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ACTIVITIES OF THE PROTECTIVE TURTLE
ECOLOGY CENTER FOR TRAINING, OUTREACH,
AND RESEARCH, INC. (ProTECTOR Inc.) IN
HONDURAS

2013 and 2014 ANNUAL REPORT

September 11, 2015



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ECOLOGY CENTER FOR TRAINING, OUTREACH,
AND RESEARCH, INC (ProTECTOR Inc.) IN
HONDURAS**

***ANNUAL REPORT OF THE
2013 and 2014 SEASONS***

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PREFACE

This report represents the ongoing work of the Protective Turtle Ecology Center for Training, Outreach, and Research, Inc. (ProTECTOR) in Honduras. The report covers activities of ProTECTOR during the 2013 and 2014 calendar years, and is provided in partial fulfillment of the research permit agreements provided to ProTECTOR by DIGEPESCA. All published articles resulting from this work have been supplied to the appropriate government agencies of Honduras with this report.

ACKNOWLEDGEMENTS

ProTECTOR recognizes that without the financial assistance of the Department of Earth and Biological Sciences (Loma Linda University), these ongoing projects could not take place. The Global Health Institute (LLU and the Students for International Mission Service (LLU) also supported various aspects of our community outreach. We are grateful to Loma Linda University graduate students Noemi Duran, Dustin Baumbach, and Christian Hayes for directing field studies, and to ProTECTOR Interns Christian Hayes, Kyungje Sung, Lelyn Castillo, Liesl Cole, Samantha Serna, Gabriela Ochoa, Marsha Wright, Linda Baeza, and Rodney Smith, for all their hard work on field projects. We are also indebted to the community of Punta Ratón and the Municipality of Marcovia for their participation in these sea turtle conservation efforts. We thank Noemi Duran for assistance in organizing and tabulating data collected from this and past years. These studies were conducted under approval from the Loma Linda University Institutional Animal Care and Use Committee (IACUC) (Protocol # 89029), and the Loma Linda University Institutional Review Board (IRB) (Protocol # 5120308 and # 5120097), and are in compliance with United States and Honduran law. We thank the Dirección General de Pesca y Acuicultura (DIGEPESCA) and the Secretaria de Agricultura y Ganadaria (SAG) for Research permits (Permit # SAG-224-2011) provided for undertaking these studies.

September 11, 2015

Cover image: *Lepidochelys olivacea* hatchlings at Punta Ratón hatchery. Photo: © S.G. Dunbar, 2011

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INTRODUCTION AND BACKGROUND

A comprehensive background regarding previous work undertaken by ProTECTOR on the Hawksbill (*Eretmochelys imbricata*) and Olive Ridley (*Lepidochelys olivacea*) sea turtles, and the need for continuing research on their status and plight in Honduran waters, has been provided in previous reports to DIGEPESCA (Dunbar 2006, Dunbar and Berube 2008, Dunbar and Salinas 2008, 2013). Those reports provided details on methods carried out by ProTECTOR under SAG permits **#DGPA/005/2006; DGPA/245/2006; DGPA/5428/2007, DGPA/707/2009, SAG/251/2010B, and SAG/224/2011**, and provided study results obtained up to November, 2012.

We provide the following report on the activities of ProTECTOR Inc. between January, 2013 and December, 2014, combining two years of activities into the current report. This report provides information on all ProTECTOR Inc. projects throughout Honduras, including the Bay Islands and South Coast. These studies continue with the aim of tagging and tracking juvenile hawksbills, nesting hawksbills, and nesting olive ridley sea turtles in our study sites, as well as community outreach and development of additional sea turtle research and conservation activities, with the aims of benefitting local communities, eco-tourism operators, and marine protected area (MPA) managers with information and recommendations for improved resource management and community outreach. Over the past two seasons, we have continued to further develop strong research, conservation, and community development ties with the community of Punta Ratón, Utila, and the West End community of Roatan. Developments within the community of Punta Ratón continue to be difficult, with the continuing issue of intra-community rivalries and lack of community direction, and little to no government oversight of the La Veda hatchery projects throughout the south coast of Honduras.

In addition to the continuing work of ProTECTOR Inc. during the veda period, ProTECTOR Inc. in partnership with the Loma Linda University Students for International Missions (SIMS) continued to provide health care services to the communities of El Venado and Punta Ratón during the 2013 summer season. Research work carried out in 2013, and analyses of these data done in 2014 has provided a basis from which new investigations can be launched into nesting

beach monitoring, hatchery development, hatchling migrations studies, and population genetics analyses.

