

light conditions affect the growth of photosynthetic organisms (which were the predominant fouling organisms in our study), we recommend considering the boats’ position in harbors for minimizing fouling. Moreover, boats that have used boat washers had significantly lower fouling at the end of the season so we encourage the use of this environmentally-friendly maintenance method. These results suggest that the maintenance habits ought to be reconsidered and that the use of toxic compounds in antifouling paints may be unnecessary for leisure boats in the northern part of the Baltic Sea.

MO038

Lethal and sub-lethal effects of aniline to the copepod *Tisbe battagliai*: simulation of Hazardous and Noxious Substances (HNS) spill profiles

M. vannoni, Cefas; T. McGowan, Cefas Lowestoft Laboratory; D. Sheahan, J. Aldridge, Cefas; M. Kirby, Hazardous and Noxious Substances (HNS) are chemicals, other than oil, which have potential hazards to human health or to other organisms. Over the last decades transportation of HNS by sea has increased significantly so emphasizing the need to be prepared to respond to incidents involving HNS. The European project ARCOPOLplus recently identified 23 HNS as having high likelihood of being present in maritime transport incidents suggesting these be the focus of efforts to fill data gaps. This study therefore considered aniline, one of the 23 chemicals listed as priority for improving existing data. The volume of chemical spilt at sea and its behaviour (e.g. whether it dissolves or evaporates) and location (e.g. open sea or nearshore) are major factors influencing extent of any impact. Chemical modelling can predict the dispersion and fate of a chemical during a spill and this can be linked to toxicity data to predict likely impact. However toxicity data is usually derived from constant exposure studies and under natural conditions this rarely occurs. This work investigates the acute and chronic toxicity of aniline using representative spill profiles. The copepod *Tisbe battagliai* was chosen as test species because of its sensitivity and importance in the marine food webs. Tests included short exposures (1 and 2 hours), simulated varying concentration spill profiles (based on modelling data) and constant exposures. Different life stages of *Tisbe* were used to obtain acute and sub-lethal effects data. Results indicate that aniline does not have prolonged effects even after exposure to relatively high peak concentrations providing exposure duration is short e.g. 1-2 hours. Preliminary data indicate realistic spill profiles may have negligible impact for chemicals that do not accumulate or persist due to the relatively rapid dilution that occurs at offshore locations. At near shore locations more significant effects are predicted. This study indicates that use of standard toxicity data could lead to an overestimate of the potential harm caused by a spill. Whilst such a precautionary approach is recommended where there are significant threats to marine resources, more realistic assessments may be of value in predicting actual levels of impact and potential for recovery. *This work has been carried out with support from Defra, Project MERR C5905 and in the frame of ARCOPOLplus project, co-funded with ERDF through the Atlantic Area Transnational Programme*

MO039

Integrated assessment of the chemical environmental state of Cartagena Bay (NW Mediterranean) in relation to marine chemical contamination

C. Martinez-Gomez, B. Fernandez, J. Valdes, C. Navarro, M. Albertosa, J.A. Campillo, V. Leon, J.M. Benedicto, Instituto Español de Oceanografía; T. Burgeot, IFREMER / BIOGEOCHIMIE ECOTOXICOLOGIE; D. Vethaak, DELTARES Cartagena bay (SE Spain) was chosen as study case of the NW Mediterranean region to demonstrated the suitability of the integrated marine environmental monitoring of chemicals and their effects proposed by European experts (ICON Project) for the North Sea. Coordinated sampling of key environmental matrices (surface sediment, fish and mussels) was performed in Autumn 2008, considering biogeographical characteristics. The benthic fish red mullet (*Mullus barbatus*) and mussels (*Mytilus galloprovincialis*) were used as target species. Due to scarcity of mussel populations on natural substrates, mussels were transplanted for a period of six weeks in a coastal site located in the bay. Contaminant related-biomarkers of exposure and effects were measured in both target species. Contaminant concentrations were analyzed in sediments and biota but also and sediment-passive samplers. *In vivo* embryotoxicity bioassays were performed using sediment elutriates. Additionally, a battery of *in vitro* gen reporter bioassays covering different mode of action of toxicants were also investigated using extracts of the sediment-passive samplers. Chemical and biological data were assessed against its corresponding assessment criteria, and then integrated over levels of matrix. Assessment was expressed with varying levels of aggregation (contaminants, bioassays, biomarkers of exposure and biomarkers of effects) to graphically represent the proportion of different types of determinants exceeding either level of assessment criteria. Subsequently, data were aggregated further into a single schematic showing the proportion all determinants that exceed assessment criteria in Cartagena bay, using a threshold of 95%

