

### The Rufford Small Grants Foundation

### **Final Report**

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

We ask all grant recipients to complete a Final Report Form that helps us to gauge the success of our grant giving. 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. 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	Jason Ray O'Bryhim
Project title	Combining Genetic and Sociological Techniques to Evaluate
	the Status of Shark Populations in Costa Rica
RSG reference	13921-1
Reporting period	October 20th 2013 – October 20th 2014
Amount of grant	£5990
Your email address	Jobryhim@masonlive.gmu.edu
Date of this report	December 1, 2014



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

	Not	Partially	Fully	
Objective	achieved	achieved	achieved	Comments
Use forensic genetic techniques to identify the types and quantities of shark species being caught in Costa Rican fisheries and sold at local markets.		X		I collected a total of 1,300 tissue samples from sharks being sold in the markets and caught in artisanal fisheries. This number is below what was originally estimated, but still provides a robust sampling of these locations and enough data to provide significant answers to the questions posed in this research. To date I have analysed approximately 1/2 of those samples and am on track to complete
Use a social survey to determine the knowledge, perceptions, and behaviours Costa Rican fishermen have toward sharks and legislation protecting sharks.			X	Attempted distribution of the social survey was conducted at six different artisanal fishing communities. Of the communities where sampling was attempted, one entire community refused to take any part in the social survey, while fishermen from several of the other communities also refused. This led to the collection of 73 social surveys. Despite the lower than expected social survey response, it is believed the surveys obtained will provide all the necessary data to answer the questions relating to this portion of the research.

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

Several difficulties did arise during the duration of this project. One such difficulty was the acquisition of the necessary permits allowing for the collection of shark tissue samples from the artisanal fishermen. The Costa Rican Government required a contract signed by all fishermen taking part in the research, via the donation of shark tissue samples. Obtaining signatures from these fishermen was very difficult for several reasons. First, the fishermen distrust the government and conservation organisations, which they see as trying to increase fisheries regulations, and are therefore, weary to sign any document that may impact their fishing rights in any way. In previous cases, the government or other organisations have come into small fishing communities to gather data from the fishermen on their fishery. They then have used this information to create no fishing zones or marine parks without consulting the fishermen, directly impacting the fishermen's livelihoods and providing no alternative sources of income. Several of the fishermen also expressed concern about signing any document due to their lack of a valid fishing license. They feared signing



these documents could expose them to legal prosecution by the government. It took many conversations between leaders of these communities, myself, and other trusted researchers who have been working in these communities for extended periods of time before we could obtain the signatures. Even though the contracts were eventually signed the fishermen still showed a great deal of concern. In the end it took strong relationships between leaders of these fishing communities and researchers who have been working there for an extended period of time to resolve this issue. Trust was built over time and we were entirely transparent with the fishermen on how the data we collected would be used.

This same issue also arose when implementing the social survey to these communities. Their distrust of the government and other organisations was shown in several communities where all the fishermen refused to take part in the survey. Fishermen in many communities are very guarded about providing any type of information on their fishery to anyone. This is the main reason we did not reach our goal of 120 completed social surveys.

Another issue that arose was obtaining samples from the market. Originally vendors were donating samples during the pilot study for this research. However, the vendors soon abandoned this practice and sought payment for the samples. They would also not take small pieces from already cut pieces of fish. Whole chuletas or filets had to be bought. The price wasn't excessive, about USD\$1 per sample, but did add up when ~850 samples were collected from the markets. This was more of a budgeting inconvenience.

DNA extraction from shark muscle also proved to be difficult and required the testing of several different methods. Originally the plan was to use a cost effective bead beater method, which utilises small glass beads to break down the tissue for extraction. However, the buffers used to help digest the tissue and the chemicals in the muscle tissue (i.e. urea) reacted in an unexpected way and resulted in very poor yields of DNA. Chelex extractions, which are also relatively inexpensive, were also tested but proved to not provide uniform enough DNA concentrations. In the end, Qiagen extractions were used and provided the most consistent DNA concentrations that proved to work well in the PCR and barcoding reactions. Using this method was more expensive than was previously estimated for extractions, but by far provided the best results. It was also found that from the shark tissues collected during the research, fins, not muscle, proved to provide the best DNA yields.

