PHD COURSE IN LIFE AND ENVIRONMENTAL SCIENCES

Report Form for PhD student annual evaluation (XXXVI and XXXVII cycles)

Name of PhD student: Matteo Fanelli

Title of PhD research: Biogeochemical cycling of contaminants in marine waters

Name of PhD supervisor: Silvia Illuminati
Research lab name: Chimica Analitica per l'Ambiente e gli Alimenti

Cycle:
[ ] XXXVI
[X] XXXVII

PhD Curriculum::
[ ] Marine biology and ecology
[ ] Biomolecular Sciences
[X] Civil and environmental protection

DISVA instrumentation labs/infrastructure eventually involved in the project:
[X] Actea Mobile Laboratory
[ ] Advanced Instrumentation lab
[ ] Aquarium
[ ] MassSpec lab
[ ] MaSBiC
[ ] Simulation/informatics lab
[ ] Other. Please, indicate: .....

ABSTRACT (1000 characters, including spaces):
Potentially Toxic Element, PTEs (e.g., As, Cd, Hg, Pb) are naturally occurring elements presents in the environment in very low concentrations. PTEs evaluation is a key parameter to reach the Good Environmental State (GES) according to the MSFD (Directive 2008/56/EC). The human activities and the globalization processes, with the introduction of elements far from their natural origin, altered the natural biogeochemical cycling of elements[1], resulting in many cases as stressor condition for the environment.

In my PhD project, to understand sources and alterations, PTEs are studied in the seawater, sediments, atmospheric depositions, and biota, in three different sites of Ancona coast. Here, methodology setup and preliminary results are reported. The first-months analysis of PTEs in the selected sites evidence a site-specific variability and, in some cases, PTEs values are close or above the quality standards.

Part 1. Scientific case of the PhD Research (2 to 3 pages, including figures)

- BACKGROUND

While the sampling and analytical techniques for the PTEs determinations in the environment have practically no issues and has been well described by many and many authors, the chemical composition of atmospheric depositions, sediments and seawater of the southern part of the Northern Adriatic Sea (NAS), in particular for the coastal region of the Marche, suffers a very huge lack of data. Furthermore, there are no studies concerning the metal apportioning given from atmospheric deposition in the marine-coastal environment for the Western part of the Adriatic Sea.

- SCIENTIFIC AIMS

This project aims to (1) chemically characterize the marine coastal area of Ancona; (2) to determine PTEs in different marine matrices (seawater, surface sediments and organisms); (3) to describe the possible influence of atmospheric pollution on the marine biogeochemical cycle of PTEs; (4) to evaluate the seasonal and inter-annual evolution of PTEs in the different matrices, (4) to identify the possible sources and the relationships or fluxes between the different matrices. The first year focused on the study of the best analytical approach and first data evaluation.

- WORKPLAN AND RESEARCH ACTIVITIES

From March 2022 samples of seawater, marine sediments and atmospheric depositions have been collected each month along the coastal area of Ancona. Three sites with different anthropogenic impact and environmental features were individuated along the coast of Ancona (Figure 1), in three different area: 1) Palombina (Pal), characterized by low hydrodynamics, very shallow waters (<1 m), the proximity of API refinery, Esino River and the presence of bathers in the summer season; 2) Marina Dorica (MD), close to the Ancona port, characterized by intense maritime traffic and close to industrialized areas and 3) Portonovo (PN), two miles far from the coastline, characterized by the absence of direct anthropogenic impact or industries proximity, except for the maritime traffic, particularly intense during the summer season.

Fig. 1. Sampling points, Coast of Ancona, Marche, Italy.