MO040

Hepatic histopathological findings in Ariidae *Cathorops spixii* submitted to

anthropogenic metal exposure in two Brazilian estuaries

J.d. Azevedo, Federal University of Sao Paulo / Biological Sciences; H.N. Silbiger, University of São Paulo USP / Biological Oceanography; J.E. Sarkis, IPEN - Nuclear and Energy Research Institute / Center for Chemical and Environmental Technology; J.F. Dias, University of São Paulo USP Histopathological lesions are used as biomarkers of contamination by organic and inorganic chemical compounds in the aquatic systems and can reflect chronic effects in sentinel species. Previous work demonstrated the ability of the catfish *Cathorops spixii* to be an efficient bioindicator species for metals contamination. Cananéia estuary is considered an area of environmental protection, and UNESCO World Heritage Site (WHS), and as such, several biomonitoring studies in the São Paulo State, Brazil, use this estuary as a reference environment due to the fact that it shows very little human influence. However, in recent years, several studies have been indicating the presence of some toxic metals, for instance mercury (Hg) and lead (Pb). Santos-São Vicente estuary has a long history of contamination and release of pollutants, for instance metals such as Pb, Cd, Ni, Mn, Cu and Hg as a consequence of different anthropogenic activities such as petrochemical, metallurgical and harbor. In the present work, histopathological lesions in liver were investigated in the Ariidae *Cathorops spixii* from Cananéia estuary and in two sites affected by industrial and domestic sewage disposal, in order to verify the possible impact of the introduction of metals to the local fish species. Fish were collected in Cananéia and in two sites of the Santos São-Vicente estuary subjected to different anthropogenic influence (industrial and domestic sewage disposal). Biometric data (total length –TL- and total weight -TW- of each fish) was obtained and the fish dissected by collection of the hepatic liver to histopathological and metals analysis. Analyses of Hg was performed by FIA-CV AAS and Pb, Cd, Cu, Mn and Ni concentrations were determined by ICP MS. Fish from Cananéia showed higher levels of Mn (80%) and Cu (66%). The most important injuries observed in individuals from the impacted areas (industrial and domestic sewage) were the presence of necrotic areas, hepatitis focal, vacuolization and rupture of blood vessels. The occurrences of lesions were significantly higher in individuals from domestic and industrial area than in fish from Cananéia estuary ($p>0.05$).

MO041

Influence anthropogenic pollution on starfish’s *Asterias rubens* at different level of biological organisation

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There is growing concern that the invertebrate test organisms commonly employed in the field of aquatic ecotoxicology may not be sufficient to accurately screen for the possible deleterious effects of contaminants discharged into the marine environment. The use of echinoderms has been proposed to redress this problem, due to their ecological importance and their evolutionary closeness to the chordates. A suite of biomarkers which operated at different levels of biological organization (sub-cellular, cellular, individual and community level) were identified for use with the common starfish *Asterias rubens* L. and copepods *Scottomyzon gibberum* Scott. Adult females of *S. gibberum* live on starfish body surface of and can induce galls formation. Specimens of the *Asterias rubens* were collected near the Biological station of Lomonosov MSU at Kandalaksha Bay of the White Sea during the summers of 2012-2013 years from 10 points which characterized by different anthropogenic pollution level and ecosystem type. Indexes which widely used in parasitological studies were calculated to characterize population: prevalence, intensity, abundance, aggregations indexes and also copepods location on host body. The population increases from mid-June to late September. Population heterogeneity is shown, which can be attributed to anthropogenic load and ecosystem type. Population of copepods on starfishes increases with depth. Ecotoxicity bioassay was made with 4 different heavy metals composite concentration in salinity 21 and 25.7 ‰ (normal). Micronuclei induction was estimated in coelomic fluid phagocytic cells, which are caused by both chromosome breakages and spindle apparatus dysfunction. Low salinity level case of decrease proliferation of phagocytic cells and bacterial infection. In normal salinity heavy metals cause a dose-dependent increase the number of micronuclei. Habitat reaction (righting behaviour) shows adaptation for influence heavy metals and salinity after 10 days of experiments.