In addition to the work on the South Coast, additional projects were undertaken along the in the Bay Islands in collaboration with the Roatán Marine Park, and the Bay Islands Conservation Association (BICA – Utila)..

This report has been furnished to all appropriate Secretariats, Ministries, and Departments of the Honduran Government, including SAG, DIGEPESCA, SERNA, and DiBio, in both Spanish and English languages. Data from this report may be included in the annual report of Honduras to the Inter- American Convention for the Protection and Conservation of Sea Turtles (IAC) with appropriate credit cited.

RESEARCH IN 2013

Studies continued in the community of Punta Ratón (Fig. 1) on olive ridley (*Lepidochelys olivacea*) nesting, hatchery management, and hatching success.



Figure 1. A map of the Gulf of Fonseca showing the distance hatchlings must travel to reach the open Pacific from their release point at Punta Ratón.

Tidal Influences on Hatchling Movements in the GOF

One of the first studies to be completed was a study on the release methods of hatchling turtles from the hatchery managed by the community at Punta Ratón. We noted that when eggs incubated in the hatchery eventually hatched, they were often held in tubs of water for up to 24 hours before being released to the open ocean. Releases were typically done at night, and very often during the later periods of outgoing tides. We undertook tests of hatchlings' drifting and swimming movements at two different times of the outgoing tide; during the mid-outgoing tide, and during the start of the outgoing tide. Because of the conditions and shallow depth of the Gulf of Fonseca, tidal movements are strong and swift.

We tracked hatchlings released with a modified “Witherington float” tethered to the hatchlings with 1 m of thread (Fig. 2). Within the floats, we placed a chemical glow stick, which could be seen from 20 – 30 m away, allowing us to approach the hatchling every 5 – 8 minutes in order to record the latitude/longitude with a GPS. The full details of the methods of this work are provided in Duran and Dunbar (*In Review*).



Figure 2. The Witherington float with chemical glow stick attached to the hatchling for night observations.

We found that hatchlings released during the mid-outgoing tides were carried along with the surface currents until the tide changed to its incoming trajectory. At that stage, hatchlings were unable to swim against the incoming current and were swept back to the point of release. In

some cases, hatchlings were swept further north than the point of release (Fig. 3) and into the Boca de la Jagua estuary.

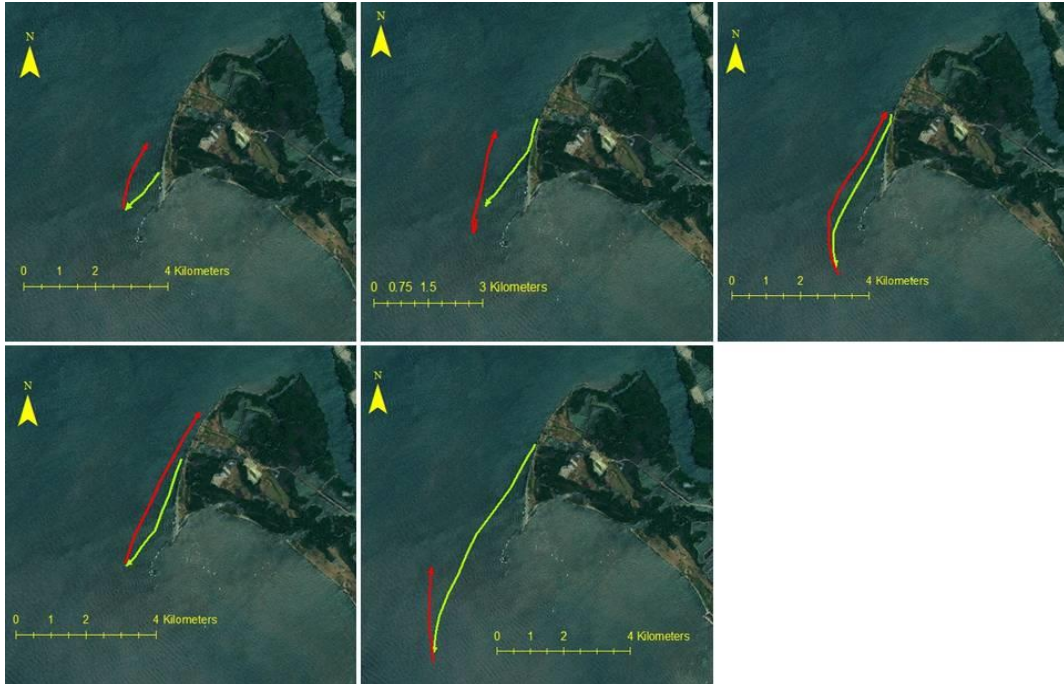


Figure 3. Swimming and drifting patterns of hatchlings released from Punta Ratón Beach at the mid-outgoing tide. Yellow lines indicate hatchling direction and movement during outgoing tide; red lines indicate hatchling direction and movement on the subsequent incoming tide

In contrast, when hatchlings were released at the start of the outgoing tide, we found that hatchlings were able to make much further progress by swimming with the outgoing tide for the longer duration. In this case, when tide turned, hatchlings were swept back to a small degree, but were not swept back to the point of release (Fig. 4).

