced in Europe each year. A subset of these chemicals may, intentionally or not, enter and affect the environment. To protect the environment and the diverse services it provides, it is important to know what the impact and risk of a chemical release may be. Underestimating the risk can have harmful effects on the environment and on human health. Overestimating the risks may, unnecessarily, increase the costs of preventing or ameliorating pollution. Hence, accurate knowledge of the effects and the associated risks is essential.

Predicting the effect of a chemical is, currently, primarily based on the results of single-species experiments with freshwater organisms that are exposed to a single stressor in a standardized (laboratory) environment. However, in reality organisms are not exposed to these standardized conditions, but live in and are exposed to a variable environment. Furthermore, inter-population differences in sensitivity may exist due to differences in local adaptation and even a single organism's sensitivity may change during its lifetime. Finally, organisms may be exposed to multiple stressors, natural or anthropogenic, simultaneously. Hence, it is suggested that it might not be possible to accurately predict the adverse effects using the currently prescribed methods.

The main objective of this research was to examine the effect of these potential sources of variation on the toxicity of chemicals on marine organisms in order to increase the realism of current environmental risk assessment procedures. This was accomplished by assessing the influence of environmental variation, mixture toxicity, population variability and life-stage variation on the accumulation and toxicity of Cu on a Cu sensitive marine test species, the mussel.

In part one of this research, the combined influence of the two main marine environmental variables, salinity and dissolved organic carbon (DOC), on the distribution, accumulation and toxicity of Cu in mussel larvae (*Mytilus galloprovincialis*) was assessed. By using synchrotron radiation X-ray fluorescence, the distribution and accumulation of Cu in mussel larvae were determined. Cu body burden concentrations varied between 1.1 and 27.6 µg/g DW larvae across all treatments and Cu was homogeneously distributed at a spatial resolution level of 10x10 µm. The 48 h Cu EC10 varied between 2.8 and 11.2 µg/L, confirming that mussels are very sensitive to Cu. Cu accumulation and toxicity decreased with increasing DOC concentrations which can be explained by an increase in Cu complexation. In contrast, an increase in salinity increased the Cu toxicity. This change could not be explained by Cu speciation or competition processes and suggests a salinity-induced alteration in physiology, resulting in a changed Cu sensitivity.

In part two a similar experiment was performed with two populations of settled mussels (North Sea and Baltic Sea). Baltic Sea mussels were chosen because previous research had indicated that the mussel population in that region is already stressed, due to the low salinity of this marine system. It was hypothesized that environmentally stressed populations would be more sensitive to anthropogenic pollution as they have to allocate more energy towards basic maintenance. The Baltic Sea population did accumulate more Cu compared to the North Sea population (both in the gills and in the total soft tissue). However, both populations exhibited an equal sensitivity to copper. This suggests that environmentally stressed populations are not necessarily more sensitive to anthropogenic pollution and that different populations may have a different way to cope with excess Cu. The influence of salinity and DOC on the accumulation and toxicity of Cu to settled mussels was very limited in both populations. Hence, it is concluded that DOC-Cu complexes are bioavailable to settled mussels. Due to the absence of a protective effect by DOC in settled mussels, implementing a DOC correction factor to determine a Cu environmental quality standard for Cu – as is done for freshwater environments – cannot be proposed for marine environments.

Organisms are not only exposed to a changing environment, but are also frequently exposed to multiple metals simultaneously. In the North Sea, for example, high Cu concentrations frequently coincide with high concentrations of other metals like Ni and Zn. Nevertheless, little information is available on the effect of metal mixtures, certainly of environmentally realistic concentrations, in the

marine environment. Therefore, in part three of this research, the effect of metal mixtures was assessed. The effect of the Cu-Ni binary mixture on *Mytilus edulis* larvae was assessed using a full factorial design. The reproducibility of the results was assessed by repeating this experiment 5 times during a 3-year period and having them being performed by different researchers. The data were analyzed using a Markov Chain Monte-Carlo algorithm (MCMC). The use, for the first time for mixture toxicity analysis, of this statistical tool enabled the estimation of both the mixture toxicity deviation from the reference models and the uncertainty on the deviation. The results demonstrated that mussel larvae were about 100 times less sensitive to Ni than to Cu (average Cu EC50: 4.1 µg/L vs Ni EC50: 414.7 µg/L). When mussel larvae were exposed to a mixture of these metals, a reproducible ratio-dependent deviation from the concentration addition reference model was observed. Antagonism was observed at high nickel concentrations (> 200 µg/L) but, more importantly, low concentrations of Ni (as low as 4.9 µg/L) resulted in a synergistic interaction with Cu. To our knowledge this is the first time that synergism (according to the concentration addition reference model) was observed at low, environmentally relevant, metal concentrations. This highlights the need to consider mixture effects in marine environmental risk assessment procedures.