WP1. Seawater

Chemical characterization of seawater along the coast of Ancona

Methods. Seawater samples were monthly collected from March 2022 (Fig. 1). Surface sea samples are collected manually by pre-cleaned HDPE bottles. Bottom waters are sampled only in the Portonovo site through a Niskin bottle and then preserved in HDPE bottles. Once collected, seawater samples were vacuum filtrate through 0.45-µm MCE filters in order to determine the dissolved (M_{diss}) and the particulate (M_{part}) concentrations of PTEs. The filtrated solution is acidified with ultrapure HCl at 2% v/v, while MCE filters were stored at −20°C. Dissolved As, Cd, Cu, Hg and Pb are determined by Atomic Spectroscopy Fluorescence (AFS)[2]; for the other PTEs (Al, Cr, Fe, Mn, Ni, Se, Pb) an analytical procedure for seawater determination by Atomic Absorption Spectroscopy (GF-AAS) will be set-up. Particulate PTEs are determined by GF-AAS, previous acid microwave assisted digestion of MCE filters.

Expected/Obtained Results: In this first year of my PhD project preliminary results on dissolved Cd and Hg are reported. Cd_{diss} and Hg_{diss} show generally low concentration in Portonovo, with respect to the other sites (Fig. 2). Marina Dorica resulted as the most polluted site for Cd and Hg, while Palombina Cd is comparable in some case to MD and for Hg comparable with PN. No statistically significative differences are observed between surface and bottom waters of PN for both metals. Concentrations determined are below the Maximum Acceptable Concentration (MAC-EQS) included in MSFD (Marine Strategy

Framework Directive) for dissolved Cd and Hg, respectively of 450 and 70 ng L^{-1}, anyway Hg_{diss} concentrations found in MD site are quite close to the MAC-EQS.

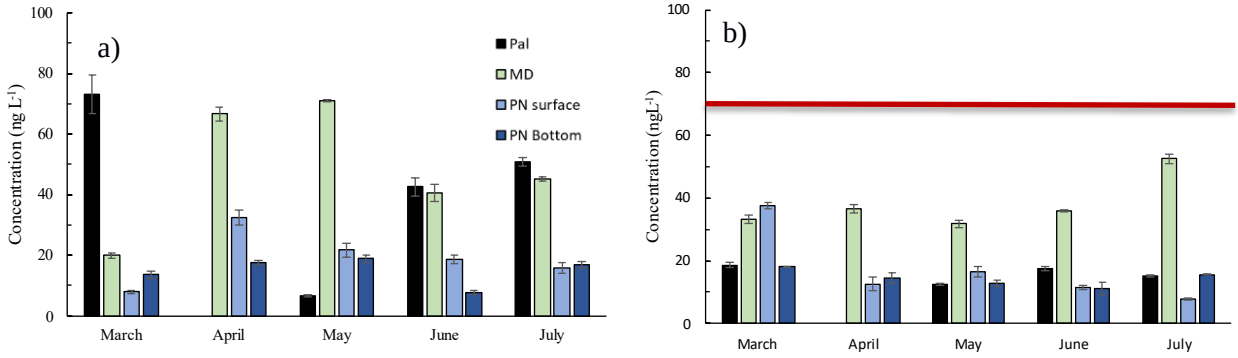

Fig. 2 Seawater dissolved Cd (a) and Hg (b) concentrations. Red line in b) represents the MAC-EQS

WP2. Marine sediments

Chemical characterization of sediments along the coast of Ancona

Methods. Samples of superficial sediment have been collected once a month from March to September 2022 in the study area, by manual core in Palombina and by grab sampler in Portonovo and Marina Dorica. Sediment samples were subjected to freeze drying lyophilization and subsequently homogenization processes. About 0.15 g was used for the Mercury quantitative determination through Directly Mercury Analyzer (DMA). Another aliquot (~0.1 g) was subjected to microwave-assisted acid digestion by adding 3 mL of suprapure HNO_3, 1 mL of suprapure HCl and 1 mL of suprapure HF^3. Metal concentrations were determined in the digested solutions by GF-AAs (for Al, As, Cd, Cr, Ni, Pb) and FAAS (Fe, Mn, Zn). The accuracy of the analytical procedure was evaluated by analysing two Certified Reference Materials (CRM PACS-2 and MESS-2), processed using the same procedure of sediment samples. All recovery % resulted above 90%.

Expected/Obtained Results: Results of chemical analysis for Al, As, Cd, Cr, Fe, Hg, Mn, Ni, Pb, Zn are reported in Table 1.