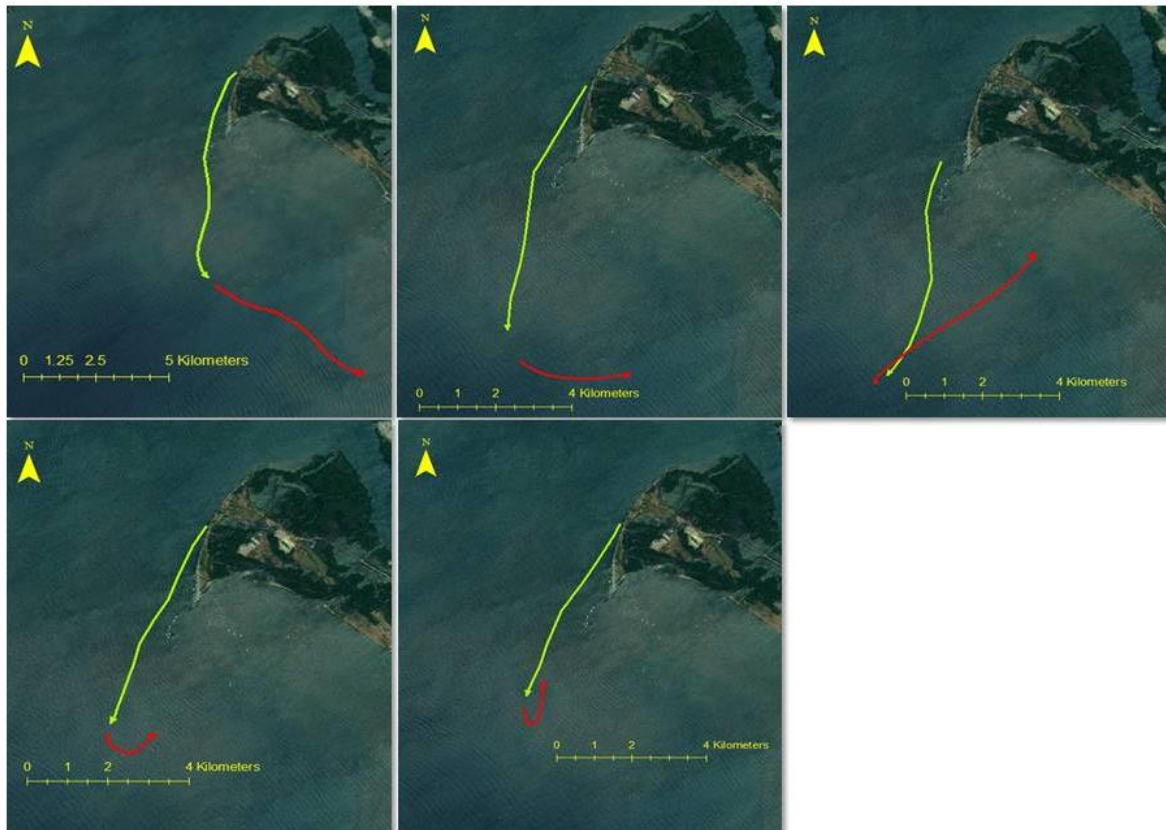


Figure 4. Movements of hatchlings released from Punta Ratón Beach at the start of an outgoing tide. Yellow lines indicate hatchling direction and movement during outgoing tide; red lines indicate hatchling direction and movement on the subsequent incoming tide.

These results highlight the important impact of currents and tides in the Gulf of Fonseca on the hatchlings released from La Veda hatchery projects. Our results suggest that without proper research into best practice methods, current La Veda methods may be leading to the demise of many of the hatchlings released as part of the Veda project implemented by the Honduran Government and overseen by the Division of Biodiversity (DiBio) in the Ministry of Environment (SERNA). This study suggests that when hatchlings are, instead, intentionally released at the start of outgoing tides, they are much more likely to make their way to the mouth of the Gulf, than are those hatchlings that are released on a mid-outgoing tide.

This study further underscores the need for continuing research that leads to understanding sea turtle behaviors and activities, and that directs conservation efforts for the benefit of the species, and the local communities that manage the hatcheries in the Gulf of Fonseca.

