

### The Rufford Foundation Final Report

Congratulations on the completion of your project that was supported by The Rufford Foundation.

We ask all grant recipients to complete a Final Report Form that helps us to gauge the success of our grant giving. The Final Report must be sent in **word format** and not PDF format or any other format. We understand that projects often do not follow the predicted course but knowledge of your experiences is valuable to us and others who may be undertaking similar work. Please be as honest as you can in answering the questions – remember that negative experiences are just as valuable as positive ones if they help others to learn from them.

Please complete the form in English and be as clear and concise as you can. Please note that the information may be edited for clarity. We will ask for further information if required. If you have any other materials produced by the project, particularly a few relevant photographs, please send these to us separately.

Please submit your final report to jane@rufford.org.

Thank you for your help.

#### Josh Cole, Grants Director

Grant Recipient Details							
Your name	Dalia Carolina Barragán Barrera						
Project title	Studying the ecological isolation and physiological response to anthropogenic stressors of a vulnerable bottlenose dolphin population in Bocas del Toro, Panama						
RSG reference	23298-D						
Reporting period							
Amount of grant	£10000						
Your email address	daliac.barraganbarrera@gmail.com						
Date of this report	December 5, 2018						



## 1. Please indicate the level of achievement of the project's original objectives and include any relevant comments on factors affecting this.

Objective	Not achieved	Partially achieved	Fully achieved	Comments
Conduct outreach activities in in several local Institutions for discussion about research activities and engage local stakeholders in management steps.				<ul> <li>Outreach activities included several talks that we conducted with local communities:</li> <li>01/Mar/18: Teacher Science training programme. Smithsonian Institution, Bocas del Toro Station.</li> <li>03/Apr/18: Guabito Middle School. Changuinola, Bocas del Toro</li> <li>03/Apr/18: University of Panama. Changuinola, Bocas del Toro.</li> <li>28/May/18: Public talk. Smithsonian Institution, Bocas del Toro Station.</li> <li>04/Jun/18: ISAE University. II International Congress of Social Science. Changuinola, Bocas del Toro.</li> <li>8/Jun/18: Teacher Science training programme. Smithsonian Institution, Bocas del Toro.</li> <li>8/Jun/18: Teacher Science training programme. Smithsonian Institution, Punta Culebra Station.</li> <li>28/Jun/18: Public talk. Environmental Ministery and International Maritime University of Panama.</li> </ul>
Determine ecological isolation of bottlenose dolphins in Bocas del Toro comparing isotopic niche width between samples from Bocas del Toro and neighbouring areas.				We conducted surveys within the archipelago and surrounding areas of Bocas del Toro in March and April. During this survey, a total of 91 animals were counted in 27 sightings. We collected a total of six new biopsies of skin and blubber. We conducted new isotopic analyses using old and new samples from bottlenose dolphins collected in Bocas del Toro. We compared these isotopic signatures with samples from dolphins collected in neighbouring areas. For this, we analysed carbon and nitrogen isotopes to assess niche for each



	population. The isotopic analysis, which will be published soon, indicates that dolphins in Bocas del Toro belongs to the "inshore form", since the prey selection is broad in terms of coastal items, which show low mercury concentrations.
Collect samples from individuals in Dolphin Bay to quantify dolphin stress levels in relation to dolphin-watching, through cortisol analysis.	During two fieldtrips conducted this year we have collected six new samples available to assess hormone analyses. In total we have collected 26 blubber samples of bottlenose dolphins in Bocas del Toro to conduct hormone analyses related to stress. We used an Enzyme Immunoassay (EIA) kit (Arbor Assays, Ann Arbor, MI, USA) to measure the levels of hormones concentrated in the blubber of each animal. Preliminary analyses showed that dolphins increased stress hormones (cortisol) during interactions with multiple boats (high touristic season).

## 2. Please explain any unforeseen difficulties that arose during the project and how these were tackled (if relevant).

Our main problem during field trips was related with dolphin collection, since animals were more evasive, and therefore skin samples collection were harder. For this reason, we decided to conduct our laboratory analyses with samples already collected in order to achieve with proposed goals.

In the lab we also had some problems mainly with hormone work. Although we used all blubber tissue from each sample to conduct hormones analyses, some of them were not enough to extract effectively all hormones. For this reason, we decided to optimise the samples and conduct hormone analyses not only using cortisol, but also including other hormones such as reproductive ones, which were effectively extracted from all samples.

#### 3. Briefly describe the three most important outcomes of your project.

a) Community outreach. We highlight the interest of local communities for dolphin conservation in Bocas del Toro. This year we can involve more Panamanian academic institutions, since of our long-term plan in conforming a local research program to monitor dolphins in the archipelago. The involvement of local community has been crucial to conservation, but we need that local government manage effectively the situation of dolphins in the archipelago. Therefore, talks with Environmental Ministry was one of the most important outcomes in outreach



activities, as one step to establishment of a dolphin centre in Dolphin Bay as one measure for dolphin conservation in Bocas del Toro.