In part 4 mussel larvae from two populations (North Sea and Baltic Sea) were exposed to Cu, Zn, Ni and a Cu-Zn mixture to assess both the influence of the mixture and determine possible inter-population differences in metal (mixture) sensitivity. The Baltic Sea mussel larvae were approximately 20 % smaller and grew slower than North Sea larvae. This agrees with previous research that suggested that settled Baltic Sea mussels are stressed by the low salinity and therefore grow slower. Mussel larvae from the Baltic Sea were three times more sensitive to Zn (as single substance) and Ni, as expected based on the proposed but untested hypothesis that the Baltic Sea mussel population would be more sensitive (due to the environmental stress) to metal exposure. However, both populations had an equal sensitivity to Cu and the effect of the Cu-Zn mixture was also similar in both populations. This indicates that inter-population variability in sensitivity is metal-dependent. It can be concluded that: all variables investigated in this study changed the accumulation and/or the toxicity of Cu in mussels. The assessed environmental variables, i.e. salinity and DOC, had a strong influence on the accumulation and toxicity of Cu in mussel larvae but not in settled mussels. Furthermore, the influence of salinity on the Cu toxicity in mussel larvae could not be explained based on complexation and competition. Therefore, using the current knowledge, the development of a universal marine BLM based only on the water chemistry is currently not possible. Next to the influence of environmental factors, we have provided evidence that synergistic metal mixture interactions can occur at concentrations currently measured in marine environments. To adequately protect marine organisms, metal mixture interactions should be included in future environmental risk assessment procedures. Finally, the two assessed populations were equally sensitive to Cu. This suggests that naturally stressed populations are not 'by default' more sensitive to pollution than unstressed populations. However, population differences in organism sensitivity to other metals (Zn and Ni) were observed, indicating that inter-population variability is pollutant-dependent and that this knowledge may need to be included in future ERA procedures.

Realistic environmental mixtures of hydrophobic compounds do not alter growth of a marine diatom (published in the Marine Pollution Bulletin (2016), 102, 58-64)

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In this paper we determine whether a realistic mixture of hydrophobic chemicals affects the growth dynamics of a marine diatom and how this effect compares to the effect of temperature, light regime and nutrient conditions. To do so, we examine the specific growth rate of *Phaeodactylum tricornutum* in a 72 h algal growth inhibition test using a full factorial design with three nutrient regimes, two test temperatures, three light intensities and three chemical exposures. Passive samplers were used to achieve exposure to realistic mixtures of organic chemicals close to ambient concentrations. Nutrient regime, temperature and time interval (24, 48 and 72 h) explained 85% of the observed variability in the experimental data. The variability explained by chemical exposure was about 1%. Overall, ambient concentrations of hydrophobic compounds present in Belgian coastal waters, and for which the passive samplers have affinity, are too low to affect the intrinsic growth rate of *P. tricornutum*.

This paper illustrates the beneficial effects of interdisciplinary research in marine sciences. The novelty of the paper not only relates to the use of passive samplers which are complementary to the conventional spot sampling campaigns. The true novelty relates to the process that all collaborators went through when performing the research. An inspiring series of interactions between field workers (deployment of the passive samplers), technical personnel (support of the laboratory experiments), chemists (characterization of the chemicals), statisticians (statistical analyzes) and engineers (interpretation of results) occurred within the scope of this research. Furthermore, the results described in this paper, i.e. that ambient concentrations of organic chemicals do not affect the growth of a marine diatom, moved the field of multiple stressor marine ecotoxicology forward. The latter is one of the reasons why this paper was published in the Marine Pollution Bulletin which is a high-ranked peer-reviewed scientific journal in the field of Marine and Freshwater Biology (6/104). The paper is published since January 2016 and has already been cited twice in other peer-reviewed scientific journals.