Hatchling Predation and Swimming Patterns in the GOF

In another study, we investigated the predation rates on hatchlings released from Punta Ratón Beach. This study was to further understand the fate of hatchling turtles that had been incubated during the Veda period and released after hatching in the community managed hatcheries of the GOF. Prior to this study, no effort had been made to investigate the fate of turtles released from the GOF hatcheries.

To undertake this study, we released hatchlings at night with Witherington floats that contained chemical glow sticks. We also released hatchlings during the day with Witherington floats in which the chemical glow stick was replaced with a small yellow balloon. Turtles were followed and their positions and behaviors recorded at regular intervals. We performed 22 trackings at night, 11 during decreasing tides and 11 during increasing tides. We also performed 3 trackings during daytime.

As a result of this study, we recognized differences in swimming patterns of hatchlings swimming at night versus swimming during the day, and carried out an extensive study of these behaviors. Detailed methods for both the predation and swimming pattern studies are provided in Duran and Dunbar (2015).

During the predation study, we recorded no successful predation events on hatchling turtles during their swimming either at night or during the day. These results are very different from a number of other studies on turtle hatchling predation reported in the literature, and may be a result of the specific conditions of the waters in the GOF. We did observe one event in which a group of hatchlings was released during the day and a seagull attacked and captured a hatchling. This hatchling, however, managed to free itself from the beak of the gull and was dropped into the water. The gull did not make another attempt to recapture the hatchling. This

event demonstrates that hatchlings released in groups may be more conspicuous to potential areal predators than hatchlings released alone.

Results of the swimming pattern study showed that hatchlings spent the majority of their time swimming at the surface during the night, only diving to depth briefly (Fig. 5A). In contrast, when hatchlings were observed during the day, they spent the majority of their time swimming at depth, only coming to the surface briefly to breathe (Fig. 5B).

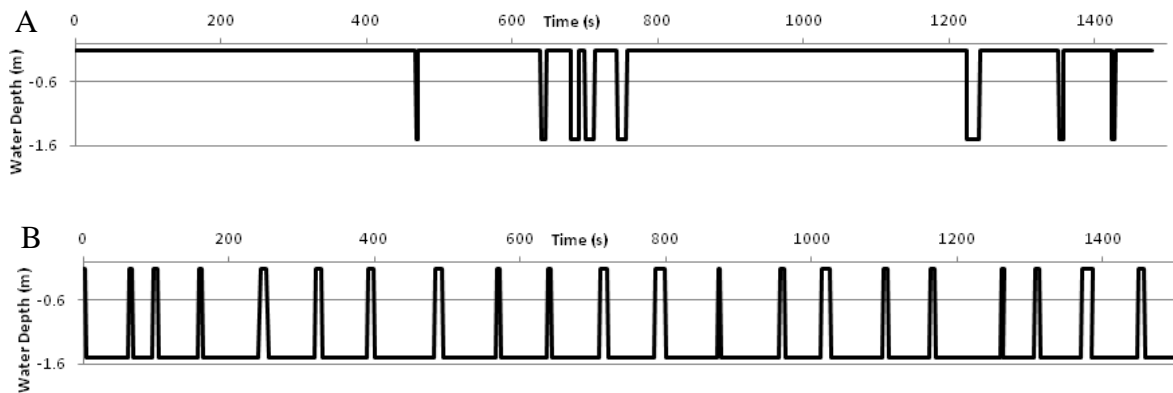


Figure 5. A: Swimming pattern of hatchling turtles released at night. B: Swimming pattern of hatchling released during the day.

These results suggest that *L. olivacea* may be adapted to the specific conditions of shallow turbid waters in the GOF and may be swimming at depth during the day as an effective anti-predatory measure. There have been no other reports in the published literature that demonstrate this behavior in other locations where swimming patterns have been reported for turtle species. Taken together with other studies, these results suggest that local oceanic conditions may drive the evolution of innate swimming behaviors. In the Gulf of Fonseca, due

to its abundance of sea birds and very turbid waters, it is clearly advantageous for the hatchlings to swim at depth as much of the time as possible during the day.

Further research is needed to determine whether this behavior is characteristic of olive ridley sea turtles in other areas or if it is a local adaptation for enhancing survival under the specific conditions of the Gulf of Fonseca. In any case, it would be of interest to perform laboratory studies to assess the actual energy investment this behavior requires of the animals, and compare it with the amount of energy used by hatchlings of other species normally swimming near the surface and during diving in response to the presence of aerial threats. In the case that this behavior appears to exist only in the Honduran population of the Gulf of Fonseca, it would be worth investigating whether hatchlings of other sea turtle species nesting in the area, such as hawksbill and green turtles, also show a similar behavior during offshore migration from beaches of Pacific Honduras. An in-depth presentation of the results of this study can be found in Duran and Dunbar (2015).