**b) Isotopic analyses**. The bottlenose dolphin population from Bocas del Toro is exposed to high disturbance levels related to boat traffic. The main concern with this situation is this population shows high philopatry and genetic isolation in the Caribbean. If this population decreases, a unique coastal linage will be lost. However, we need more evidence that supports the isolation and coastal habits of this population. Isotopic niche width comparisons with surrounding populations indicated that dolphins in Bocas del Toro effectively belongs to the "inshore form", since the prey selection is broad in terms of coastal items, which show low mercury concentrations. These results have high implications for dolphin's conservation, since we confirm coastal habits of this unique linage in the Caribbean. For this reason, these findings will be presented to stakeholders as review, including all data that we have generated until now, in order they know current conservation situation of these dolphins. We expect that all these scientific data, will be taking account to manage and change local conservation status of this bottlenose dolphin population in Panama.

c) Hormone analyses. Previous work indicated that dolphins have affected their distribution, behaviour, and communication due to exposition to high disturbance levels related to boat traffic. Consequently, dolphins in Bocas del Toro can be exposed to high levels of stress. We validated, extracted and measured four steroid hormones from each of the 26 samples: cortisol, corticosterone, progesterone and testosterone. Our preliminary data showed that cortisol hormones tend to be higher during the high season of tourism compare with the low season (figure 1). High levels of this hormone are related to stress, so these findings agree with previous results that indicate boat traffic has negative effects on individual's health. We are planning to go back to the field the next year in order to get more samples to confirm statistically the stress via cortisol analyses; however, current findings will be the key to demonstrate negative effect of boat traffic on dolphins' health in Bocas del Toro.

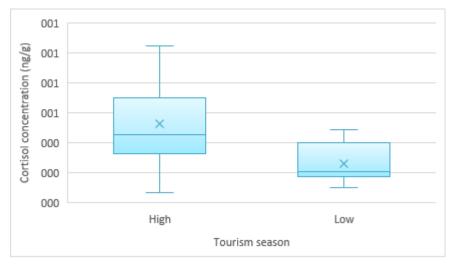


Figure 1. Cortisol concentration (ng/g) during the high (n=10) and low (n=6) tourism season.



## 4. Briefly describe the involvement of local communities and how they have benefitted from the project (if relevant).

Involvement of local community for successfully dolphin conservation in Bocas del Toro is one of the most important outcomes of our project. Our long-term plan is establishing a local research programme. For this season, we conducted talks in several local institutions such as:

- Smithsonian Institution-STRI: To show our main findings to scientific and local community.
- The Gabito Middle School University of Panama, Changuinola University: To involve Panamanian students in dolphin research.
- Environmental Ministry: To show main findings to governmental institution in order they priorities management plans for conservation of Bocas del Toro dolphin population.
- Teacher Science training programme: To discuss with local teachers how we can help to conserve this unique dolphin population.

We highlight the link created with Environmental Ministry as one of the most important outcomes in outreach activities, in order to establish effectively a dolphin centre in Dolphin Bay as one measure for dolphin conservation in Bocas del Toro.

#### 5. Are there any plans to continue this work?

Absolutely! As part of our long-term goals, we plan in the long-term:

- Creation and establishment of a dolphin centre, which will be located in Dolphin Bay in order to control the number of boats interacting with dolphins. For this, it is needed to continue with monitoring programme within archipelago, and outreach activities presenting dolphin situation to Environmental Ministry, since this is the most important environmental institution in Panama.
- Maintain the long-term research programme involving local institutions. It is needed to conduct abundance analyses to assess if this dolphin population has increased during years, and collect more samples to assess effectively health status of individuals related to stress. With this data, we plan to publish a review paper in Spanish compiling all biological data and conservation status of bottlenose dolphin in the archipelago. This document will be presented to governmental institutions (such as Environmental Ministry) in order they present an addendum under resolution ANAM No 0051-2008 (Gaceta Panamá, 2008) to this dolphin population will be included in the Panamanian Red List. This will be first step to guarantee their conservation in the long term.

