

COPROT

SEASON 2021 - 2022

ANNUAL REPORT OF NESTING ACTIVITY PLAYAS CARATE, RIO ORO Y PEJEPERRO

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I. RESUMEN

El presente informe pretende hacer una valoración del monitoreo de la temporada de anidación 2021- 2022 llevada a cabo por el proyecto comunitario de conservación de tortugas marinas COPROT en las playas Carate, Rio oro y Peje Perro entre el mes de junio de 2021 hasta el 15 de marzo 2022.

Durante este periodo se registró un total de 6,278 nidadas de tortuga lora (*Lepidochelys olivacea*) y 379 de tortuga verde (*Chelonia mydas*) de las cuales de reubicaron respectivamente 245 y diez nidadas.

El porcentaje de emergencia para las nidadas exhumadas de tortuga lora fue de 60 % (n=307) y 83.4 % (n=130) para la tortuga verde. El estimado de neonatos liberados para todas las nidadas exhumadas fue de 15, 490 neonatos, de los cuales 8,983 fueron de tortuga lora y 6,507 de tortuga verde. Se estima la producción global de neonatos a 183, 697 para la tortuga lora y 18,970 para la tortuga verde.

Un total de 48 nidadas marcadas (de las 398 nidadas marcadas) de lora fueron depredadas mientras se registró 4 de tortuga verde. El porcentaje de saqueo sigue siendo bajo (1.9%) sin embargo es importante resaltar la presencia de personas involucradas en narcotraficante presentes en la playa y la amenaza que representa para la perennidad del proyecto de conservación.

II. ABSTRACT

This report aims to present the results of the monitoring of the 2021-2022 nesting season carried out by the community project for the conservation of sea turtles COPROT on the Carate, Rio Oro and Pejeperro beaches between June 2021 and March 2022.

During this period, a total of 6,278 Olive Ridley turtles (*Lepidochelys olivacea*) and 379 Pacific green turtles (*Chelonia mydas*) clutches were recorded, of which 245 and 10 clutches were relocated respectively.

The emergence percentage for the excavated Olive Ridley clutches was 60 % (n=307) and 83.4 % (n=130) for the Pacific green clutches. The estimated number of hatchlings released for all excavated clutches was 15,490 hatchlings, of which 8,983 were Olive Ridley hatchlings and 6,507 were Pacific green hatchlings. The estimated total production of neonates across all our beaches for the Olive Ridley species is 450,000 individuals and 25,000 for the Pacific green species.

A total of 48 Olive Ridley nests and 4 Pacific green nests (out of our sample set of 398 marked nests) were recorded as predated (13% predation rate). The percentage of illegal extraction remains low (1.9%), however it is important to highlight the presence of people involved in drug trafficking on the beach and the threat it represents for the sustainability of the conservation project.

III. INTRODUCTION

Sea turtle populations today suffer threats of a different nature, including direct human activities (hunting and consumption of eggs) and indirect human activities (coastal development, overfishing and pollution), as well as other factors such as nest depredation by wild animals and dogs, in addition to the effects of climate change (high tides, floods, extreme temperatures and ocean acidification).

The study area is one of the most important beaches for solitary nesting of the Olive Ridley sea turtle at the regional level, and there are also other species that are priorities for conservation. Playas Carate, Pejeperro and Río Oro have nesting sites for 4 species of sea turtles; the Olive Ridley (*Lepidochelys olivacea*) and the leatherback (*Dermochelys coriacea*) which are found with vulnerable populations, the Pacific green or black turtle (*Chelonia mydas*) which is found with populations in danger of extinction, and the hawksbill (*Eretmochelys imbricata*), which is found Critically Endangered. Therefore, it is imperative that, through the monitoring of turtles, we develop studies that help us formulate proposals for mitigation and adaptation to the different threats that are due to their importance as umbrella species such as illegal poaching of eggs and hunting, predation by dogs, erosion and loss of beaches for nesting, the effects of climate change on species and the ecosystem, among other factors.

The territories of Carate and Rio Oro are part of the Golfito canton, which in 2011 had a population of 39,150 people, which represents 35% of the territorial population corresponding to the Brunca region and, at the district level, is part of Puerto Jiménez, which registered at the time of the census a low district social development index (INDER, 2015), this can translate into low employment opportunities, basic living conditions, low level of schooling, among other things.

According to INDER (2015), in the district of Puerto Jiménez some economic activities are livestock, tourism, artisanal fishing, forestry and commercial activities. However, there are also other activities that people in the area engage in, and Rio Oro and Carate have been the scene of activities that are harmful to the environment, such as illegal hunting. The historical illegal gold mining for survival in the Corcovado National Park and on the banks of the Carate and Rio Oro rivers, as shown in the documentary Nosotros las Piedras (Torres-Crespo, 2018), and the looting of turtle eggs with a total of XX registered nests poached for this nesting season 2021 2022.

However, the territory has characteristics that can favour the development of the population and begin to generate changes in the current socio-environmental situations since it represents one of the most biodiverse areas in the country, if not the world.

Thus, a monitoring and research project such as COPROT offers the opportunity to involve different local actors, volunteers and conservation organisations, to contribute to the protection and monitoring of endangered species.

