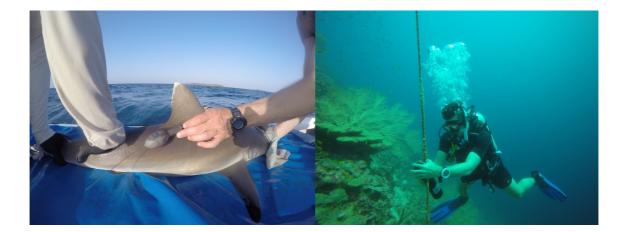
BIOT Array Servicing & Reef Shark Tracking Expedition

Final Report, March 2018

DMP Jacoby¹, D Curnick¹, RJ Schallert², D Tickler³, N Truelove², T White² & TK Chapple²

¹Institute of Zoology, Zoological Society of London ²Stanford University, Hopkins Marine Station ³Centre for Marine Futures, Oceans Institute, University of Western Australia



Executive Summary

The British Indian Ocean Territory (BIOT) is among the largest no-take contiguous marine protected areas in the world. In recent years the marine ecosystems of BIOT have faced growing challenges posed by increasing water temperatures and more extreme climatic events threatening to disrupt the complex balance of life harboured by the remote atoll systems that comprise the Chagos Archipelago. As a component of the Bertarelli Program in Marine Science (BPMS), researchers from Stanford University, the University of Western Australia and the Zoological Society of London have established one of the key monitoring platforms for assessing how some of the largest predatory fish in BIOT are responding to their changing environment and threats posed by illegal fishing activity. Using acoustic and satellite tracking technologies the team can observe the movements, distribution, residency and connectivity of mobile marine predators both close to the reef and during excursions into deeper pelagic waters. A spatially extensive network of acoustic receivers has been monitoring tagged fish, sharks and manta rays for approximately six years providing invaluable long-term data for addressing changes in these marine megafauna communities. In addition, new genetic techniques (eDNA) are now being employed alongside our technology to explore community composition over broad spatial scales and monitor the physical and biogeochemical processes shaping the

ecosystems of BIOT. In order to maintain the continuity of data gathered via our archival acoustic monitoring array our receivers require regular servicing to ensure continuous functioning of the array. In March 2018 we undertook a research expedition aboard the *RV Tethys Supporter for X days*.

Similar to previous years, the expedition objectives included servicing the entire receiver array and deploying a new cohort of long-term acoustic and shorter-term satellite tags. With the assistance of the crew aboard R/V Tethys and under somewhat challenging time and weather constraints, the team deployed tags on 71 sharks from four different reef associated species. In total, 81 individual tags were deployed, with 10 individuals double tagged with both pop-up satellite tags in addition to the acoustic tags. The team was able to recover, download and redeploy 43 VR2W receivers and 1 underwater modem unit, yielding over 370,000 new detections to contribute to the time-series. In addition, two VR4 Global units were serviced with new moorings, ensuring continued live feeds of acoustic data. Following the success of the previous deployments and retrieval of acoustic release receivers (VR2AR) deployed beyond diveable depths - for example on the submerged seamounts of Sandes' and Swart's four AR units were deployed near the original locations from 2016. The team collected water samples for eDNA from 20 separate receiver and tagging locations. The samples were filtered on site and frozen for next generation sequencing at Stanford. In addition the team took still images, video, 360° and drone-based footage to contribute to the BPMS media library and for all up and coming outreach events. The expedition was covered on several social media platforms with three blogs written from the field ensure that the work of the BPMS in BIOT reaches a broad audience. The data gathered from this recent expedition will contribute to many research projects and papers as the BPMS moves forward into its next phase. Importantly, these continued data, combined with environmental monitoring and enforcement data, enable us to assess the impact the MPA and the changing climate is having on our marine predator guilds and continue to make BIOT a benchmark for ocean monitoring in the Indian Ocean.