Community Hatchery Management Study

To understand the actual success of the government-sponsored, community run hatchery at Punta Raton, we conducted a group of experiments comparing nest conditions, hatching success, and hatchling characteristics within the hatchery nests versus semi-natural nests. Because no nests on the beaches of Punta Raton are left to naturally incubate due to egg poaching both during and outside of the Veda period, we had community members gather nesting female turtles from the water's edge (a practice regularly undertaken by the community), and place the turtle in a protected enclosure at the upper reaches of the beach. The turtle was allowed to wonder within the enclosure and naturally, dig, lay, and cover the eggs before leaving. These eggs were then left *in situ* and were thus considered "semi-natural" nests.

As noted previously, when turtles hatch, it is common practice for the turtles to be held for up to 24 hours before being released. We investigated several parameters associated with the conditions of the nests (nest temperatures), the hatching success (how many eggs successfully hatched and emerged alive relative to the number of eggs laid or buried), and the hatchling condition both immediately after hatching compared with 24 hours after hatching.

To compare nest conditions, we measured nest temperatures with temperature data loggers placed within each experiment nest, as well as within pseudonests (controls) both in the hatchery, and in the semi-natural nests. Temperature data loggers collected temperature data over the entire incubation period.

To compare hatching success, we excavated nests 46 days after being laid. We counted live hatchlings, dead hatchlings, and unhatched eggs. The sum of these three numbers was considered the total number of eggs for the clutch. We calculated hatching success as the total number of hatched neonates (hatchlings both live and dead) divided by the total number of eggs. Hatching success was calculated for four experimental nests at the beach site (B1, B2, B3 and B4) and eight nests at the hatchery, for a total of twelve nests. The eight nests from the hatchery included four nests containing thermal dataloggers (H26, H67, H93 and H94), two additional nests used for the hatchling body condition and performance experiments (H101 and H108), and two nests not used in the experiments that were excavated while we were present in the hatchery (H64 and H65).

To evaluate if there were differences in body condition after 24 hours of holding the hatchlings in a container, we investigated running speed and swimming ability immediately after hatching, then again after 12 hours. From the 15 hatchlings previously weighed and measured from each nest, we randomly assigned six to be tested for running speed and six to be tested for swimming style. Running speed was assessed outside at night under natural light conditions. We used a 1-m long PVC gutter lined with sand and placed seaward on the beach with the natural beach slope. Hatchlings were placed at the upper part of the gutter and allowed to crawl to the lower end.

For the swimming style test, we used a transparent glass tank (30×30×55 cm) filled with 15 cm of sea water from the beach at Punta Ratón, and kept at ambient temperature. We fitted each hatchling with a 5-mm Velcro band around the widest part of the carapace, and attached it to a wood pole located on the top of the tank *via* a monofilament tether. The band did not touch any of the flippers during movement. The tether was adjusted in a manner such that the hatchling could swim freely. We observed 10 minutes of swimming for each hatchling, and counted the

number of power strokes (synchronous movements of the frontal flippers), and the time to the nearest second performing dogpaddling swimming style (alternate movement of the four flippers), for 60-sec periods beginning at 1, 5, and 9 minutes. With these three values, we calculated the average number of power strokes per minute, and the average dogpaddling time per minute for each individual. Detailed explanations of the methods and results can be found in Duran et al. (*In Review*)

We found the mean incubation temperature of nests in the hatchery were not significantly different from those on the beach in any of the incubation thirds (Fig. 6). However, there was a significantly higher metabolic heating in nests at the hatchery over the nests on the beach during the second and third thirds of the incubation period (Fig. 7).