Table 1: Metal concentrations in sediment samples. Results are reported as mean ± standard. Environmental Quality Standard for coastal marine sediments included in D.Lgs 13/10/2015 n.172 is also reported for each metal, when available.

Site	Metal concentration									
	mg/g			µg/g						ng/g
	Al	Fe	Mn	As	Cd	Cr	Ni	Pb	Zn	Hg
Palombina	6.4±4.3	5.9±0.3	0.81±0.02	7.1±0.4	0.55±0.14	9.4±0.5	14±0.4	9.2±0.8	13±0.1	3.7±0.1
Marina Dorica	7.4±3.5	1.4±1.3	1±0.1	14±5	0.76±0.01	17±3	37±10	15.6±2.2	67±5	15±2
Portonovo	4.6±0.8	19.6±1.1	0.91±0.02	13.4±0.8	0.79±0.04	24±3	57±5	19.0±0.9	92±3	43±9
EQS				12	0.3	50		30		300

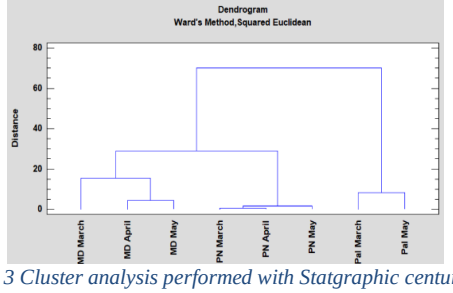

Fig. 3 Cluster analysis performed with Statgraphic centurion

As evident from Table 1 and the cluster analysis (Fig. 3), there is a significant difference between the chemical composition of the three sites. Unexpected higher concentrations of Hg, Ni and Pb were measured at Portonovo with respect to the other two sampling sites. This may be due to a different granulometric composition of Portonovo sediments, constituted by silt/clay or mud with respect to Palombina/Marina Dorica where sand is prevalent.

WP 3. Atmospheric Depositions

Chemical characterization of atmospheric deposition in Ancona

Methods. From July 2021, atmospheric depositions were collected by means of passive samplers (sampling bulks) in only one site of the study area (Marina Dorica). An additional sampling site was considered in the first part of my PhD project (campus of Università Politecnica delle Marche) to evaluate different emission sources. The bulks are made of a PE bottle (volume, 10 L) with a PE funnel (volume, ~10 L; surface area = 0.034 ± 0. 002 m^2) following the EN 15841:2009. Each sampling bulk was placed at 1.5 m above the ground, avoiding the collection of resuspended soil particles. Samples were shaken, and vacuum-filtered through pre-weighed 0.45-µm membrane filters to separate the soluble and insoluble fractions. A second pre-weighed mixed cellulose esters (MCE) filter was used to "clean" the inner surfaces of the funnel to collect any particles eventually remained on it. The washing solution was further filtered by using the same filter of the deposition. The total particle load from atmospheric deposition were measured according to the gravimetric method, following a procedure previously set-up and optimized[4]. Soluble and insoluble metal fractions were determined by analyzing filtered and residual fractions. A quarter of each bulk-set filters (¼ filter with residue + ¼ filter with particulate from the funnel walls) was microwave (MW) digested in Teflon bottles, after addition of ultrapure HNO_3+H_2O_2+HF, 5+2+1 mL. Digested solutions were analyzed by GF-AAS for metal determination. The total mercury content in residual fractions was quantified by Direct Mercury Analyzer (DMA). Soluble concentrations of metals were determined by AFS previous sample acidification with ultrapure HCl 2% v/v.

Expected/Obtained Results: In this first-year report of my PhD preliminary results of soluble metal fluxes in atmospheric depositions are reported. For the fluxes evaluation, soluble mercury (Hg_{sol}) and Cadmium (Cd_{sol}) concentrations determined by AFS were related with the bulk surface area, the filtration volume and sampling days. Results are reported in Fig. 4. The fluxes resulted generally higher in the port of Ancona (CNR) than those recorded at the UNIVPM campus, especially for Hg_{sol}, which showed results about 3 times higher. This could be related to the proximity of the industrial area in Ancona port. Accuracy tests have been performed using Certified Reference Material (CRM), recovery = 91%. Limits of Detection has been evaluated through calibration curve method: 6.1 ng/L for Hg and 3.5 ng/L for Cd.

