Mar del Plata, Argentina

September 29, 2025

Dear Rufford Foundation Team,

Through this letter, I would like to explain how the project titled “A Non-Lethal Tool for Monitoring Fish Health of Wetlands From South Pampas in Argentina” is progressing.

As we mentioned in a previous email message, the sampling campaign was re-scheduled for January–February 2026. Over the past few months, we have started working on tasks related to the project´s third objective and have begun purchasing the materials that we will require at a later stage. Since the waterbodies are located in different municipalities, we have also initiated communication with the municipalities in relation to the third objective:

1) **General Pueyrredón:** First, we have had a meeting with the Environmental Management Director of the Municipal Entity of Urban Services of General Pueyrredón, Eng. Marcelo Ragonesse. In this first meeting, we explained to him what The Rufford Foundation is and what the objectives of the project are. He showed interest about the potential risks to the bodies of water selected for the project and informed us of some municipal resolutions on wetlands and the limitations they impose on work in these areas. He found our work interesting, and has now realised that our data could be used to improve management in these areas. After the meeting, the possibility was left open to carry out some activity with the fauna department within the framework of an agreement already established between the University and the Municipality.

From left to right: Myself (Lic. Gastón Ojeda), Dr. Mirta Menone (Director of my PhD thesis) and Eng. Marcelo Ragonesse (Environmental Management Director of the Municipal Entity of Urban Services of General Pueyrredón)

2) **Mar Chiquita:** On the other hand, we have also established connections with the Director of the Environmental Department of Mar Chiquita, Lucía Bogarín. Our first contact was made during the "Second Participatory Workshop of the Mar Chiquita Reserve" held on April 28. In this context, we recognize the importance of the Mar Chiquita Municipality in relation to environmental policies and the concern of local communities for their environment. Following this initial contact, we held another meeting with Lucía Bogarín, during which we explained what The Rufford Foundation is and outlined the objectives of the project. After that, we scheduled a meeting with the municipal technicians and the residents of La Caleta town, a community living near Los Cueros Creek.

The meeting was held in the Visitor Information Center of Mar Chiquita and it was very fruitful. Both the technicians and residents expressed strong interest in the conservation of Los Cueros Creek. They showed interest not only in the project but also in the broader objectives of The Rufford Foundation; even they showed interest in further studies on contamination and species conservation in the area. They shared their questions and inquiries about the project, the species inhabiting Los Cueros Creek, the non-lethal techniques we will use, and how they could participate and learn these tools. We established a date to organize a laboratory practice an the National University of Mar del Plata, during which we will explain how we will sample and how to proceed with these species.

Below, I have attached some pictures of that day:

Visitor Information Center of Mar Chiquita. The place where the meeting took place.

Presentation of the project with technicians and residents of La Caleta. At the front: myself (Gastón Ojeda) explaining the objectives of the project and the role of the Rufford Foundation. Seated: technicians and residents of La Caleta, invited by the Environmental Department Director of Mar Chiquita, Lucía Bogarín.

Participants of the first meeting with Mar Chiquita teamwork. On the left: myself (Gastón Ojeda). In the center: Mirta Menone, director of my PhD thesis. On the right (with the squared scarf): Lucia Bogarin, Environmental Department Director of Mar Chiquita.

In addition to the mentioned work, we conducted a study to determine the appropriate concentration of Eugenol for the effective anesthesia of fish, a necessary preliminary step for the development of the project. This compound will be used as anesthetic in the field sampling of my project, in order to minimize the stress in the fish. Eugenol is a natural anesthetic widely used in veterinary medicine and offers several advantages: it is of natural origin (extracted from clove oil), and possesses antiseptic and vasodilatory properties, which make it particularly suitable for non-lethal blood extraction. We prepared a scientific poster based on this information, which we are presenting at the XXIV Congress of the Argentine Toxicology Association (ATA) this week. This information enables us to perform blood extractions in accordance with an appropriate ethical framework for animal research.

Adult *Australoheros facetus* anesthetized with Eugenol. Under this degree of anesthesia (deep anesthesia), the fish exhibited only opercular movements and did not experience stress from stimulation or handling, which allowed us to collect blood and mucus samples for the determination of health biomarkers without compromising their health.

Adult *Australoheros facetus* recovered from anesthesia. These specimens are form the laboratory stock.

We have also begun acquiring the necessary materials for the project. However, we must inform you that we are facing difficulties in importing the “Vitellogenin ELISA Kit,” which had been included in the

approved budget. This item is appropriated for the quantification of VTG protein in blood samples. In Argentina, to import research-related items without paying extra costs and taxes, it is necessary to submit a request through ROECyT (Register of Science and Technology Entities and Organizations). The current government has reassigned the personnel responsible for processing these requests, and the system has not been operating for the past four months. These requests always take six months to be approved. We discovered this situation when we attempted to initiate the request in May. We explored alternative solutions, but the only available option is to purchase the kit through a custom courier, who charges four times the original price—an expense that compromises the entire project budget. We regret to inform you that we do not consider acquiring this item in the near future. This item has been suggested by one of the Referees of my application, and although we have considered, at this point it is not viable. Nevertheless, we deem it necessary that you are aware of this situation. However, we are not giving up; we will seek to replace this information with another fish health indicator. We have started thinking about some options and will inform you of our choice of new biomarker in future updates.

After all the mentioned information, we would like to emphasize that we are truly pleased to carry out this project. The opportunity to work directly with local communities, as well as the insights gained about how society deals with these issues, are invaluable to my PhD research. We are genuinely grateful for this opportunity.

Regards,

Lic. Gastón Ezequiel Ojeda Rossi