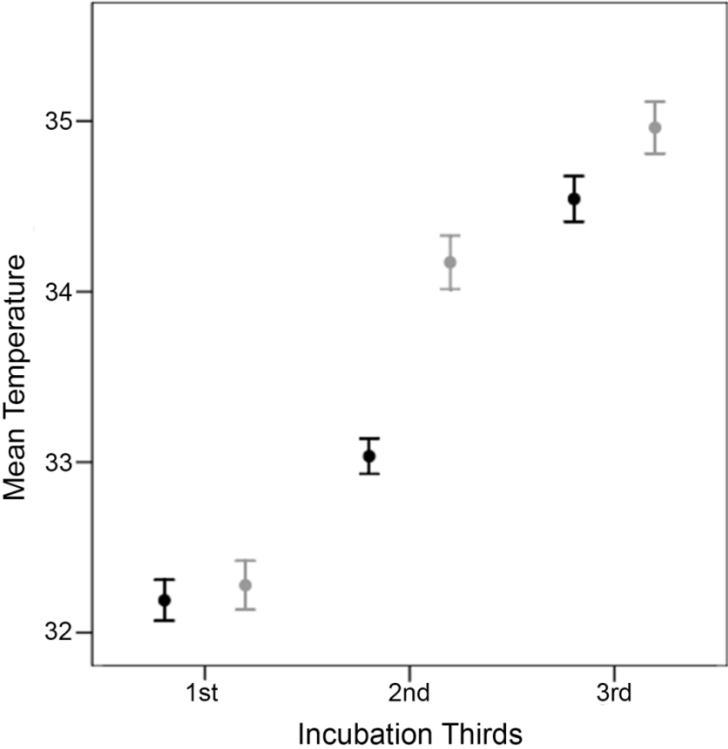


Figure 6. Mean nest temperatures for nests in the hatchery (grey) and semi-natural nests at the beach (black) over each third of the incubation period.

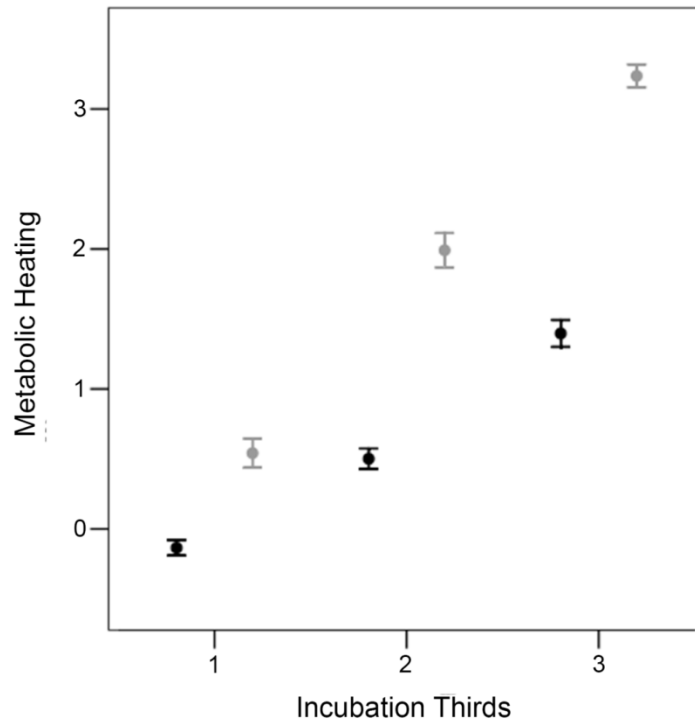


Figure 7. Mean metabolic heating of hatchery nests (grey) and semi-natural nests (black) over each third of the incubation period.

With respect to hatching success, we found that mean clutch hatching success differed significantly between the nests at the beach site and the nests in the hatchery site (beach: $83.22\% \pm 4.04$ SE; hatchery: $24.08\% \pm 6.00$ SE, $t_{10} = 6.818$, $p < 0.001$) (Fig. 8).

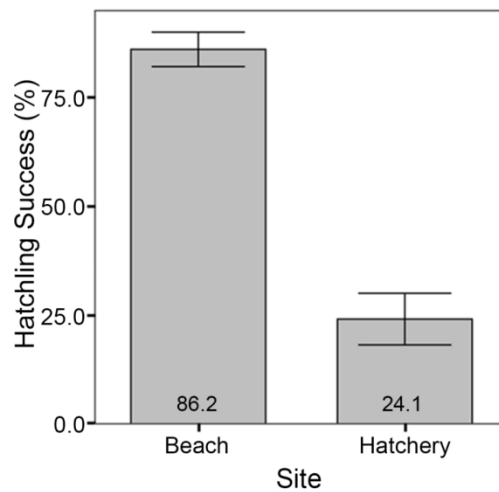


Figure 8. Hatching success rates of semi-natural beach nests, compared with hatchery nests.

When we investigated running speed, power strokes, and the amount of time undertaking dog paddles immediately after hatching compared with 24 hours later, we found differences in all measures with the second measures significantly lower than the first measures (Table 1).

Table 1. Locomotion performance results.

Means and SE for running speed (RunSpeed), number of power strokes (PS) per minute, and time per minute swimming dogpaddling style (DPTIME), are shown for hatchlings from the two experimental sites.