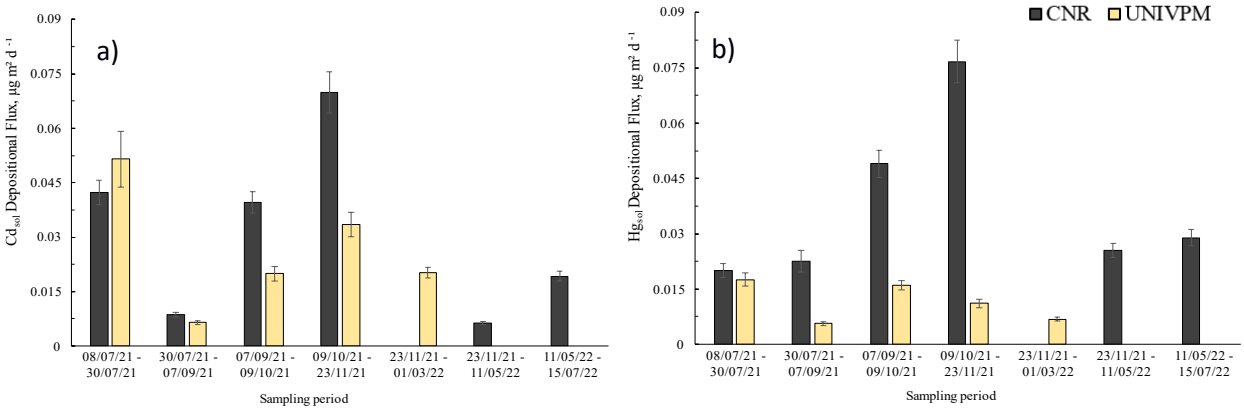

Fig. 4 Depositional fluxes for Soluble Cd (a) and Soluble Hg (b) in CNR and UNIVPM bulk samplers, expressed as µg m^2 d^{-1}.

Future perspectives:

Further studies on the chemical characterization of the coastal area of Ancona are necessary to obtain information on the different sources and anthropogenic inputs of metals. The second year of my PhD project will focus on:

- Metal determination in all the matrices studied
- Macronutrient evaluation in seawater and atmospheric depositions
- Microbiome analysis in atmospheric depositions
- Assessment of metal depositional fluxes (both soluble and insoluble)

- Physical-chemical characterization of marine sediments
- Metal concentrations in target organisms (sponges)
- Statistical analysis and Chemometric data treatment to evaluate source apportionment of metals in the study area

- REFERENCES

1. Rauch, J. N., & Pacyna, J. M. (2009). Earth's global Ag, Al, Cr, Cu, Fe, Ni, Pb, and Zn cycles. *Global Biogeochemical Cycles*, *23*(2).
2. Girolametti, F., Fanelli, M., Ajdini, B., Truzzi, C., Illuminati, S., Susmel, S., ... & Annibaldi, A. (2022). Dissolved Potentially Toxic Elements (PTEs) in Relation to Depuration Plant Outflows in Adriatic Coastal Waters: A Two Year Monitoring Survey. *Water*, *14*(4), 569.
3. GÜVEN, D., & Akinci, G. (2011). Comparison of acid digestion techniques to determine heavy metals in sediment and soil samples. *Gazi University Journal of Science*, *24*(1), 29-34.
4. Illuminati, S., Bau, S., Annibaldi, A., Mantini, C., Libani, G., Truzzi, C., Scarponi, G., (2016). Evolution of size-segregated aerosol mass concentration during the Antarctic summer at Northern Foothills, Victoria Land. Atmos. Environ. 1254. GÜVEN, D., & Akinci, G. (2011). Comparison of acid digestion techniques to determine heavy metals in sediment and soil samples. *Gazi University Journal of Science*, *24*(1), 29-34.