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

- 1) DNA barcoding results show that at least seven species of shark and one ray (*Alopias pelagicus, Carcharhinus falciformis, Mustelus lunulatus, Nasolamia velox, Rhizoprionodon longurio, Sphyrna lewini, Sphyrna zygaena, Dasyatis* spp.) are being sold in local markets, with *C. falciformis* representing 80% of samples tested. This is in accordance with findings from previous studies using observer data from long-lining vessels in Costa Rica. The endangered scalloped hammerhead, *S. lewini*, and the smooth hammerhead, *S. zygaena*, were also identified in the market samples (6.6% and 1.4% respectively). Of particular interest is the fact that the majority of *S. lewini* samples discovered came from one specific vendor, therefore, investigating whom they receive their sharks from and how they fish may provide valuable information on fishing practices endangering this species.
- 2) Within the artisanal fisheries, six species of shark were identified (*A. pelagicus, Carcharhinus limbatus, M. lunulatus, N. velox, R. longurio, S. lewini*), with *S. lewini* accounting for ~85% of all sharks landed. Estimated total lengths for *S. lewini* in these



fisheries, based on observer data, suggests that each of the sharks sampled were either juveniles or neonates. Based on these data it is likely that the areas from which these samples were collected represent important nursing grounds for *S. lewini*. The quantity of young *S. lewini* being removed from these areas, and ones similar to them, by artisanal fisheries could be of great importance when determining future conservation measures for this species. Our findings also indicate that sharks being caught in these artisanal fisheries do not appear to represent those being sold in markets around San Jose.

3) Data from collected social surveys is still being analysed, however, preliminary results have demonstrated that perceptions and behaviours of artisanal fishermen toward sharks, the impact of their fishery on shark populations, and shark conservation vary between the different fishing communities and on the fishing methods (i.e. gillnet, hand-line, long-line) in which they use.

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

A large portion of this research was conducted in small fishing communities along the Pacific coast of Costa Rica. These communities contributed to this research by donating shark tissue samples to determine species composition in their fishery and in some cases taking part in a social survey to gauge their attitudes and potential behaviours toward sharks in their fishery and shark conservation that could impact their fishery. The social survey provided a means for the local fishermen to express their views and knowledge of sharks and shark conservation anonymously. The results from these surveys will be distributed to the fisheries governing body within Costa Rica (INCOPESCA), via the conservation organisation PRETOMA, to show INCOPESCA the current support for shark fishing legislation within these communities and their willingness to work with officials to reduce shark bycatch and other activities causing recent shark population declines.

Working with the NGO PRETOMA we are also working to distribute the results on the species of sharks being sold in local markets to the government to show the need for further monitoring of large-scale (industrial) fishing vessels to protect threatened species. Our results show that the vast majority of the sharks being sold in markets around San Jose do not come from small-scale artisanal fisheries, but rather from larger vessels that can travel farther from shore. We found that none of the artisanal fishermen travelled farther than 10 km from shore, and no silky sharks, a semi-pelagic species that accounted for 80% of sharks being sold in the markets, were ever documented in their catches. Many of these larger vessels are foreign owned and there have been issues in the past with regulating their catches (i.e. shark finning). PRETOMA has already been working with fishery regulators through workshops on how to ID shark fins being sold and there are plans to also run these workshops on how to use genetic tools to continue with the identification of products being sold in the markets, and landed by large-scale fisheries, as a means of further fisheries monitoring. Better regulations on these large-scale fisheries can also help with the sustainability of the artisanal fisheries in Costa Rica by reducing pressure on adult shark populations responsible for replenishing the juvenile sharks found along the coast.

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

There are plans to continue this research with the focus shifting slightly to rays being caught and sold in the fisheries and markets. Based on recent studies many ray species are highly threatened with over exploitation and it is important we identify the species that are currently at risk. During my



research, increases in ray sales in the markets were documented. Establishing a DNA barcode library for Costa Rican ray species would be a first step, followed by the monitoring of markets and artisanal fisheries using the forensic genetic techniques from this research. The development of two to three social surveys that would target the different groups (fishermen, vendors, consumers) utilising rays in Costa Rica has also been proposed. The goal of these surveys would be to understand why there is a growing market for rays in Costa Rica and how we can work with these groups to establish sustainable usage of these animals. We have already heard stories from fishing communities in Costa Rica about other areas within the country where fishing for rays started and within a few years had caused complete population crashes.

Within the artisanal fisheries juvenile and neonate scalloped hammerheads represented the largest proportion of shark landings. We now plan to use population genetics to look at the connectedness (structure) of this species in Costa Rica and other areas in the eastern tropical Pacific. Specifically we want to look at how nursing areas along the Costa Rican and Central American Pacific coast are connected to one another, as well as to the adult populations that school at various geological structures in the open ocean (I.e. Cocos Island, Galapagos Islands).

There is also a push for the Costa Rican fisheries governing body, INCOPESCA, to begin to use some of the genetic forensic techniques to monitor shark fisheries to help develop better management practices in the future. Currently, PRETOMA is working with members from INCOPESCA to help with the implementation of these techniques.