The "*Alcanivorax* paradox": mild hydrostatic pressure impacts deep-sea oil bioremediation

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The relevance of oil spill contamination for the North Sea and the Belgian coast

Belgium coastline is one of the shortest among North Sea costal States. Yet, it is subjected to an intense naval traffic and is considered of strategical relevance ecologically and socioeconomically (Schallier and Van Roy, 2016). The presence of several major harbors in the South part of the North Sea together with illegal oil discharge exposed this area to chronic oil pollution. Since policy measures as the MARPOL 73/78 Convention (1983) and the multilater Bonn Agreement (1969/1983) were established, aerial surveillance and oil spill response sensibly reduced oil pollution of surface and shallow Belgian waters (Lagring et al., 2012), this target met also for other surrounding countries (e.g., The Netherlands [Camphuysen and Vollaard, 2016] and Denmark [Christensen and Carpenter, 2016]). Nonetheless, when one speaks of petroleum and sustainable development, maritime transport with oil cargo will continue to exist along with the risk of accidents involving oil and by-product spills (Barrow and Rothschild, 2002). A quick look at the multiple petroleum reservoirs studding the North Sea indicates that this remains a major threat for Belgium as for other North Sea costal State.

Alongside naval traffic-related pollution, the recent Deepwater Horizon (DWH) disaster in the Gulf of Mexico (April 2010) opened a new era for oil spills worldwide. The blowout of a well rig at 1500 m below surface level (bsl) resulted in the largest marine oil spill in history (780'000 m^3 of spilled oil). Oil deep-sea platforms are defined as those operating in the mesopelagic zone (200 to 1000 m bsl, subjected to mild hydrostatic pressure [HP] of 2 to 10 MPa, or 20 to 100 bar) and further below (bathypelagic zone). There are several deep-sea oil fields in the world, two being located in the North Sea (Troll A and Gullfaks, in the Norwegian sector, 472 and 380 m, respectively). Limited maturity of deep-sea technologies was claimed as a critical factor for the DWH spill (Jernelov, 2010; Thibodeaux et al., 2011) which in fact lasted for 3 months. The implications for the Gulf of Mexico were impressive: dispersant application at the well rig, alongside deep-sea conditions of temperature and HP, formed multiple oil plumes (Kujawinski et al., 2011), with underwater currents enlarging the deep-sea area directly impacted by oil. The estimated deep seafloor contaminated area is 3200 to 8000 km^2 large (Chanton et al., 2014). The oil reaching the surface was eventually delivered to the coast and directly impacted an area of 180'000 km^2 and more than 25'000 km of shorelines (Norse and Amos, 2010). While the range of techniques to remove oil on surface and shallow waters is relatively wide (e.g., booms and skimmers, dispersants, oil burning), at deep-sea conditions microbial biodegradation represents the only (bio)technology available.

With continued political and economic pressure to get access to deep-sea oil the risk for future spills in the deep sea is not expected to decrease. A full understanding of the physiological requirements enhancing bioremediation at deep-sea condition is therefore pivotal to provide policymakers with critical information to elaborate proper protocols to combat deep-sea oil spills. This is particularly true when considering the metabolic network entrenched in deep-sea marine life. Following the DWH, the implications for the latter were outstanding: deep-sea sediments were classified as low to moderately polluted (Romero et al., 2015); deep-sea pore-water exerted high toxicity levels and DNA mutagenesis (Paul et al., 2013); primary production and carbon export to the deep sea was reduced (Prouty et al., 2016); in sea-food, concentration of certain petroleum-hydrocarbons was 1000 times above the threshold for human consumption (Sammarco et al., 2013). While another oil spill of the proportions of the Gulf of Mexico is unlikely, the impact of a deep-sea uncontrolled oil release within the much smaller area of the North Sea would expose oil remediation operators to a number of novel different issues, including the lack of meaningful long-term solutions for oil degradation.

Research environment supporting the present project

The text corpus presented here was elaborated in the frame of the 33-partner EU project Kill Spill (grant agreement N. 312139, Integrated Biotechnological Solutions for Combating Marine Oil Spills). Belgian science was represented by the Center for Microbial Ecology and Technology (CMET) of the University of Gent (UGent). The data discussed here refers to the 3-year research period of the present candidate, Alberto Scoma, as postdoctoral researcher at CMET-UGent, which resulted in three research

articles and one review in Q1 international peer-reviewed journals (i.e., Scientific Reports, impact factor [IF] 5.5, and Frontiers in Microbiology, IF 4.2), all published between March and August 2016. Following the "Alcanivorax paradox" case we received two invitations for a paper by EMBO Reports (IF 7.7) and Trends in Biotechnology (IF 12.1), currently under preparation. We expect the publication with these four different editors (Nature, Frontiers, EMBO and Cell) to widen the impact of our results. It is worth mentioning that the present body of work is completed by two further manuscripts presently under revision in Frontiers in Microbiology and Environmental Science and Technology (IF 5.4) and one under preparation for ISME Journal (IF 9.3). These latter findings are not discussed here. A complete list of the material published, under revision or preparation is provided in chapter six of my attached CV. The present results put CMET-UGent atop the field of HP microbial oil degradation physiology, an emerging topic with a rising number of competitors, with current developments involving a PhD candidate who is applying synthetic communities and in situ conditions of both HP and low temperature.