#### 6. How do you plan to share the results of your work with others?

The main source to share our results have been the outreach activities. We plan to still be working on with local community doing talks, workshops and forums. Additionally, we posted on social media (Facebook) our activities and important findings. Our main interest next year is working actively with Environmental Ministry,



so we plan to publish a review paper in Biología Tropical, an important local journal, in which we compile and present all our scientific data showing dolphin situation in Bocas del Toro as a scientific referent to stakeholders. As a scientific support to dolphin isolation in the Caribbean, we have submitted a new paper with our ecotoxicological results, which is entitled as "Foraging habits and levels of mercury in a resident population of bottlenose dolphins (*Tursiops truncatus*) in Bocas del Toro Archipelago, Caribbean Sea, Panama". Additionally, we plan to present our findings in two conferences: the RSG Conference in Galápagos (Ecuador) and the 2nd World Marine Mammal Science Conference in Barcelona (Spain).

### 7. Timescale: Over what period was The Rufford Foundation grant used? How does this compare to the anticipated or actual length of the project?

The funds from RSG were expected to be used in the field during three trips in a period of a year (15 days for each fieldtrip for 45 days in total) and in laboratory analyses which included isotopic and hormone measurements. All hormone analyses were conducted at the endocrinology laboratory of the University of Alaska Fairbanks, Juneau Centre under the guidance of Dra. Shannon Atkinson. Because we had to prepare the trip to analyse hormones, and due to sample collection was difficult, we decided conduct two fieldtrips and prioritise laboratory work. Consequently, we completed 30 days in the field, but we can complete both isotopic and hormone analyses in the estimated period. With funds saved by shorten our fieldtrips, we could cover additional funds related to use of material and reagents in hormone analyses not included in the initial budget. Additionally, we could cover residual cost of hormone analyses because we had additional funds from Smithsonian and McGill University.

## 8. Budget: Please provide a breakdown of budgeted versus actual expenditure and the reasons for any differences. All figures should be in £ sterling, indicating the local exchange rate used.

Item	Budgeted Amount	Actual Amount	Difference	Comments
Isotopic measurements	395	999	604	This value describes the cost of isotopic analyses, which include technical services of isotopic measurements for all samples collected. This value increased because individual cost of each sample increased, and because we had to include here cost of laboratory materials and reagents to prepare samples to isotopic analyses.



				RSG spent = 998.6
Hormone analyses (including Enzyme Immunoassay kit)	3,950	4,698 + 359 = 5,057	1,107	This value describes laboratory material, the hormone extraction kit (Enzyme Immunoassay kit), and reagents. This cost increased due to individual cost of reagents to extract additional hormones from all samples, since size of some samples were not enough to extract cortisol hormones. Therefore, we decided to optimize samples extracting other hormones useful to assess stress on individuals. We also included here the cost of liquid nitrogen to conserve samples to conduct these analyses. Residuals were paid with external funds. <b>RSG spent = £ 4,463 + £ 359 = £ 4,822</b>
Sexing analyses (PCR expenses)	118	197	79	This value describes laboratory material such as gloves, PCR tubes and tips used to prepare PCR mix. The value of these reagents and materials increased in Colombia due to changes in the TRM. <b>RSG spent = £ 197</b>
DNA extraction (including DNeasy kit QIAGEN)	232	398	166	This value describes DNA extraction kit. The value of this kit increased in Colombia due to changes in the TRM. <b>RSG spent = £ 398</b>
Digital Camera with Zoom Lens 75-300mm	1,777	1,777	0	This cost was the same because we already had this equipment. <b>RSG spent = £ 0</b>
Binoculars w/ Range	174	174	0	This cost was the same because we already had this equipment. <b>RSG spent = £ 0</b>
GPS Navigator	234	234	0	This cost was the same because we already had this equipment.



				RSG spent = £ 0
Research permits	103	103	0	This value describes the fee of permits for research. We did not pay this item with the Booster Rufford grant. <b>RSG spent = £ 0</b>
Travel Insurance (two people)	158	158	0	This value describes the travel insurance for the researchers. All of them paid for their own insurance. Therefore, we did not pay this item with the Second Booster Rufford grant. <b>RSG spent = <math>\pounds 0</math></b>
Facility fee (one person, Panamanian people are free)	355	355	0	This value describes costs of facility use in STRI station. This cost was the same because this fee has a standard price during the year. We did not pay this item with the Second Booster Rufford grant. <b>RSG spent = £0</b>
STRI Admin and Bench fee (one person, Panamanian people are free)	47	47 + 127	127	This value describes costs of administration in STRI station. The differences in the value was because we included here the cost of darts fee. Only this last fee was paid with RSG funds. <b>RSG spent = £ 127</b>
Research boat and gasoline	3,200	1,382	1,818	In this item we include both boat and gasoline because we did one payment to STRI for all. The differences in the values are because we were in the field less days (one month). <b>RSG spent = £ 1,382</b>
Boat operator	2,133	1,277	856	This value describes the payment of boat operator during the fieldtrip. The differences in the values are because we were in the field less days (one month). <b>RSG spent = £ 1,277</b>
Meals	1,422	500	923	This value describes food expenses for the researchers during 30 days. The differences in the values are because we