	1st measurement			2nd measurement (after 24hours)		
	RunSpeed (cm/s)	Number PS per min	DPTIME per min (s)	RunSpeed (cm/s)	Number PS per min	DPTIME per min (s)
Beach	1.48 ± 0.11	38.9 ± 5.5	19.2 ± 1.7	1.06 ± 0.13	31.7 ± 4.8	13.5 ± 1.3
Hatchery	1.23 ± 0.15	25.7 ± 3.1	14.4 ± 1.1	0.68 ± 0.11	20.4 ± 3.8	13.4 ± 1.5

This study has demonstrated that, under the current conditions of hatchery management in Punta Ratón, Honduras, the olive ridley nests in the hatchery incubated at higher temperatures, especially during the second third of incubation (34.2 °C vs 33.0 °C), experienced higher metabolic heating (1.9 °C vs 0.6 °C), and showed lower hatching success (24.08 % vs 83.22 %) than the semi-natural nests buried by female turtles on the upper part of the beach and kept *in situ*. The high incubation temperatures observed at the hatchery were likely the result of metabolic heating in individual nests affecting neighboring nests due to the close proximity of nests to each other. High incubation temperatures are known to affect hatching success. However, our results suggest that mean temperature alone may not be the best predictor of hatching success. In our case, we found that mean temperature during the second third of incubation had a higher influence on hatching success than the overall mean incubation temperature.

Although statistically significant differences were not demonstrated in all cases, our results suggest that nests at the hatchery produced smaller and less fit hatchlings than the nests at the beach. Hatchlings from the beach averaged 0.9 (6.5 %) g larger, ran 0.3 cm/s (42 %) faster, and swam using 12 (61%) more power strokes per minute than hatchlings from the hatchery. We

suggest that these excessively high temperatures were a consequence of the metabolic heating of nests affecting near neighbor nests. According to this hypothesis, the temperatures at each nest would have differed depending on the specific position of the nest, how many nests were nearby, the state of development of embryos in those nests, and the distances separating them.

The results of our holding study clearly show that retaining hatchlings for 24h after emergence or nest excavation may be detrimental. The effects of retention were similar for hatchlings from both the beach and the hatchery. After 24 h of retention, hatchlings had lost an average of 0.9 g (6.5 % of their body mass), and they ran much slower (0.2 vs 0.7 cm/s), and swam fewer (13.6 vs 19.9 power strokes per minute and 9.5 vs 12.5 s using dogpaddling style) than just after emergence. Negative effects of retention time on running speed, swimming speed, and swimming style have been reported in previous studies, even for much shorter periods of retention. Our data indicates that retaining hatchlings for long periods of time after emergence or removal from the nest, prevents them from optimally performing these natural behaviors, and threatens their survival by extending the time spent on land and in shallow waters.

Sea turtle conservation practices consisting of relocating eggs laid on the beach to hatcheries may have detrimental results for embryos and hatchlings when these hatcheries are not appropriately built and managed. Hatchery nests in this study experienced shared metabolic heating and reached excessively high temperatures during the second third of incubation, likely causing much lower hatching success, as well as smaller and less fit hatchlings than those nests incubated on the beach. We thus recommend nests to be kept *in situ* as much as possible, even if it is necessary to use creative solutions, such as moving the incoming females to safe parts of the beach to nest.

Photo-Identification and Automated Searching

We tested a series of photographs collected over the past 7 years of hawksbill research in Roatán and subjected several of these to testing using the photo-identification software I³S (Spot). Photographs were tested against a database of photographs from previous research. Both the computer-assisted matching program and manual visual matching were compared to

each other for true positive and false positive matches. Details of the methods and results can be found in Dunbar, et al. (2014).

This study found the computer photo-ID software, I³S a useful tool for re-identifying individual turtles whose photos had been taken above water with specific protocols as part of the overall research method. We concluded that using in-water photos may present more challenges because it is not always possible to take photographs underwater at the same angle and under similar light and water conditions from one time to another. Thus, we suggest that the program be tested using a suite and database of underwater photographs taken in situations during dive sightings. However, the use of observational research, even when using photo-identification, cannot take the place of other research techniques in which hand capture, blood/skin/scute sampling, flipper/radio/satellite tagging are needed. These techniques are required to answer many research questions that cannot be answered by purely observational activities alone. A full presentation of the results and discussion of the study can be found in Dunbar, et al. (2014).