IV. MATERIAL AND METHODS

A. SPECIES

The species encountered in this area are mainly the Olive Ridley (*Lepidochelys olivacea*) and Green (*Chelonia mydas*) sea turtle species; however, since the project was founded in 2019, nesting activity from Hawksbill (*Eretmochelys imbricata*) and Leatherback (*Dermochelys coriacea*) sea turtles were also recorded. The following tables provide a summary of each of these 4 species.



B. STUDY AREA

COPROT monitored the beaches of (2.5km), Río Oro (2.5km) and Peje Perro (3km), located on the Osa Peninsula (town of Carate, Puerto Jiménez, province of Puntarenas, Costa Rica) to collect data on nesting female sea turtles (Fig. 1). Our research station is located just 400m behind the beach in between beaches Río Oro and Pejeperro behind the river, Río Oro (coordinates 564232, -932071).



Figure 1. Map of monitored beaches. Playa Carate (Yellow), Playa Rio Oro (Red), Playa Pejeperro

C. BEACH PREPARATION

We've placed posts every 25m with sequential numbers to locate our activity with visual ease on the beach. We always record the sector to the West of the nesting activity. The zone records the relation of the activity to the tides. Zone 1 is an activity under the high tide line. Zone 2 is an activity between the hide tide line and the vegetation, and zone 3 is an activity behind live vegetation (Fig. 2).



Figure 2. Map of tidal zone placements.

D. NIGHT PATROL AND MORNING CENSUS

Between 01 June 2021 and 15 March 2022, COPROT staff patrolled bi-daily field surveys: one at night and one in the morning. Each survey is complete when at least one person has walked the length of the entire beach and recorded all sea turtle track activity. Night patrols are conducted 6 days a week, and the time of the patrol depends on both the season and the tide height. During the rainy season (August–November), night patrols

leave 3 hours before low tide, for the safety of the assistants due to unpredictable river swells at high tides. During the dry season (June–July and December–March), patrols leave 2 hours before high tide, as more turtle activity in certain populations has been documented with respect to an increasing tide (Reina et al. 2002). The time spent on the beach will depend on the number of turtle activities on the beach but typically lasts between 3–8 hours. The primary purpose of night patrols is to collect accurate population data on nesting females: to identify individuals (Inconel external tags), record biometric data (CCL, CCW), and assess injuries on the female. Because sea turtles predominantly nest at night, these data could not otherwise be gathered through morning census.

Morning census are conducted 7 days a week, leaving at 5am from the station each day. These patrols last between 2–6 hours and serve as a final monitor of the beach to ensure no turtle activities were missed from the previous night. Additionally, during morning surveys, research assistants check for emerging nests, evidenced by hatchling tracks, and perform a full nest excavation (see excavation protocol below).

To assess patrol effort, we record the date and the start and end time of each patrol.

E. DATA COLLECTION

When we encounter a nesting female, we first record the time we encountered the female, which matches the current turtle activity. All sea turtles follow a regimented nesting process. First, the turtle emerges from the water and searches for a spot to lay her eggs. Second, she prepares the nesting area by moving dry sand from the nesting area; this clears beach debris and prevents nest collapse. Third, she digs her nest chamber. Fourth, she lays her eggs. Fifth, she covers her eggs. And sixth, she returns to the water. If the turtle has left tracks on the beach, but has returned to the sea before we've collected data, we still collect spatial data on the track activity: recording the activity as "no turtle," and recording the time we encounter the track.

Once the turtle has begun laying eggs, we can begin to collect our data without disturbing the turtle. During the moment before the turtle begins depositing her eggs, we record the depth of the nest from the bottom of the nest chamber to the base of the turtle's plastron to the nearest centimetre. We record the total number of eggs laid, counting as they fall. Once the turtle has laid 25 eggs, we measure the curved carapace length (CCL), from the edge of the turtle's carapace, where the shell meets the soft tissue to the notch of the turtle's shell, and the curved carapace width (CCW), the widest part of the turtle's carapace to the nearest decimetre (Fig 3.). We assess the turtle for any injuries (missing flippers, missing chunks of carapace, or injuries to the head area). Once the turtle has begun to cover her nest, we place a small metal marker that contains the date, time encountered, and species of the encountered turtle, which is unique to each nesting activity. This is used to verify and pair the nest at the time of hatching with the data from the female. As the turtle is covering, this is also when we will apply any new metal Inconel tags to the turtle's two front flippers in the 1st scale. During the 2021–2022, we focused out tagging efforts on the more endangered Pacific Green turtles, but we've applied for a tagging grant to begin applying tags to Olive Ridleys for the upcoming 2022–2023 season to gain a better understanding of our solitary Olive Ridley nesting population.

Occasionally, the female will decide to return to the sea prior to depositing her eggs. This is called a false crawl. In this case, we will gather all available data once the female begins returning to the water (CCL, CCW, any tagging codes, evidence of previous tag, and any visible injuries).