Introduction

The British Indian Ocean Territory (BIOT) is among the world's largest marine protected areas and effective long-term biodiversity and fisheries management strategies are only possible with extensive knowledge of how and when species utilize this region. In this respect the acoustic monitoring array funded by the BPMS is essential for assessing how elasmobranchs and teleost fishes utilize this area. Researchers from Stanford University, the University of Western Australia (UWA), and the Zoological Society of London (ZSL) have used electronic tag technology to study the residency, habitat use and connectivity of fish, sharks and mantas, within and around BIOT. Not only is this research vital for understanding the importance of the archipelago as a refuge for pelagic fish, elasmobranchs and the more residential reef associated animals but through the application of novel analytical techniques such as network analyses, insight is also being gained on the behavioural strategies of animals and their predictability for informing management strategies. In addition, telemetry data obtained from both acoustic and satellite tags of marked animals can be used to estimate home range and habitat use of the focal species. From such analyses we can also identify aggregation hot spots and estimate shark density throughout BIOT. Electronic tag data are also a critical component of mark recapture models that are being used to assess the population size of different species as well as aiding in the development of new technology, such as the FAST tag. In addition, the tagging effort in BIOT provides information on large-scale, long-term movement patterns and connectivity of populations of reef and pelagic species across with wider Indian Ocean basin. This BPMS research program is facilitating the development of the BIOT Marine Reserve as an ocean observatory for monitoring important abiotic variables pertinent to climate change by placing instruments with a long-term environmental data acquisition potential. New projects are currently being undertaken that will work to incorporate these environmental data with shark tracking data to better understand the drivers of movement and population distribution, in addition to assessing the impact of recent coral bleaching events on mobile marine predator communities.

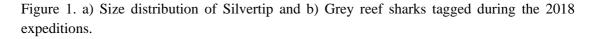
To complement telemetry data, recent advances in next-generation sequencing (NGS) and bioinformatics have led to the development of powerful new tools to detect the presence of BIOT elasmobranchs and teleosts by sequencing environmental DNA (eDNA) from seawater samples. eDNA present in seawater may originate from a variety of sources, such as damaged tissue, shed scales, sloughed cells, blood, mucous, or metabolic waste. All molecular methods for identification of species using eDNA from seawater rely on five key steps: (1) filtering seawater onto a fine membrane to collect eDNA (2) extracting eDNA (3) amplifying eDNA by PCR (4) sequencing using a NGS platform and (5) using bioinformatics to match eDNA sequence data to the correct species by accessing large DNA sequence databases. Recent eDNA studies have revealed hidden diversity that was not detected by dive surveys or baited remote underwater video and such techniques will be invaluable for sampling species distribution beyond what can be inferred from the telemetry data. The team use eDNA to identify the presence of sharks and coral reef fish in the Chagos, particularly those that are not typically encountered during tagging and survey expeditions. Results can be compared with other BIOT teams using traditional methodologies for identifying presence absence of vertebrates.

To support these goals, an expedition was undertaken in March 2018 aboard the Seychellesbased vessel, RV Tethys Supporter. During a 16 day expedition the scientific team aimed to maintain the existing acoustic monitoring array and deploy additional acoustic, satellite and biologging tags on pelagic and reef associated elasmobranch fishes. In addition, the team also collected water samples to detect the genetic signatures (eDNA) of animals associated with the archipelago.

Results and Discussion

The expedition afforded the team 16 days within BIOT to achieve our primary goals of fully servicing and downloading the array data to ensure at least another continuous year of tracking data. We also aimed to deploy acoustic tags on a new cohort animals to replace those lost from the study through natural or fishing mortality and departure from the areas under surveillance, and to opportunistically deploy pop-up satellite tags on the larger individuals captured. The tagging teams deployed tags on a total of 71 animals with 81 tags: 40 grey reef sharks (*Carcharhinus amblyrhynchos*), 26 silvertip sharks (*Carcharhinus albimarginatus*), four silky sharks (*Carcharhinus falciformis*) and one whitetip reef shark (*Triaenodon obesus*) (Table 1, Figure 1&2). The tagging of the hitherto elusive silky sharks was a significant accomplishment since we continue to have a poor understanding of their spatial use of the marine reserve, but hypothesise that they have the largest activity space of any of the shark species tagged to date and therefore are a vital umbrella species for spatial management planning in BIOT. We hope

that the four silky sharks will reveal novel information on these threatened ocean wanderers (Vulnerable, IUCN Red List).



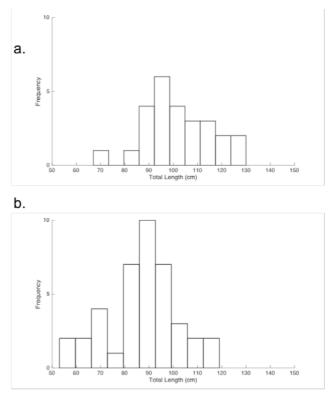


Table 1. The new cohort of animals tagged in 2018 with 10 year acoustic and satellite tags.