Researchers that assisted with this research from the University of Costa Rica and PRETOMA are also continuing to monitor shark landings from several artisanal fisheries included in this study.

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

The work from this project will be shared in several different formats. Preliminary results have already been shared with members of the University of Costa Rica's fisheries department via an invited presentation at the school, as well as, a presentation to the fishing community in Ojochal on the research. There are plans for more presentations and workshops to disseminate this information among government officials, the fishing communities, and market vendors. Many of the vendors selling sharks in the markets showed interest in learning about the species of sharks they were selling (many were unsure on the exact species) and concern with the overexploitation of some sharks. Informing vendors on the types of species they are selling and the conservation concerns related to those species could be a first step in them making more sustainable decisions in the future. The results from this work will also be submitted to peer reviewed journals for publication and appear together as a dissertation for the Department of Environmental Science and Policy at George Mason University. Much of this research will also be submitted for presentation at the Joint Meeting of Ichthyologists and Herpetologists and the American Elasmobranch Society in the summer of 2015.

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

The funds from the RSG were used over a full calendar year. It was necessary to perform several sampling trips to Costa Rica that were relatively evenly spaced throughout the year to account for any seasonal changes that may occur in landings of specific shark species. Several pelagic species of



sharks use the coastal waters of Costa Rica as pupping and nursing grounds but only appear during specific times of the year. Therefore, sampling only during one time of the year could either inflate the estimated number of these species being caught or not record their occurrence in the fishery at all. The anticipated final length of the project, which includes submission of manuscripts to journals, is approximately 21 months. The overall time necessary to complete all necessary lab work after the final samples were collected was 14 months in total. An additional 2 to 3 months beyond this are anticipated for final data analysis.

# 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
Air Travel	1536	1732.95	196.95	Airline tickets are subject to price fluctuations and I mistakenly did not include the exit tax associated with leaving Costa Rica.
Ground Transport	374	1199.66	825.66	My original plan for ground transport was to rent a car. However, the price displayed online apparently is never what is charged by the company when you get there. They have hidden fees that do not appear until you get there (i.e. insurance). Due to this I only rented a car for the first trip (total cost £288). After this I used taxis which, were much easier to use in San Jose, and buses to get to the coast where I didn't need a vehicle once there. This proved to be more cost effective than renting a car.
Subsistence	960	451.14	-508.86	
Lodging	0	222.58	222.58	The Reserve that I would stay at in Ojochal changed its policies after the research had begun and started charging £6.4/night.
Shark Tissue Collection Supplies	496	488.26	7.74	These funds were used to get materials to collect samples and also for shipping the samples to the U.S.
DNA Extraction from Tissues	1280	1664	384	Ultimately less samples than predicted were collected, however, a more expensive DNA extraction process was needed to yield the concentrations necessary for this research. This resulted in the price exceeding what was predicted despite requiring DNA extraction from fewer samples.



Genetic	Analysis	of	1344	504	-840	Due to there being less samples the
Samples						price of genetic analysis was reduced
						and the money saved helped fund the
						higher ground transportation costs,
						lodging and DNA extraction.
TOTAL			5990	6262.59	272.59	

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

This research helped to answer several questions about shark fisheries in Costa Rica, but also created more that need to be answered. As previously mentioned, one fact that was clearly evident is that there is a growing market for ray meat in Costa Rica. As the project progressed, more of the market locations where shark samples were being collected were beginning to sell ray meat. Rays have been deemed some of the most vulnerable and least studied elasmobranchs making this a concerning finding. In many cases rays can become extirpated from an area due to increased fishing pressure even faster than sharks. During this project several samples of ray meat were collected from the markets to determine species ID. I found that almost all the rays in Costa Rica do not have reference DNA barcodes in Genbank for species ID. For these animals an important next step would be the development of DNA barcoding reference sequences for the rays in Costa Rica. This would allow for monitoring of these species in the fisheries and at the markets. As discussed above, we found that scalloped hammerheads made up a majority of the sharks being caught in the artisanal fisheries. An important next step would be looking at the population genetics of this species within the waters along the Pacific coast of Central America to look for population structure. This would encompass collecting samples from more artisanal fisheries both inside of Costa Rica and in other countries, as well as, samples from open water location around areas like Cocos Island.

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

The RSGF logo was used printed on all the social survey forms distributed to the local fishermen. The logo was also shown during a presentation describing the research project to the CIMAR in San Jose, Costa Rica. During said presentation the RSGF was also thanked for funding this project.

### 11. Any other comments?

I would like to sincerely thank the Rufford Foundation for their financial support for this research. Without their help this work would not have been possible. Their funding provided the means to answer several pertinent questions for shark conservation in Costa Rica and clarify the next steps to help conserve these animals.