Figure 3. Biometric measurements for shelled turtles, CCW and CCL

1. SPATIAL DATA

We record spatial data in four different ways: 1) Sector, 2) Zone, 3) GPS, 4) Triangulation. If there is a nest, this data will be taken for the nest location; if the turtle left a false crawl, these data will be taken at the highest point of her crawl.

For all Pacific Green nests and a subsample of Olive Ridley nests, we record GPS coordinates to 3m accuracy of the nest. Additionally, for these nests we will triangulate to have a precise location of nest placement. To the nearest centimetre, we measure from the west sector post, the centre post (which we write on with the nest marker information), and the east sector post to the nest. We can reverse these measurements to relocate the nest prior to its predicted hatching date.

In summary, for all encountered track activity (False Crawls and nests), we record the date of the activity, species (as identified by the track pattern), time of encounter, the west sector of the beach, the zone of the activity, the activity of the turtle (if not seen, "No turtle"), and we indicate whether the activity was a nest or a false crawl. (Table. 1)

Table 1. Data Sample.

SPECIES	SECTOR	ZONE	HOUR	ACTIVITY	NEST	DEPTH	PROT	PREDATION	NOTES
L	81.5	2	05:51	PON	ND	v	1	PIZ	
v	81.75	3	09:45	NT	Ν	×	PALOS	1	

2. OLIVE RIDLEY VS PACIFIC GREEN

Due to our large abundances of nesting Olive Ridley turtles, we do not collect data on every nest. We take a subsample of nests to track and monitor in order to obtain the most data depending on the nesting stage when we encounter the female. Once we encounter an Olive Ridley, we wait with her for 10 minutes. If she has not begun nesting after that time, we continue patrolling the rest of the beach. Regardless of whether she has begun nesting, we will check for any visible injuries, tags, and any evidence of previous tagging and will return to the activity later to check if she has deposited eggs. However, because Pacific green turtles are considered critically endangered in the Eastern Pacific, we take all possible data on the female turtle and on the nest. If we miss a green nesting activity, we will use a probing stick to locate the eggs and we will mark the nest and take all spatial data on the nest.

Olive Ridley nests are shallow, and we protect those nests from major predators on our beach (dogs, coati, and racoon), by creating a log covering over the nest using driftwood on the beach (Fig. 4).



Figure 4. Example of a log covering protecting all Olive Ridley Nests

Due to our predation analysis from last year, we determined that we did not need to continue protecting green nests, as they are much deeper and not predated by mammalian predators on our beaches.

F. RELOCATION PROTOCOL

Occasionally, a turtle will lay her eggs in the intertidal zone (Zone 1). These nests are extremely vulnerable to being washed out by the tides with a 0% emergence success. If we encounter any nests in zone 1, we relocate the nest to a safer zone. It's important that all relocations are performed as close to the deposition time as possible because after 6–8 hours of laying, the embryo of the turtle forms and attaches to the shell for development (Pintus *et al.* 2009). To avoid severing this connection, we perform all of our relocations at night

or between 5–7am during the morning patrol and to handle eggs with great care to avoid rotation. For all relocations, we dig a new nest chamber to move the eggs, record all available data on the female, as well as record the new location data, the time the eggs have been relocated, and the number of eggs. We place a metal marker and monitor all relocations to ensure that our conservation effort is successful.

G. PREDATION

During morning patrols, we check our nests in the ground for any predation activity. If there has been a predation on one of our marked nests, we record the type of predator (as assessed by tracks), and we check if the entire clutch has been lost. If there are still viable eggs, we will record a half predation in our database, place the metal marker back in the nest, and re-protect the nest. If the entire clutch has been lost, we record the full predation and will not return to the nest for excavation.

Further, as a designated protected area by SINAC, we tally all predation events and predator species on sea turtle nests daily specifically on the Rio Oro beach as part of the PRONAMEC data collection initiative.

H. NEST EXCAVATION

Just before each nest has reached its incubation duration, we locate and mark the nest to be checked daily for hatchling tracks. Once there are tracks or the nest has been incubating for 75 days, we will check the nest contents. If there are still viable eggs or more than 10 live hatchlings in the nest, we will cover the nest and wait for hatching. If there are rotting eggs or fewer than 10 live hatchlings in the nest, we will excavate the nest.

During an excavation, all nest contents are removed from the nest chamber and the metal nest marker is found. We record the nest code on the metal marker to pair with our nesting female data. We then record the depth of the nest to the nearest centimetre from the bottom of the nest to the surface of the surrounding sand. Then we count all empty eggshells, unhatched eggs, live and dead hatchlings in the nest and record. For all unhatched eggs, we open them to determine the stage of development when the turtle stopped developing. Stages of development are shown below (Fig. 5). We also record how many unhatched eggs have evidence of bacteria or fungus, presenting as a thick and discoloured yolk. Once the excavation is finished, we put all of the nest contents back in the nest and cover it with sand. We remove our centre post to remove any nesting evidence from the beach.



Figure 5. Stages of embryonic development. Each unhatched egg is opened and the stage of development is recorded.