MO042

Early maternal exposure to estrogens cause malformations in eelpout fry

J.E. Morthorst, University of Southern Denmark / Department of Biology; N. Brande-Lavridsen, B. Korsgaard, University of Southern Denmark; P. Bjerregaard, University of Southern Denmark / Biology Recently malformations among eelpout (*Zoarces viviparous*) fry living in North European coastal areas with high anthropogenic input have been observed. The specific chemicals or group of chemicals causing the observed malformations are unknown. In oviparous fish species fry malformations can be induced by exposure to chemicals including endocrine disrupters. The eelpout is a viviparous and stationary fish and maternal exposure to chemicals including endocrine disrupters might explain the fry malformations observed in nature. The aims of the present

experiments were to investigate mother-offspring interactions e.g. teratogenic effects upon maternal exposure to 17β-estradiol (E2) and environmental chemicals with known endocrine disrupting effects; the PAH pyrene, the synthetic hormone 17α-ethinylestradiol and 4-*t*-octylphenol and to investigate if a teratogenic window for E2 could be established. Wild pregnant eelpout with newly fertilized eggs were either (1) exposed continuously to 17β-estradiol (E2) (5.7-133 ng/L), 17α-ethinylestradiol (≈20 ng/L), 4-*t*-octylphenol (≈6.25-50 ng/L) or pyrene (≈ 500 ng/L) for six weeks or (2) exposed to E2 during different weeks of pregnancy to investigate if a teratogenic window for E2 could be established. None of the chemicals influenced the survival of the females. Plasma levels of E2 and the yolk protein precursor vitellogenin were increased in mothers exposed continuously to E2 and an increased abundance of fry malformations was observed at the highest E2 concentration. If exposure takes place within the first two weeks of pregnancy ovarian function and fry development is severely affected. As eelpout are fairly stationary during their pregnancy individual populations could be differentially influenced as the local exposure scenarios are different and most likely vary from year to year.

MO043

Effects of 107Ag and 63Cu stable isotope sublethal exposure in oysters

***Crassostrea gigas* using cell and tissue level biomarkers**

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Marine bivalves such as oysters are known to be good sentinel organisms. Their sedentary way of life and their ability to accumulate pollutants with little metabolic transformation make them ideal organisms for this purpose. Many marine pollution monitoring programs have used oysters to obtain chemical and biological data. Previous studies have demonstrated that cell and tissue level biomarkers are reliable and sensitive tools to assess organisms and environmental health status. Silver and copper are both elements present in the environment that in high concentrations impair biological processes and affect environmental health status and even human health. In order to study the biological effects produced by these metals and their accumulation kinetics and pathways, a 28 d laboratory experiment was carried out. Oysters were exposed directly to a range of environmentally relevant concentrations of silver and copper stable isotopes (¹⁰⁷Ag and ⁶³Cu). The use of stable isotopes allowed precise and rapid tracing metal accumulation in five different tissues. Different endpoints were measured in oysters at different levels of biological complexity including induction of metallothioneins, alterations in lipofuscin and lipid contents, metal accumulation and distribution in tissues and target cells, histopathological alterations at tissue level, and changes in organism condition and mortality rates. Results indicated higher mortality in oysters exposed to high concentrations of Ag and the combination of Ag and Cu together with a general decrease in the condition index after 20 days of exposure. Histological examination and quantification of alterations in the digestive gland indicated a higher digestive gland atrophy and over time tissue degeneration in animals exposed to high Ag and Cu concentrations. A similar pattern was observed for histopathological lesions. Moreover, in the more affected oysters a progressive arrest of the gamete development occurred. Autometallographical screening for metal ions indicated the presence of metals mainly in the gill epithelium, the digestive cell lysosomes and in the basal layer of the digestive tubule epithelium of oysters exposed to Ag and Cu. In general, obtained results indicate a close relationship between metal exposure at environmentally relevant concentration levels and physical degradation of oysters. Acknowledgements: Funded by the Basque Government (grant to Consolidated Research Groups; GIC07/26-IT-393-07).

MO044

Intertidal biota and the rock shell populations after accidents of Fukushima Dai-ichi Nuclear Power Plants

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To investigate possible adverse ecological effects caused by accidents of the Fukushima Dai-ichi Nuclear Power Plants (1F) accompanied with big earthquakes and Tsunami in March 2011, we conducted field surveys at intertidal zones of 43 sites along the coastal line of eastern Japan from April to August 2012. The number of species of intertidal biota seemed to get smaller as the site was close to 1F. No rock shell (*Thais clavigera*) specimens were collected at 8 sites of Fukushima prefecture, which were located around 1F. Because the rock shell specimens were collected at many sites in Miyagi and Iwate prefectures, where enormous Tsunami attacked, it is unlikely that smaller number of intertidal species and no rock shell

specimens around 1F were caused by Tsunami. Contaminated cooling water leached from the nuclear reactors to the sea may have given any impacts to intertidal biota including the rock shell populations around 1F.