Part 2. PhD student information on the overall year activity (courses/seminars/schools, mobility periods, participation to conferences)

List of attended courses/seminars/schools

1. Technology transfer and innovation A.A.2021/2022 (curricular)

2. Design of Research: European projects. A.A. 2021/2022 (curricular)

3. VI Scuola di Monitoraggio Ambientale "i Siti Contaminati", Online, 24-26 Novembre 2021. Società Chimica Italiana, Divisione di Chimica dell'Ambiente e dei Beni Culturali

List of periods spent abroad

1. None.

List of conferences/workshops attended and of contributions eventually presented

1. X Convegno Nazionale sul Particolato Atmosferico 2022, Bologna 18-20 Maggio 2022. "Seasonal evolution of atmospheric aerosol in the city of Ancona". Annamaria Falgiani*, Flavio Vagnoni, Sébastien Bau, Matteo Fanelli, Federico Girolametti, Giada Giglione, Behixhe Ajdini, Cristina Truzzi, Anna Annibaldi, Silvia Illuminati. Oral presentation by A. Falgiani.

2. Ninth International Symposium – Livorno 14-16 June 2022. Monitoring of Mediterranean coastal areas: problems and measurement techniques. "Water column phosphatase activity assessment in a marine coastal environment and its relationship with rain events" M. Fanelli*, F. Girolametti, B. Ajdini, C. Truzzi, S. Illuminati, A. Annibaldi, C. Totti, S. Accoroni. Poster presentation.

3. Congresso ABC – XIX Congresso Nazionale della Divisione di Chimica dell'Ambiente e dei Beni Culturali – Torino, 20-23 giugno 2022 "Vertical distribution of Hg in sediment cores of the western-central and southern Adriatic Sea", M. Fanelli*, A. Annibaldi, C. Cerotti, F. Girolametti, B. Ajdini, S. Illuminati, E. Prezioso, C. Truzzi, R. De Marco, E. Frapiccini, A. Gallerani, M. Tramontana, G. Baldelli, F. Spagnoli. Oral presentation by M. Fanelli.

4. XXIX Congresso della Divisione di Chimica Analitica della Società Chimica Italiana (SCI), Milazzo 11-15 September 2022. "Influence of the Antarctic Convergence on the distribution of fatty acids associated with surface marine suspended particulate matter". F. Girolametti, S. Illuminati, A. Annibaldi, F. Ardini, M. Fanelli, B. Ajdini, C. Truzzi. Poster presentation.

5. VII MS Food Day, Florence 5-7 October 2022. Omega-3 enriched house cricket (Acheta domesticus) as novel and eco-sustainable food product in Europe. Behixhe Ajdini, Irene Biancarosa, Silvia Illuminati, Anna

Annibaldi, Federico Girolametti, Matteo Fanelli, Francesca Tulli, Gloriana Cardinaletti, Cristina Truzzi. Poster presentation.

6. Partecipation as exhibitor with UNIVPM-DISVA at Sealogy 2021, 18-20 November 2021.

Part 3. PhD student information on publications

List of publications on international journals

J1. Fanelli, M.; Girolametti, F.; Truzzi, C.; Illuminati, S.; Ajdini, B.; Susmel, S.; Celussi, M.; Šangulin, J.; Annibaldi, A. Impact of Depuration Plants on Nutrient Levels in the North Adriatic Sea. *Water* **2022**, *14*, 1930. https://doi.org/10.3390/w14121930.

J2. Girolametti, F.; Fanelli, M.; Ajdini, B.; Truzzi, C.; Illuminati, S.; Susmel, S.; Celussi, M.; Šangulin, J.; Annibaldi, A. Dissolved Potentially Toxic Elements (PTEs) in Relation to Depuration Plant Outflows in Adriatic Coastal Waters: A Two Year Monitoring Survey. *Water* **2022**, *14*, 569. https://doi.org/10.3390/w14040569.

List of publications on conference proceedings

C1. None.

List of other publications (books, book chapters, patents)

B1. None.

14/10/2022

Student signature

Supervisor signature