I. AVAILABLE NESTING SPACE

Additionally, as part of the PRONAMEC data collection requirements, we collect tide data once a month on Rio Oro beach. We take measurements every 50m from the high tide line to the vegetation directly behind the metre marks, and always take this measurement during the third quarter of the moon cycle in order to be consistent with tide height. We use these measurements to gauge dynamic changes in available nesting space from month to month. This is coupled with our spatial nesting data to better understand female nest site selection.

J. RESEARCH ASSISTANT TRAINING

We have nesting turtles year-round on our beaches, and our research assistants stay for a minimum of 3 months. Upon arrival, research assistants have a series of presentations on sea turtle biology and our data collection methods. Additionally, for the first two weeks of night and morning surveys the new assistants will train with the Project Coordinator or more experienced assistants. We accept research assistants on a rolling basis, so there is always someone available to train new incoming research assistants. If comfortable with data collection and surveys after two weeks, new assistants will begin leading surveys and training volunteers on patrols. We have weekly meetings to discuss data collection methods on the beach and discuss irregular events on the beach as they arise.

K. MINI ARRIBADAS

To assess the potential occurrence of mini arribada events on the beaches of Rio Oro, Pejeperro and Carate, the data was analysed for spikes in Olive Ridley nesting numbers throughout the entire season of 2021/2022. Arribadas are mass nesting events which were defined as a 24h window with over 100 nesting events (Beange, Clift & Arauz, 2015). Only successful nesting events were included within this analysis. The events were

classified by week with the dates recorded as YYYY/MM/DD and weeks starting on a Sunday. The data arribada events were also grouped according to the lunar cycle to determine potential synchronicities between the two main arribada nesting beaches, Ostional and Nancite, also located on the pacific coast of Costa Rica (Beange, Clift & Arauz, 2015; Koval, 2015). If the arribada event occurred in two lunar phases, it was classed within the lunar phase in which majority of the event days occurred.

L. TAGGING PROGRAM

While the turtle is laying, we check all of the turtle's flippers for any existing tags, in which we would record the tag placement and code, and for any evidence of previous tags (this typically presents as a hole or a small missing chunk/rip in the flipper).

When a turtle finished nesting and tagging was a viable option, a research assistant prepared the tagging by disinfecting the metal tag and applicator with Uterine Antiseptic (Chlorhexidine Gluconate). The Inconel brand was inserted into the applicator, always keeping the tool with the male part facing up (Fig. 4 and 5). The mark with the highest number is always applied on the right fin, while the lowest number always goes on the left fin and leaving a space of approximately 5 to 8 mm corresponding to the last two numbers of the mark between it and the left. tortoise scale to prevent future damage from the growth of the reptile's skin as indicated (Chacòn, Sánchez, Calvo, & Ash, 2007).

The tagging programme, which was initiated in December of 2020, is used to assess the nesting activities and patterns of individual turtles including information such as the re-migration interval (the time between reproductive events of an individual female), the re-emergence interval (the time between turtles visiting the beaches) and the nesting interval (the time between individual clutches in one reproductive season). To investigate the above parameters, simple descriptive analyses were used in R-Studio to assess the average amount of time between nesting events, visits to our beaches and the interval between clutches for individual tagged turtles.



Figure 6. Inconel tagging placement in 1st scale of fore-flippers.



Figure 7. Correctly applied, closed Inconel tag.

M. NEST EXCAVATIONS

To analyse a nest hatching success, two parameters were used: hatching and emergence. This whole section focuses on monitored nests only. In total, 295 nests were tracked from lay date to excavation. For some sections of the analysis, nests were omitted as a result of inaccurate data input, or missing data. hatching success is the total number of hatched eggs and the emergence, the number of these hatched neonates that successfully left the nest, both expressed as percentages. These two functions of success are equally analysed throughout the 'Hatching Success' results section.

To understand the spatial trends in hatching success, each sector (25m) was categorised into the following categories: no vegetation, low vegetation, and trees. To statistically test the differences, an ANCOVA was conducted, along with a Tukey HSD, to identify which groups there were significantly different.

The abiotic conditions of both monitored beaches dramatically change throughout the year, from the wet to the dry season. Considering, the effect of these two seasons on hatching success was analysed using a t-test. For this analysis, the wet season was from June 2021 – 15 December 2021 and the dry season 16 December 2021 – 28 February 2022. Season classification was based on what the majority of eggs were incubated in.

Olive Ridley turtles, unlike Pacific green turtles, regularly nest in the intertidal region of the beach (Zone 1), which often requires nest relocation. To test whether relocated nests' success differs from *in-situ* nests, a t-test was used to firstly test the differences in hatching success and secondly to test the prevalence of fungus and bacterial infection in the relocated and *in-situ* nests.

Fungus and bacteria are known causes of infection in turtle nests. To analyse any spatial trends in infection the average number of unhatched eggs that were infected with fungus or bacteria was observed graphically.

At the beginning of the season, green nests were protected in the same way as Olive Ridley nests. However, from 12 November 2021, green nests were no longer protected, and instead only marked for ease of finding. To analyse the potential effect protection had on green incubation and subsequent hatching success, a t-test was conducted.

