



Joint IAATO/COMNAP Antarctic Fellowship Report

In partnership with COMNAP

Title of Fellowship Project:

Understanding the consequences of microplastics introduction in the Antarctic environment: the Plastisphere Resistome.



Miguel González-Pleiter taking samples for this project in Lake Uruguay. Photo: Evelyn Krojmal.

Name of Fellow:

Joint IAATO/COMNAP Antarctic Fellowship 2020

Home Institution:

Univerisdad Autónoma de Madrid (UAM)

Host Institution:

Universidad de la República Uruguay (Udelar)

International and national project collaboration:

This fellowship allowed me to participate in the Antarctic Campaign 2021-2022 thanks to the collaboration with the Uruguayan Antarctic Institute (IAU) through the principal

investigators of the project “Plastics and microplastics in marine-coastal areas of the Fildes Peninsula, King George Island (Antarctica)” (AntarPLAST), which is led by Uruguayan researchers Franco Teixeira de Mello (Udelar and *Centro Universitario de la Región Este*; CURE) and Juan Pablo Lozoya (Udelar and *Centro Interdisciplinario Manejo Costero Integrado del Cono Sur*; C-MCISur). In addition, this project has allowed me to establish new collaborations with other Uruguayan researchers that resulted in the international research stays of three Uruguayan researchers (Evelyn Krojmal, Barbara De Feo and Camila Vidal) in my laboratory (*Universidad Autónoma de Madrid*, Spain) this year. I would also like to point out the collaboration with the Spanish Polar Committee (CPE) the support allowed me to transport the material to the Artigas Scientific Antarctic Base and the samples to Spain as well as the return flight from Antarctica. Finally, this project has allowed me to meet the APECS Spain group and to be part of it.

Dates of Activity:

Home quarantine in Uruguay: from January 3, 2022 to January 11, 2022

Institutional quarantine in Uruguay: from January 12, 2022 to January 27, 2022

Artigas Scientific Antarctic Base: from January 28, 2022 to February 12, 2022

Research objectives of the project (~ 200 words):

The main objective of the project was to investigate the antibiotic resistome, the collection of all the antibiotic resistance genes (ARGs), in the Antarctic freshwater plastisphere to understand the consequences of the introduction of (micro)plastics (MPs) in the Antarctic environment as MPs may be vectors of ARGs. The study was carried out in two freshwater lakes (Uruguay Lake and Ionosferico Lake) located in the Northeast of Fildes Peninsula (King George Island, Antarctica). The proposal was divided into four sections, which have the following objectives: 1) to collect, quantify and identify MPs in water, sediments and/or the shore of two freshwater lakes; 2) to detect the presence of ARGs on the collected MPs using SmartChip Real-Time PCR System (TAKARA) and to compare their abundance with that found in the bacterial populations of each lake (water, sediments and sand of the shores); 3) to identify the antibiotic resistance bacteria (ARB) attached to MPs collected to correlate with the presence of ARGs; and 4) to analyze the occurrence of antibiotics in water, sediments and sand of the shores of each lake and in the MPs collected to try to correlate with the presence of ARB/ARGs.

Methods and Activities:

What was the nature of the research and activities undertaken? Did everything go as you and your host had hoped? What results were generated and how do they reflect expectations?

In Antarctica, we collected MPs in water, sediments and/or the shore of two freshwater lakes. We used the same methodology that we have employed in previous studies. The samples had their respective blank control and procedural control. Regarding this sampling, we had problems collecting the smaller fractions in water and sediment due to their low concentration. Furthermore, contrary to our expectations, it is worth noting that the highest concentration on the shores of the lakes was of plastics and the lowest concentration of microplastics. Therefore, we

obtained more samples of plastics (which could eventually become microplastics) than of microplastics. Samples will be stored at -20°C.

Once the samples stored at -20°C arrived in Spain aboard the BIO Hesperides (Spanish polar research vessel) in mid-June 2022, DNA extraction from MPs started. Several challenges had to be overcome to obtain the quantity and quality of DNA needed for the ARG analysis. To solve this, several extraction methods and DNA extraction kits were tested until we found one with which we obtained the required DNA qualities and quantities in mid-October 2022. It should be noted that due to the timing (summer) some of the kits we needed took a long time to be delivered. At the same time, I kept in contact with TAKARA to carry out the analysis because the laboratory in Finland that offered this service closed. Finally, I was able to find another laboratory to perform the analysis in France (La Plate-forme Génomique EcogenO, Université de Rennes). At that time, I had also some problems finding a company to ship the DNA from Spain to France and to pay for the analysis. Finally, I sent the samples in November 2022, and I received the first results in December 2022. However, in mid-December 2022, the laboratory in France has informed me that I had had troubles with the SmartChip Real-Time PCR System, and they would not be able to send me the rest of the results until January or February 2023. The results (I have so far) indicate the presence of mobile genetic elements (MDR-mobile, integrase and plasmids) and antibiotic resistance genes (aminoglycoside and fluoroquinolone) in the bacterial populations of Antarctic lakes. I am waiting results from the rest MPs collected in both Antarctic lakes.

Project outcomes and achievements:

What do you feel were the significant outcomes in terms of the research but also in terms of personal development?

We carried out the analysis using the SmartChip Real-Time PCR System that performs high-throughput screens of antibiotic resistance genes. Therefore, we believe that our main results will be to describe for the first time the resistome of the Antarctic plastisphere. Once we know the resistome of the Antarctic plastisphere, we will be able to compare it with that of their surrounding environment. This will help us to understand the role of the MPs in spreading ARGs in pristine ecosystems.

Publications, Presentations and Products:

Are there papers or articles submitted or in preparation as a result of the Project? Have you made presentations as a result of the Fellowship? Are there significant products as a result that will have use beyond the Fellowship for yourself or others?

I am currently waiting for the rest of the results. Once we have them, we will submit a scientific publication of the first resistome of the Antarctic plastisphere in a journal in Q1. This scientific publication includes the identification of the MPs that we are performing using micro-FTIR and the identification of the antibiotic resistant bacteria that we will grow in different culture media using the antibiotic detected in the water and sediments. Also, this fellowship allowed me to establish collaborations with

Uruguayan researchers that have led me to participate as an author in three presentations at the international conference MICRO 2022.

Capacity Building, Education and Outreach Activities:

As a result of the Fellowship did you engage in educational and/or outreach activities before/during/after your visit? Did you meet with students to explain your work? Did you give a public lecture? Was there any publicity about your visit - either through your host or home institute?

As a member of APECS Spain, I participated in the cycle of conferences of scientific divulgation “Café con Hielo” in March 2022. Furthermore, I have told my Antarctic experience in two schools in Spain to students from 8 to 16 years old.

Future Plans and Follow-ups:

Do you plan to continue contact with the host institute and others you met as a result of the project? What will be the nature of the future work?

This fruitful collaboration has led to the fact that we are looking forward to expanding our research collaboration in the Antarctic Campaign 2022-2023 and the future with several Uruguayan researchers.

Personal Impact:

How do you feel the Fellowship has and will continue to impact your research and career objectives? What was the main impact for you personally?

Infections caused by microbial carrying antibiotic resistance genes will overcome cancer deaths by 2050. However, the fate and transport of antibiotic resistance genes around the world are poorly known. My main goal over the next five years is to understand how antibiotic resistance genes travel around the world and enter the food chain until they reach humans. My hypothesis is that MPs could play a key role in the spreading of the antibiotic resistance genes. This fellowship has helped me to take the first steps to understand how antibiotic resistance genes reach and travel through pristine regions.

Acknowledgements and References:

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