Lodging	1,066	799	267	were in the field less days (one month), and we saved money cooking at station. We did not pay this item with the Second Booster Rufford grant. <b>RSG spent = £ 0</b> This value describes accommodation for researchers during 30 days. The differences in the values are
	150	10		because we were in the field less days (one month) <b>RSG spent = £ 799</b>
Weekly transport to STRI in Bocas del Toro	158	60	98	This value describes daily transportation from Smithsonian Institute to downtown (roundtrip) in Bocas del Toro. Initial cost was estimated, so final cost was different. We did not pay this item with the Second Booster Rufford grant. <b>RSG spent = £ 0</b>
In-country air travel (Panama - Bocas del Toro, roundtrip - two people, three trips)	869	433	436	This value describes airplane tickets for two researchers in three trips to the study area. The differences in the values are because we conducted two trips. We did not pay this item with the Second Booster Rufford grant. <b>RSG spent = £0</b>
Flight from Colombia - Panama (roundtrip - one person, three trips)	551	472	79	This value describes airplane tickets for one researcher in their three trips to the study area. The differences in the values are because we conducted two trips. We did not pay this item with the Second Booster Rufford grant. <b>RSG spent = £0</b>
TOTAL	16,942	14,548	6,559	RSG spent = £ 10,000.6

#### 9. Looking ahead, what do you feel are the important next steps?

Ecological results found during this year provided more evidence that bottlenose dolphin population in Bocas del Toro is isolated and formed only by the "inshore" individuals. Additionally, preliminary hormone analyses indicated that boat traffic is



affecting individuals' health, so these dolphins are vulnerable to boat traffic. Although we plan to present these results to stakeholders next year, we recognize that we need to generate abundance data to establish if population has increase or not in order to IUCN and local government change the conservation status of this population. Additionally, we need to collect more samples to conclude hormone analyses. Therefore, next steps within the archipelago include maintaining the longterm monitoring with local students to generate more biological information that include:

- Population estimation to assess number of individuals present in Bocas del Toro.
- Reproductive parameters to determine reproductive success and calf mortality in Bocas del Toro.
- Collection of more samples from individuals during both high and lows touristic seasons to assess stress hormones in these biopsies.
- Finally, maintaining good links with Environmental Ministry in order they prioritise conservation of this bottlenose dolphin population.

# 10. Did you use The Rufford Foundation logo in any materials produced in relation to this project? Did The Rufford Foundation receive any publicity during the course of your work?

The RF logo was displayed in several materials that we distributed and presented in the community during outreach activities. Furthermore, RF logo was posted in our social media (webpage and Facebook). The logo was also displayed in poster and oral presentations in two scientific events: the XII Congreso de la Sociedad Latinoamericana de Especialistas en Mamíferos Acuáticos SOLAMAC – RT 18 and the V Congreso Colombiano de Zoología. Additionally, the RF was mentioned in the acknowledgment sections of the manuscript "Foraging habits and levels of mercury in a resident population of bottlenose dolphins (*Tursiops truncatus*) in Bocas del Toro Archipelago, Caribbean Sea, Panama", recently submitted in a peer-review journal.

## 11. Please provide a full list of all the members of your team and briefly what was their role in the project.

**Dalia C. Barragán-Barrera.** I was working in the field collecting data. My role in the project was focused on preparing samples to conduct isotopic measurements, and analysis of isotopic data as well.

**Betzi Pérez-Ortega.** She worked in the field collecting dolphins' samples. She also established links with local people and institutions such as Smithsonian Tropical Research Institute, Environmental Ministry and local Universities to conduct talks and outreach activities. Additionally, she conducted hormone extractions and posterior analyses on blubber samples.

**Chelina Batista.** Although she was not included in our initial proposal, she supported us actively collecting data in the field trips.



**Demetrio Georget.** Although he was not included in our initial proposal, his support as boat captain is very important to collect samples, since he has a lot of experience working with dolphins.

#### 12. Any other comments?

As I mentioned in previous reports, the support that generously the Rufford Small Grants has provided us allowed the generation of relevant data for conservation bottlenose dolphins in the Bocas del Toro Archipelago in Panama. Therefore, we are really grateful for all support and confidence that the RSG board committee has given us to achieve the most important outcomes in our project. We really appreciate your support in conservation projects in developing countries, since you are contributing to conservation of many species and ecosystems in our continent, and also you are empowering young Latin American researchers to follow their careers and promote conservation issues in developing countries. Thank you for all.



The PAXARMS system. ©Chelina Batista.





A dolphins group. ©Betzi Pérez





Evasive dolphin behaviour. ©Betzi Pérez.

Dolphins surfing the wave. ©Betzi Pérez.