V. RESULTS

A. LEPIDOCHELYS OLIVACEA, OLIVE RIDLEY TURTLE

1. TEMPORAL DISTRIBUTION

Between June 2021 and 15 March 2022, 6,278 *L. olivacea* nests were recorded. There was a lower proportion of *L. olivacea* false crawls compared to nests with 1,578 false crawls. Peak nesting month for *L. olivacea* was October (Fig.6). The distribution of *L. olivacea* nests and false crawls had a greater spread throughout the season compared to *C. mydas*, whose nests and false crawls were mostly concentrated between December to February.



Figure 8. Temporal distribution of Lepidochelys olivacea

Throughout the season, the most common time for *L. olivacea* and *C. mydas* sightings was 22:30-23:00 (Fig. 7). There were notable reductions in sightings at 02:00 for *L. olivacea* and 01:00 for *C. mydas*. No individuals of either species were sighted between 19:00-19:30.



Figure 9. Hourly occurrence and encounter of nesting females on Pejeperro and Rio Oro.

2. MINI ARRIBADAS

There were two mini arribada events which occurred, one at the end of September and one at the end of October during the same span of days (Table 2). They lasted around four days and occurred during the third quarter of the moon cycle, one week before new moon (Fig. 8).

Table 2. The descriptive statistics relevant to the mini arribada events which occurred on Playa Rio Oro and Playa Pejeperro in2021 including start date, duration, arribada size and lunar phase during which the arribada occurred.

Month	Start Date	Duration	Number of Nests	Lunar Phase
September	27 th to 30 th	4 days	496	Third Quarter
October	27 th to 30 th	4 days	358	Third Quarter



Figure 10. Top: The weekly number of Olive Ridley (*Lepidochelys olivacea*) nesting events during the entire season of 2021/2022 on the beaches of Rio Oro and Pejeperro, Carate. In green are the weeks with potential arribada events due to extraordinarily high nesting frequency. Bottom: The daily nesting events in September, October and November displaying the arribada events for the weeks highlighted in the top graph. Specifically, the end of September (27th Sept. until 30th of September 2021) and end of October (27th of October until 30th of October 2021) saw two longer mass nesting events lasting several days.







Figure 12. Vertical spatial distribution of nesting activity for L. olivacea

4. PREDATION

A total of 48 Olive Ridley nests and 4 Pacific green nests (out of our sample set of 398 marked nests) were recorded as predated (13% predation rate). The predators observed and their special distribution across the beaches are shown in Figure 13. The percentage of illegal poaching from humans remains low (1.9%), and is highly associated with peak times for fishing.



Figure 13. Spatial distribution of predation events in Rio Oro and Peje Perro a) dogs, b) Raccoons, C) Coatis D) unknown i.e., the predator could not be identified.



Figure 14. Percentage of nests of L. olivacea predated by predators

5. NEST EXCAVATIONS

313 excavations were executed on marked nests throughout this season, 113 more than last season; 138 of these were green and 175 Olive Ridley. Overall, the mean hatching success for both beaches was 62.1% and the mean emergence success was 60.0% (Fig.13). The results from this season cannot be compared to the ones registered during the previous seasons as the nests were randomly excavated when emergence was detected.

It is important to highlight the significant difference between the results obtained from PRONAMEC protocols in comparison with the ones obtained through ours. This difference is due to the non-inclusion of nests that have been predated or eroded away or unsuccessful since the PRONAMEC protocols suggests random excavation of successful nests but fails to follow nests from oviposition to emergence (i.e. any nests lost to predation, poaching or washed by the ocean are not taken into account in the PRONAMEC data sets). Our protocols also dictated that all relocated nests would be marked and therefore included into the sample date set, and we saw a very high predation rate on relocated nests (of 64 nests predated, 50 were relocated nests).



Figure 15. Emergence and hatching success for L. olivacea on Rio Oro and Pejeperro using PRONAMEC and COPROT protocols.

When vegetation cover was analysed, there were significant differences among hatching success in the different vegetation categories; no vegetation, low vegetation and trees (one-way ANOVA, $F_{2,285}$ =3.65,P=0.02; *Fig.*14). The hatching success was greater in areas with trees compared to areas of no vegetation (Tukey HSD: P=0.01), but there was no difference in hatching success between trees and low vegetation (Tukey HSD: P=0.78) or low vegetation and no vegetation (Tukey HSD: P=0.39). Similarly, there were significant differences among emergence success in the three different vegetation categories (one-way ANOVA, $F_{2,285}$ =5.24, P<0.01; *Fig.*2). The emergence success was greater in areas with trees compared to areas of no vegetation (Tukey HSD: P<0.01), but there was no difference in emergence success between trees and low vegetation (Tukey HSD: P<0.01), but there was no difference in emergence success between trees and low vegetation (Tukey HSD: P<0.01), but there was no difference in emergence success between trees and low vegetation (Tukey HSD: P<0.01), but there was no difference in emergence success between trees and low vegetation (Tukey HSD: P<0.01), but there was no difference in emergence success between trees and low vegetation (Tukey HSD: P=0.51) or low vegetation and no vegetation (Tukey HSD: P=0.41).



Figure 16. Mean hatching and emergence success (%) for each species for both rainy and dry season.