Species	Total Individuals	Acoustic	Satellite
Silvertip Shark	26	26	4
Grey Reef Shark	40	40	3
Silky Shark	4	4	3
Whitetip Reef Shark	1	1	0
Total	71	71	10

The BPMS acoustic dataset now comprises over 1.2 million individual detections from 310 animals, over a total of 56,970 detection days (Figure 3). Downloads from the units retrieved during this expedition resulted in >370,000 new detections. Overall the mean monitoring period to date for all animals is 452 days (1.3 years), and mean residency index (days detected per days monitored) is 41%. Incredibly, one female grey reef shark tagged in the first year of the

project has been detected for a total period of 1,642 days (4.5 years) with a 36% residency index; two additional silvertip sharks tagged in 2013 and 2014 have been monitored for 1,403 and 1,353 days with residency indices of 80% and 77%, respectively, indicating the importance of the reef habitat around the atolls where our receivers are situated.



Figure 2. A 135cm grey reef shark leaves the tagging boat equipped with both an internal acoustic transmitter and archival satellite tag (miniPAT – pop up archival transmitter), set to release after 90 days of high resolution temperature, depth and light level tracking.

Following the success of acoustic release VR2AR deployments in 2016, nine ARs were deployed in similar locations as before enabling acoustic coverage of diver inaccessible locations such as the seamounts, that will help to access deep water passage of some of our tagged animals between reef locations. The new locations of these ARs is given below. Finally, the VR4 Global units were both serviced and redeployed, ensuring another year of real-time acoustic data from Peros Banos and Egmont lagoon.

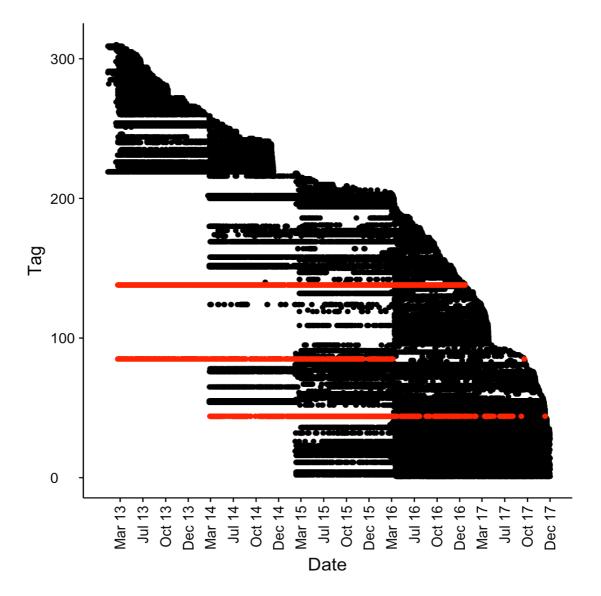


Figure 3. Abacus plot of daily detections on the BPMS acoustic array from February 2013 to December 2017; tag plots are ordered in sequence of most recent detection date. Longest detection sequences are highlighted in red.

Changes to the acoustic receiver array

The array has been reduced to 56 elements from 92 (Table 2). In total, the team dove at least once on 69 locations (VR2W, VR4-UWM, VR4G) of the existing receivers in the array, 43 of which were retrieved and downloaded for the last two years-worth of data and redeployed. In order to streamline the maintenance on future expeditions and in consideration of recorded data, 9 receivers were removed without replacement as some were considered redundant. The

prevailing weather conditions prohibited us from retrieving receivers placed opportunistically around the GCB in 2016. These will be serviced by future expeditions. A number of moorings were lost since our previous expedition due to the length of time between servicing trips. Given the locations of the missing receivers, we suspect the large storm that carried a VR4G (complete with anchor) out and away into the Indian Ocean was likely responsible for the loss of these receivers. As a result, we have developed a new mooring design, which we plan to deploy on our next expedition. We expect this new design will prevent such losses in the future.

Receiver type	April 2016	March 2018
VR2W	69	41
Acoustic release	16	9
VR4-UWM	4	4
VR4G	3	2
Total	92	56

Table 2. Receiver types and numbers in the BPMS acoustic array as at April 2016 and March 2018.

The current acoustic array maintains the high density of receivers in the core section of the study area around Peros Banhos and Salomon Atolls, and the adjacent features at Benares Shoal, Blenheim Reef, Speakers Bank and Victory Bank. The array at Egmont has been reduced to focus on the lagoon and the sheltered eastern reef to continue monitoring sharks and mantas tagged in those locations. Remotely retrievable receivers fitted with acoustic release (AR) mechanisms were redeployed at depth on the seamounts (four units) as the data retrieved in 2017 showed these locations to have high numbers of resident animals. AR units were also used around Speakers Bank (three units) as well as at Blenheim Reef (two units) to simplify servicing requirements in 2019. Two of three live transmitting VR4 Global units were serviced *in situ* with both moorings being replaced. The third was completely torn from its mooring during a storm in 2017 and has subsequently been retrieved from the coast of Tanzania. This will be redeployed in 2019. The four VR4-UWM (underwater modem) units have been left in situ until 2019, when they will be serviced with new batteries and moorings. A summary of the current receiver distribution, compared to 2016, is given in Figure 4.