Physiology of hydrocarbons degradation at mild hydrostatic pressure

Due to its unique nature, the DWH spill was extensively investigated either through in situ studies employing next-generation sequencing techniques or by ex situ experiments simulating microbial biodegradation in laboratory conditions. Despite supplying unprecedented information, both approaches failed to describe the exact metabolic routes and constraints in deep-sea bioremediation. In situ studies using molecular techniques can only provide information on potential biological activities while, with no exception, ex situ studies neglected the impact of one of the major drivers for biodiversity in marine environments, i.e., HP (Ghiglione et al., 2012).

Pressure increases linearly with depth about 0.1 MPa (or 1 bar) every 10 m of seawater column. The relative HP increase experienced in the mesopelagic zone (2 to 10 MPa) is much higher with respect to deeper zones, as HP doubles 5 times within 800 m, suggesting this might be a transition area for HP resistance. Other critical factors such as pH and salinity are not as remarkably affected by depth as HP, while temperature varies significantly in this zone although to a lower extent with respect to pressure and depending on latitude and season (Garrison, 2015). Very little is known about the physiological requirements and metabolic features of HP-adapted oil-degraders, with only four papers in the whole literature, two of which dating back to the 1970's (Schwarz et al., 1974; Schwarz et al., 1975; Grossi et al., 2010; Schedler et al., 2014). To investigate the impact of HP on oil biodegradation we tested three independent axenic cultures of the marine obligate oil degrader (i.e., hydrocarbonoclastic) Alcanivorax genus, namely A. jadensis KS_339, A. dieselolei KS_239 and the reference strain A. borkumensis SK2, using three HPs (0.1, 5 and 10 MPa, equivalent to surface water, 500 m and 1000 m bsl) and n-dodecane as sole carbon source. Sequences belonging to these species are ubiquitously detected in a wide range of oil-contaminated environments, from surface to deep sea (Lai et al., 2011), in both seawater and sediments (Wang et al., 2010; Kostka et al., 2011). In particular, A. borkumensis typically dominates oil-contaminated surface marine waters worldwide (up to 90% of the whole population within 30 days after the oil spill, Yakimov et al., 2007; Head et al., 2006), background to why it is adopted as a model organism to investigate hydrocarbon degradation pathways in marine environments (Kasai et al., 2002). In our experiments, HP significantly reduced cell replication in all Alcanivorax strains to a different extent (ranging from a factor 3 to 5), with the most severe effects observed for the predominant A. borkumensis where 5 MPa was sufficient to suppress culture growth (i.e., cell number at the end of the incubation was lower than at the onset). This was mirrored by a lowered CO_2 production by the cultures, a proxy for hydrocarbon degradation capacity, as reflected in a reduced pH acidification owe to CO_2 hydration in water as $HCO_3^- + H^+$ (there are no fermentation products affecting pH under aerobic hydrocarbons degradation). Cell damage increased at increasing HP, with the major impact being observed in A. borkumensis, where intact cells where reduced to 10% already at 5 MPa. In this strain, a further HP increase to 10 MPa triggered some resistance mechanism, as cell intact and final number significantly improved with respect to 5 MPa, while remaining largely lower than at surface water pressure. Such a dramatic effect on hydrocarbon degradation and culture growth imposed by mild HP was confirmed by transcriptome analysis, with large downregulation affecting the whole genome expression at 10 MPa as compared to surface pressure in the examined A. dieselolei KS_293 and A. borkumensis SK2. While a genomic-based approach has been proposed for piezophilic adaptation (i.e., increased microbial growth at increasing HP) (Simonato et al., 2006), piezosensitive and piezotolerant bacteria might cope with HP through a physiological response. Given the piezosensitive nature of Alcanivorax, the few upregulated genes highlighted by transcriptomic analysis were indicative of the major physiological requirements imposed by mild HP shifts. The most interesting results concerned the hydrocarbon degradation pathway. Following alkanes activation by introduction of an oxygen atom, the generated fatty acid normally enters beta-oxidation to be fully mineralized to CO_2. None of the genes related to beta-oxidation was upregulated at HP, contrary to several biosynthetic pathways related with fatty acid elongation. Lack of upregulation of beta-oxidation genes does not entail that this was not active to some extent, provided that n-dodecane was the sole carbon and energy source. However, the capacity to withdrawn oxidized n-alkanes from the