There is a significant difference in the emergence success of Olive Ridley neonates between the two seasons, with nests being significantly more successful in the dry season than the rainy (t_{122} =-2.42, *P*=0.01; *Fig.*3), however, there is no significant difference in Olive Ridley nests' hatching success during the seasons (t_{122} =-1.78, *P*=0.07). Moreover, there is no significant difference in green nests' hatching or emergence success in the seasons (t_{29} =-0.34, *P*=0.73 and t_{29} =-0.40, *P*=0.69, respectively; *Fig.*15).



Figure 17. Mean hatching and emergence success (5) for each vegetation category; no vegetation (NV), low vegetation (LV) and trees (T) for both Rio Oro and Peje Perro.



Figure 18. Mean prevalence of fungus and bacterial infections (%) in unhatched eggs for both C. mydas and L. olivacea in each sector range

This season saw 146 relocations of which, 50.68% were relocated from areas at high risk of flooding due to lagoons and rivers. These areas were also characterised by no vegetation. However, there is no significant difference in relocated Olive Ridley nests' hatching or emergence success (t_{145} =-0.13, P=0.89 and t_{145} =-0.35, P=0.72, respectively). Moreover, there is no significant difference in the presence of fungus or bacteria in relocated nests than in in-situ nests (t_{250} =-0.04, P=0.96). It is important to note; 41 nests were disregarded due to either data input errors or the nest containing no unhatched eggs. Additionally, there was no significant difference in fungus and bacterial infections in the three vegetation categories: tree, low vegetation and no vegetation (one-way ANOVA, $F_{2,245}$ =0.67, P=0.51). However, unlike the vegetation categories, some sector ranges did display on average higher infections rates, such as between sector 99.25 to 108.5, which had an average rate of infection in unhatched eggs of 64.43% (*Fig.*16).

Finally, of the 6 protected and 25 unprotected green nests that were successfully tracked and excavated this season, there was no significant difference in protected and unprotected green nests hatching or emergence successes (t_{29} =-1.28, P=0.21 and t_{29} =-1.25, P=0.21, respectively).

B. CHELONIA MYDAS, GREEN TURTLE

A) TEMPORAL DISTRIBUTION

The peak nesting activity for green sea turtles was detected in December and January. *C. mydas* showed the opposite trend to Olive Ridley turtles with a greater number of false crawls to nests with 1,114 false crawls.



■ Number of nests ■ Number of false crawls





Figure 20. Hourly encounters and hourly frequency of *C. mydas* in Rio Oro and Pejeperro.



Figure 21. Spatial distribution by sector of C. mydas



Figure 22. Vertical distribution of nesting activity of C. mydas

C. TAGGING PROGRAM

Using tagging information, we recorded the information of 101 tagged sea turtles on our beaches in the season 2021/2022 (Table 1). Out of these, 95 turtles were tagged within this season.

20211/2022		Tagged Turtles	New Tags	Old Tags	Nests	Emergence Interval	Nesting Interval	Tagging Loss
Species	Green	72	68	4	62	15.77 days	30.17 days	21
	Olive Ridley	29	27	2	27	-	-	-

Table 3. Summary statistics from the 2021/2022 tagging programme including information on the number of tagged turtles, newly tagged turtles, turtles tagged in previous seasons which returned, emergence and nesting intervals.

Out of the 101 tagged turtles, 89 turtles nested amounting to 88.11%. 17 tagged greens nested more than once this season (amounting to 23.61% of greens returning) with no tagged Olive Ridley turtles observed nesting more than once. One female green turtle nested a total of four times (Fig. 1). Nesting events were 30 days apart on average (Table 1) with individuals ranging from 11 to 62 days between nesting events. In total 21 turtles, all of which were greens, showed signs of tagging loss with 17 showing previous evidence of tagging in the form of flipper scars and four only having a tag on one flipper.



Figure 23. The tagged green turtles which nested more than once this season including information on how many nests per individual and the average amount of time between an individual's clutches.

VI. DISCUSSION

A. TEMPORAL DISTRIBUTIONS

The temporal distribution of nests shows similar patterns to last season. Last season however, the month with the greatest number of *L. olivacea* nests was September compared to October this season. The month with the greatest number of *C. mydas* nests last season was February, but was January this season. There was a notable reduction in *C. mydas* nests during September of last season, which is not present in this season's data.

A large number of turtles from both species were sighted one to two hours either side of 22:30. Sightings were less common up until 21:00 and no turtles were sighted between 19:00-19:30 for either species. Based on our data it appears that beginning the night census at 19:00 offers little results in the early parts of the census but starting patrols past 22:30 poses the risk of missing turtles. Narrowing the range of our start times (19:00-00:00) may optimize turtle sightings, allowing us to grow our database further.