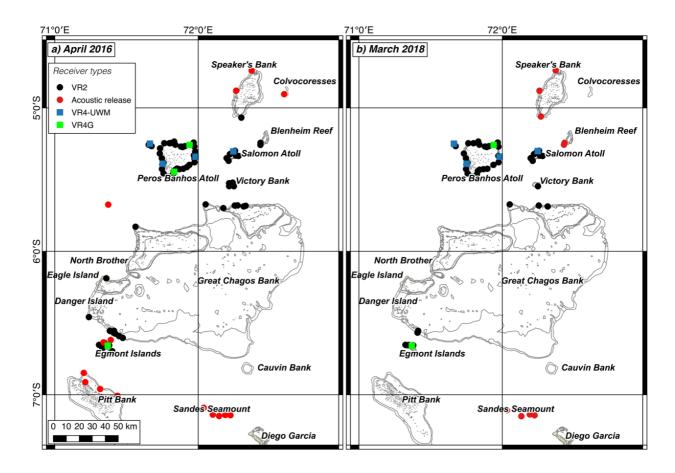


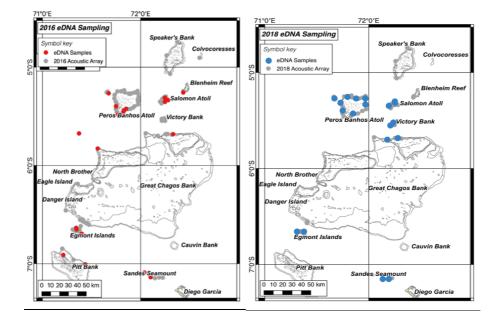
Figure 4. Map of BIOT showing locations of elements of the BPMS acoustic receiver array in a) April 2016 and b) March 2018.

In addition to servicing the telemetry equipment and deploying tags, 6 liter of seawater was filtered at each of 18 receiver locations and two seamounts in BIOT (Table 3). Seawater samples were stored on ice after collection, then stored at -20C after filtration, and transported to Stanford University on ice to avoid DNA degradation. Protocols developed by the Block Lab at Stanford University will then be used to detect the occurrence of shark and bony fish diversity from samples gathered. The results of the 2018 eDNA survey will be compared with 2017 work conducted by Dr. Luke Gardner.

Date	Sample Location	Location ID
3/8/18	Salomon	SA03
3/8/18	Salomon	SA01
3/9/18	Salomon	SA05
3/9/18	Peros Banhos	PB07
3/10/18	Peros Banhos	PB15
3/10/18	Peros Banhos	PB21
3/10/18	Peros Banhos	PB10
3/11/18	Peros Banhos	PB03
3/11/18	Peros Banhos	PB28
3/11/18	Peros Banhos	PB27
3/11/18	Victory Bank	VB01
3/12/18	Victory Bank	VB02
3/12/18	Nelson Island	NI02
3/12/18	Grand Chagos Bank	GCB02
3/13/18	Benares	BE02
3/15/18	Egmont	EG04
3/15/18	Egmont	EG05
3/17/18	Swartz Seamount	
3/17/18	Sandes Seamount	

Table 3. eDNA sample locations. Two samples were taken at each site, at 20m depth and at the surface.

Figure 5. Map of eDNA samples collected from the BIOT in 2016 (left) and 2018 (right).



Communications

The expedition team comprehensively documented the work from the field sending daily tweets and images for both live media interaction and subsequent outreach and education events. Three blogs were written during the course of the 16 days. Links to these can be found below:

https://www.zsl.org/blogs/science/being-a-rubber-duck-in-the-indian-ocean https://www.zsl.org/blogs/science/did-curiosity-kill-the-shark https://www.zsl.org/blogs/science/against-the-elements

Several hours of GoPro and drone footage were recorded, documenting our activities both above and below the water. In addition, we recorded the first 360 degree footage taken that we intend to showcase at this year's Royal Society Summer Science Exhibition, amongst others, where visitors will be able to engage with BIOT as a virtual reality experience, diving on our receivers and witnessing the reefs almost first hand. Extracts from the footage taken by the team is currently being edited as part of a trailer for the Royal Society that will be hosted on their YouTube channel in the coming weeks.