MO045

Application of European C14 method (OECD 215) on early life stage fish

growth response to marine species *Dicentrarchus labrax*

A. Tornambe, L. Manfra, S. Canepa, ISPRA; A. Rotini, Tor Vergata University; F. Oteri, G. Martuccio, ISPRA; M. Mannozi, ISPRA / ISPRA; A. Cicero, ISPRA C14 Method as described in the EU Regulation 440/2008 on juvenile fish growth toxicity test is designed to assess the chronic effects of chemicals on freshwater species. However, the use of a marine species widely distributed and well known as the sea bass can be considered more appropriate to assess the impact of dispersed chemical substances in the sea. The purpose of this study is to analyze the adaptation of the C14 Method to the marine species *D. labrax*. Toxicity tests were carried out exposing *D. labrax* to sodium dodecyl sulfate (SDS). This is an anionic surfactant widely employed in industry, agriculture, and domestic use and therefore it is found in abundance in the environment, particularly in the sea. Adaptations of the method are reported and results of growth response of *D. labrax* to the SDS are showed.

MO046

Evaluation of the detoxification mechanisms of metals in aquatic organisms by characterization of hepatic metallothionein (MT)

J.d. Azevedo, Federal University of Sao Paulo / Biological Sciences; J.E. Sarkis, IPEN - Nuclear and Energy Research Institute / Center for Chemical and Environmental Technology; S.O. Rogero, J.R. Rogero, IPENCNENSP Metallothionein (MT) has been thoroughly used as a biomarker of metals exposure. MT is a low-molecular-weight protein which has many sulfhydryl groups due to the large amount of cysteine in the molecule. These sulfhydryl groups bind a variety of metals and therefore, presumably, make them less toxic to other cellular constituents. However, biochemistry aspects of the protein as isoforms identification and quantification and their specific mechanisms of detoxification in sentinel species are, as yet, weak. Previous data showed that fish, such as the catfish *Cathrorops spixil*, are efficient bioindicator species to metals contamination in coastal aquatic areas under anthropogenic influence, for instance inputs of Pb, Cd, Hg, Ni, Fe, Zn, Cu and Mn. *C. spixil* is the most common catfish in the Brazilian coast and has a feeding habit mainly of materials and organisms upon the sediment, where the availability of contaminants is high. In order to understand intrinsic aspects of the detoxification process of toxic metals in sentinel species in natural and anthropogenic conditions, hepatic samples of *C. spixii* were collected in a non-polluted (Cananea), as well as one polluted, estuary (Santos-São Vicente) and tested under different assays in order to establish an effective bioanalytical technique to purify the protein like-MT, identify and quantify the specific isoforms and the metal contents in the cell. Therefore, hepatic samples were submitted to ultracentrifugation, thermocoagulation and a chromatographic purification and identification of MT isoforms by size-exclusion-HPLC and anion-exchange-HPLC, respectively. The elution of the protein was made with online UV/Vis detection. Metals in hepatic cytosols were also quantified by HR-ICP-MS. Obtained results showed that the established protocol of bioanalytical technique was effective to purify MT-like protein by SE-HPLC and to identify its isoforms by AE-HPLC. With these analytical strategies, it was possible to identify two kinds of MT isoforms (MT-1 and MT-2) in the *C. spixil*. Fish MT-1 from polluted areas showed a strong linkage with the levels of Cu, Hg, Pb, Fe and Ni suggesting that the MT-1 is an effective biomarker of metal contamination.

MO047

Assessment of the environmental impact of the dumped chemical warfare agents at the Baltic Sea using caged blue mussels (*Mytilus trossulus*)

R. Turja, Finnish Environment Institute SYKE / Marine Research Centre; M. Brenner, Alfred Wegener Institute / Biosciences; J. Barsiene, University of Vilnius; K.K. Lehtonen, Finnish Environment Institute / Marine Research Centre Chemical weapons dumped into the sea after World War II possess growing concern for the marine environment; metal shells of different chemical munitions lying on the bottom are severely corroded and dangerous contents pollute the sediments. Chemical warfare agents (CWAs), such as mustard gas and various arsenic-based compounds (e.g., Clark I and Adamsite) and their degradation products have been detected in noticeable concentrations in sediments at the major dumping sites at the Baltic Sea. Blue mussel caging approach was applied to assess environmental impact of thousands of tons of CWAs at the main dumping site at the Bornholm Basin. Due to the patchy occurrence of the CWAs in the sediments mussel caging method was chosen to deploy the organisms exactly at sites where high CWA concentrations were detected in sediments and to one reference site. Biomarkers representing different biological functions including antioxidant defence, biotransformation, neurotoxicity, lysosomal membrane stability, geno- and cytotoxicity, cellular energy allocation and condition index were investigated. Moreover, tissue concentrations of different CWAs and the possible metabolic