B. MINI ARRIBADAS

Interestingly, our nesting beaches have recorded both solitary and mini arribada nesting strategies. These mini arribadas seem to occur with a four week nesting interval between September and December, where up to 160 Olive Ridley turtles can be recorded laying in a single night. They occurred within the final quarter of the lunar cycle, one week before the new moon and span across a typical arribada span of three to four days (Matos et al., 2012). This directly coincides with the timing of the biggest arribada events in Costa Rica in Nicoya. Our beaches are located within feasible migratory distance from these beaches. It is therefore important that more research effort is placed on assessing these mini arribada events and investing more into tagging Olive Ridleys to assess similarities between the nesting populations and to an further understand both spatial and behavioural nesting plasticity in Olive Ridleys. Olive Ridley turtle populations are declining and considered vulnerable by the IUCN red list (Abreu-Grobois & Plotkin, 2008), but remain one of the least threatened turtles. However, as they are also one of only two species exhibiting the mass nesting behaviour their protection is vital.

C. TAGGING PROGRAM

Overall, the results from our tagging programme show that the beaches Rio Oro and Pejeperro present good nesting habitat with around one quarter of all greens returning to nest a second time. The data also seems to show that greens turtles nest more often than Olive Ridleys however far less Olive Ridleys were tagged during the season due to their IUCN Red List status being lower ('Vulnerable'; Abreu-Grobois & Plotkin, 2008) than the green's status (Endangered; Seminoff, 2004). Additional financial and capacity driven limitations led to greens being prioritised within the tagging programme though we plan to incorporate both species next year with additional funding. Only 4 green and two Olive Ridley individuals from the previous year were observed returning to nest. However, this is not atypical with green's exhibiting a common re-migration interval of two to three years (Troëng & Chaloupka, 2007) whereas Olive Ridley re-migration intervals have been found to range between one to eight years depending on the location (Tripathy, 2007). Comparing the emergence

interval and the nesting intervals it shows that for greens they emerge on the beaches approximately twice the amount that they nest, presenting a nesting success rate of around 50%. Other studies have found that a nesting interval is typically around two weeks between clutches (Chen & Cheng, 1995), suggesting that the turtles with an interval higher than around two to three weeks may either be nesting on another beach or that we missed their nesting event.

D. NEST EXCAVATIONS

Broadly, hatching success was consistent throughout the season, with spatial and temporal nest distribution having little effect. Similarly, when compared to last year, this season's hatching success for both beaches and for both species displayed little variance. However, due to some intrinsic faults with data handling and human error during data recording and excavations, many nests were lost or could not be included in the data analysis. Moving forward into the next season, a more robust and rigid protocol should be adopted to mitigate these issues along with more standardised procedures with data input. This year's results, coupled with last seasons represent a promising sign that Pejeperro and Rio Oro may be beaches with high hatching success rates.

From scientific literature and our own data, vegetation is known to be an important feature of nesting beaches for sea turtles, most notably for greens (eg. Turkozan *et al.*, 2011). This season's excavations have also demonstrated the importance of vegetation, especially trees in both the eclosion and emergence success of nests. Vegetation has also been suggested to be important in reducing the effects of climate change and the subsequent manipulations it has on skewed sex ratios, a pertinent issue that is ubiquitous in sea turtle populations globally (Kamel and Mrosovsky, 2006; Blechschmidt *et al.*, 2020). The findings of this season and from the literature, display the importance of the vegetation, mainly the trees, that line large areas of Pejeperro and Rio Oro for the future of nesting turtle populations and display the need for continual protection. Both eclosion and emergence success was significantly greater in sectors with trees. Vegetation increases nest shade and will subsequently reduce the variance in the sand and therefore nest temperatures (Kamel and Mrosovsky, 2006). These factors may allow for more successful incubation and reduce the chance of neonates exceeding their thermal capacity of 37°C.

In general, the wet and dry seasons generally did not affect nest success, a concurrent finding to Lolavar and Wyneken (2015). Although, the emergence success of Olive Ridleys was significantly higher in the dry season than the wet. A large proportion of Olive Ridley nests are laid in zone 2, an area above the high-tide mark but not in the vegetation. Therefore, the seasonal fluctuations will be larger for Olive Ridley nests on open areas of the beach, than for green, which lay many of their nests in the vegetation (Lolavar and Wyneken, 2015). Vegetation will regulate abiotic conditions, such as rain, wind, and temperature, which again leads to the understanding of the importance of vegetation for the stabilisation of nest conditions for greens. It is important to note that the green nesting season began at the end of the wet season, so a large proportion of the excavations were conducted on nests from the dry season.

Nest relocations are an essential part of many turtle projects globally (Wyneken *et al.*, 1988). However, because of poor field techniques, the introduction of fungus and bacteria into nests is common. Additionally, incorrect handling of eggs or relocating the nest too long after laying can result in early egg failure. Positively, this season's results display that the field techniques used are robust and had little negative effect on the success, or fungus and bacteria prevalence, in both Olive Ridley and green nests. Although, some sectors

displayed larger average fungus and bacterial infections than other sectors. This could be down to a multitude of factors, including large volumes of relocations and precedent pathogens.

It is also important to discuss that we had a very high predation rate observed on relocated nests (78% of the predations in our sample set were on relocated nests). This is likely due to a large proportion of our nests that were prioritized for relocation being laid in front of or around the Rio Oro river mouth. These were all relocated to a similar safe area on either side of the river, resulting in a much higher nest density at these sites than you would generally find on other areas of the beach. This high nest density seems to have drawn in a greater interest from predators, and therefore resulted in a lot of those nests being predated before hatching. In order to improve this next season, we will look to develop a small hatchery space for nest relocations at one side of the river, so that these nests can be protected and also serve for scientific studies that are difficult to undertake on nests left *in situ*.