RESEARCH 2014

Influence of SCUBA Diving on Sea Turtle Behavior

In collaboration with the Roatán Marine Park (RMP), we launched an observational study in 2014 with the aim of understanding the behavioral responses of sea turtles to recreational SCUBA divers. Although previous studies had investigated the behavior of turtles in response to snorkelers, no study had previously investigated turtle responses to divers. Since diving is a major tourism industry in the Bay Islands of Honduras, we enlisted the assistance of the RMP and the dive shops in the area of the West End, to carry out observations on the approaches of divers toward the hawksbill sea turtle (*Eretmochelys imbricata*) at frequently used dive sites.

We used weekly dive sightings logs, as well as older records from participating dive shops to calculate the frequency of dives at each dive site. We also undertook direct observations of turtles while diving using methods by Dunbar et al (2008), and also utilizing dive tourists as assistants in approaching turtles while diving. All behaviors were characterized into six solitary and two social behavior categories. We also conducted repeated in-water observations for

turtles (as able) to test for turtle habituation to diver presence. For a detailed presentation of the methods and results of this study, see Hayes, et al (*In Review*).

We collected turtle sightings information from 14 dive operations in the West End. Dive operations recorded 701 dives at 46 sites between June 9 and August 29, 2014. A total of 666 hawksbills, 420 greens (*Chelonia mydas*), four loggerhead (*Caretta caretta*), and 22 unknown turtles were reported during the study. Of the hawksbills, 393 (59%) were reported as adults and 273 (41%) as juveniles. Spatial distribution of sightings and divers indicated that divers tended to make more sightings between West End and West Bay and fewer between West End and Sandy Bay.

From 12 June to 2 September, 2014, we conducted 6092.0 min of surveys at 23 sites in the Sandy Bay West End Marine Reserve (SBWEMR). The average number of hawksbills observed per dive was 0.7 ± 0.1 . We obtained repeated observations of 11 turtles, with nine individuals observed twice and two individuals observed three times. Total initial observation time was 823.9 min. and total time for repeated observation (not including initial observation time) was 203.4 min. Mean number of divers ($n = 221$) observing turtles was 3.0 ± 1.0 (1–10). Although 22 turtles (36.1%) exhibited an obvious reaction (indicated by a rapid change in turtle swimming direction or activity) when approached by divers, 42 (68.9%) did not.

Diver approach did not impact the median number of bouts that hawksbills ($n = 42$) engaged in swimming, eating, investigating, and breathing behavior (Wilcoxon Signed Rank: $S < 41$; $p > 0.05$). Conversely, the mean time turtles ($n = 53$) spent eating and investigating was significantly lower during diver approach than when divers were at baseline position (Fig. 5B; eating: $F_{(1, 43)} = 4.31$, $p = 0.044$, β estimate = -1.79; investigating: $F_{(1, 43)} = 5.12$, $p = 0.029$, β estimate = -2.48).

Our results suggest that small groups of intermediate to experienced divers (1-4 divers) in MPAs can significantly reduce the amount of time hawksbills spend foraging and breathing. Conversely, we found that current levels of recreational diving within the SBWEMR do not significantly impact hawksbill abundance.

RECOMMENDATIONS

Based on these findings we make **the following recommendations:**

Firstly, additional in-water observation studies should be conducted both inside and outside MPAs to determine if policies and management enforcement within MPAs protect sea turtles from the potential impacts of recreational diving. Specifically, foraging and flight response behaviors of turtles within and outside MPAs should be compared to quantify the effect of recreational diving policy on sea turtle behaviors.

Secondly, additional long-term sightings and dive log surveys should be conducted in MPAs, particularly in areas heavily impacted by diving. These surveys should be combined with habitat assessments of local sea turtle foraging grounds to evaluate if recreational diving pressure indirectly impacts sea turtle population levels through the degradation of foraging habitats.

Thirdly, long-term sea turtle photo-identification surveys using software systems, such as I³S Spot and I³S Pattern should be implemented in MPAs to facilitate accurate species identification and long-term studies of individual turtles. If implemented over an entire MPA, long-term photo-identification surveys would enable management officials to estimate sea turtle population sizes, monitor changes in sea turtle populations over multiple years, and re-identify resident and migrating individuals.

Fourthly, additional studies, such as regular health assessments through the capture and blood/skin/scute sampling of individuals should be undertaken on a regular basis to assess potential contamination issues in both the habitat and the turtles in the protected area. While there is good merit in observational studies (i.e. tracking individuals and estimating populations through observational and photo-ID studies), there is no substitute for tracking the health, genetics, and movements of turtles, which may only be accomplished through standard techniques of capture and re-capture, blood and tissue sampling, flipper/radio/satellite tagging. These measures should never be undertaken by those not holding appropriate permits, and without appropriate experience in handling and sampling sea turtles.

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