degradation pathway and incorporate them as their corresponding fatty acids in the membrane has been shown for Alcanivorax (Naether et al., 2013) and other hydrocarbon-degrading microbes (e.g., Marinobacter hydrocarbonoclasticus [Doumenq et al., 2001] and Rhodococcus erythropolis [de Carvalho et al., 2009]). Alternative hydrocarbon utilization pathways may be explained as a way to counterbalance the impacted energy-production metabolism. The multicomponent ATPase complex was largely upregulated indicating that generation of ATP was affected by HP along with cytochromes, whose nature shifted from oxidases to reductases. The latter resembles a strategy adopted by Shewanella piezophila which possesses two respiratory chains for ambient and high HP (Kato and Qureshi, 1999). The central role of electron and proton transport at HP is majorly highlighted by the model piezophile Photobacterium profundum SS9 which possesses two complete operons for the F_0F_1 ATPase and multiple cytochrome sets (Vezzi et al., 2005). Finally, the effect of concomitant stresses imposed by increased osmotic and HP on the model organism A. borkumensis SK2 was tested. A synergistic effect enhancing cell activity at HP was noted, but this resulted into a dramatically lowered culture performance, confirming the piezosensitive nature of this strain (Scoma and Boon, 2016).

The environmental significance of the Alcanivorax paradox

While predominating in oil-polluted surface waters worldwide, the bioremediation capacity of the hydrocarbonoclastic genus Alcanivorax is reduced by mild HP equivalent to only 500 m bsl (Scoma et al., 2016a,b). When Alcanivorax species were isolated from up to 5000 m bsl (up to 50 MPa, Liu and Shao, 2005; Lai et al., 2011) they could not grow <10°C, i.e., at much higher temperatures than those registered for these depths (<4°C). As such isolation protocols did not apply HP, one could conclude that HP relief reestablishes Alcanivorax capacity to grow. As a matter of fact, Alcanivorax isolates were obtained from decompressed DWH water samples (Gutierrez et al., 2013), resembling results for DWH surface water and sediment samples where Alcanivorax enrichment in situ was noted based on sequence analysis (Kostka et al., 2011; Liu and Liu, 2013). On the contrary, in situ Alcanivorax abundance in bathypelagic water (Gutierrez et al., 2013), oil plume (Hazen et al., 2010; Dubinsky et al., 2013; Yang et al., 2014) and deep-sea sediments was low (Kimes et al., 2013), unrelated to hydrocarbons (Kimes et al., 2013) and its contribution to deep-sea bioremediation considered negligible (Gutierrez et al., 2013). Upon reviewing the literature related to the DWH, we noted that the Alcanivorax paradox actually extends to all hydrocarbonoclastic bacteria (Scoma et al., 2016c) such as Marinobacter or the several Oceanospirillales genera as Thalassolituus, Oleiphilus, Neptunomonas and Oleispira, which were not enriched in deep-sea oil contaminated samples (Hazen et al., 2010; Dubinsky et al., 2013; Kimes et al., 2013; Mason et al., 2014). Low temperature was proposed to account for this (Redmond and Valentine, 2012), but species as O. antarctica are psychrophilic (Yakimov et al., 2003) and sequences belonging to these genera have been retrieved from hydrocarbon-seeps in the Gulf of Mexico (King et al., 2013).

Most importantly, impacted beta-oxidation at deep-sea HP was indicated by in situ studies on DWH water and sediment samples based on metabarcoding and other metaomics approaches (Hazen et al., 2010; Lu et al., 2012; Kimes et al., 2013; Mason et al., 2012; 2014; Rivers et al., 2013). Upregulation of alkane-1 mono-oxygenases responsible for n-alkane activation was detected in all studies, consistent with our experiments with Alcanivorax (Scoma et al., 2016a,b). However, we argued that genes related to beta-oxidation were only partially upregulated in two studies out of six (reviewed in Scoma et al., 2016c), in sharp contrast with the high concentration of n-alkanes detected in plume and sediment DWH samples up to 1.5 year after the spill.

While the reason why predominant hydrocarbonoclastic genera and beta-oxidation are impaired by deep-sea HP remains unknown, we showed that HP represents a major factor driving microbial community structures and pathways associated with oil-degradation, both distinct from those occurring at surface water pressure. The physiology of microbial oil degradation is still in its infancy, with the present published data by CMET-UGent already constituting 50% of the whole literature on this topic. Understanding the metabolic routes governing oil degradation at HP is key to develop successful bioremediation strategies in the deep sea. This would improve understanding of bioremediation pathways and assist policymaker in assessing protocols challenging biodegradation at deep-sea HP.