VII. BIBLIOGRAPHY

Abreu-Grobois, A & Plotkin, P. (IUCN SSC Marine Turtle Specialist Group). (2008) Lepidochelys olivacea. TheIUCNRedListofThreatenedSpecies2008:e.T11534A3292503.https://dx.doi.org/10.2305/IUCN.UK.2008.RLTS.T11534A3292503.en.Accessed on 15 March 2022.

Beange, M., Clift, A. & Arauz, R. (2015) PRETOMA 2015 Report: Southern Nicoya Peninsula Sea Turtle Nesting Beach Conservation Projects.

Blechschmidt, J., Wittmann, M.J. and Blüml, C., 2020. Climate change and green sea turtle sex ratio—preventing possible extinction. *Genes*, *11*(5), p.588.

Chacòn, D., Sánchez, J., Calvo, J. J., & Ash, J. (2007). *Manual para el manejo y la conservación de las tortugas marinas en Costa Rica; con énfasis en la operación de proyectos en playa y viveros*. San José - Costa Rica: Sistema Nacional de Áreas de Conservación (SINAC) Ministerio de Ambiente y Energía (MINAE).

Chen, T.-H. & Cheng, I.-J. (1995) Breeding biology of the green turtle, Chelonia mydas, (Reptilia: Cheloniidae) on Wan-An Island, Peng-Hu Archipelago, Taiwan. I. Nesting ecology. *Marine Biology*. 124 (1), 9–15. doi:10.1007/BF00349141.

Eckrich, C.E. & Owens, D.Wm. (1995) Solitary versus Arribada Nesting in the Olive Ridley Sea Turtles (Lepidochelys Olivacea): A Test of the Predator-Satiation Hypothesis. *Herpetologica*. 51 (3), 349–354.

Lolavar, A. and Wyneken, J., 2015. Effect of rainfall on loggerhead turtle nest temperatures, sand temperatures and hatchling sex. *Endangered Species Research*, *28*(3), pp.235-247.

Kamel, S.J. and Mrosovsky, N., 2006. Deforestation: risk of sex ratio distortion in hawksbill sea turtles. *Ecological Applications*, *16*(3), pp.923-931.

Koval, J. (2015) Use of microsatellites to compare solitary vs arribada nesting olive ridley turtles (Lepidochelys olivacea) along the Eastern Pacific coast of Costa Rica. *Purdue UniversityProQuest DissertationsPublishing*. <u>https://www.proquest.com/openview/c90a1fa3848ebefed15d4da79745b515/1?pq-</u> <u>origsite=gscholar&cbl=18750</u>. Matos, L., Silva, A.C.C.D., Castilhos, J.C., Weber, M.I., Soares, L.S. & Vicente, L. (2012) Strong site fidelity and longer internesting interval for solitary nesting olive ridley sea turtles in Brazil. *Marine Biology*. 159 (5), 1011–1019. doi:10.1007/s00227-012-1881-1.

Pintus, K., Godley, B., Mcgowan, A., & Broderick, A. (2009). Impact of clutch relocation on sea turtle offspring.JOURNALOFWILDLIFEMANAGEMENT73(7),1151–115.Retrievedfromhttp://www.seaturtle.org/PDF/PintusKJ_2009_JWildlManage.pdf

Reina, R. D., Mayor, P. A., Spotila, J. R., Piedra, R., & Paladino, F. V. (2002). Nesting ecology of the leatherback turtle, Dermochelys coriacea, at Parque Nacional Marino las Baulas, Costa Rica: 1988–1989 to 1999–2000. *Copeia*, 2002(3), 653-664.

Seminoff, J.A. (Southwest Fisheries Science Center, U.S.). (2004) Chelonia mydas. The IUCN Red List ofThreatenedSpecies2004:e.T4615A11037468.https://dx.doi.org/10.2305/IUCN.UK.2004.RLTS.T4615A11037468.en.

Troëng, S. & Chaloupka, M. (2007) Variation in adult annual survival probability and remigration intervals of sea turtles. *Marine Biology*. 151 (5), 1721–1730. doi:10.1007/s00227-007-0611-6.

Turkozan, O., Yamamoto, K. and Yılmaz, C., 2011. Nest site preference and hatching success of green (Chelonia mydas) and loggerhead (Caretta caretta) sea turtles at Akyatan Beach, Turkey. *Chelonian Conservation and Biology*, *10*(2), pp.270-275.

Williamson, S.A., Evans, R.G., Robinson, N.J. & Reina, R.D. (2019) Synchronised nesting aggregations are associated with enhanced capacity for extended embryonic arrest in olive ridley sea turtles. *Scientific Reports*. 9 (1), 9783. doi:10.1038/s41598-019-46162-3.

Wyneken, J., Burke, T.J., Salmon, M. and Pedersen, D.K., 1988. Egg failure in natural and relocated sea turtle nests. *Journal of Herpetology*, pp.88-96.