The movement and distribution of pregnant spotted ragged-tooth sharks, *Carcharias taurus*, in the iSimangaliso Wetland Park, South Africa.

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#### **Abstract**

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The spotted ragged-tooth shark, Carcharias taurus, is widely distributed in subtropical continental coastal seas. In South Africa, it is commonly found along the entire south and east coasts, including the iSimangaliso Wetland Park (IWP) in the far north, which is the largest Marine Protected Area on the South African coast. Pregnant females occur there for much of the year, with the largest aggregations in summer. It is here we used remote Remote underwater Underwater photography (RUP) and underwater Underwater visual Visual census Census (UVC) surveys to photo-identify individuals, using unique spot patterns. Three known aggregation sites (Raggie Reef, Quarter-Mile Reef and Mushroom Rocks) were monitored between 2018 and 2023. We photo-identified 574 individuals (569 females and 5 males) and registered 1200 sightings, using images of the right flank. The identification of new individuals persisted throughout the study, with the discovery curve showing no signs of reaching an asymptote. A total of 97% (n = 550) of females observed were noticeably pregnant. Individuals were consistently identified across all sample years and at all three reefs, exhibiting movements between among the three monitored sites. The reproductive cycle is generally regarded as two years, but some females appeared to have a two-year rest between pregnancies. Raggie Reef, which lies in the sanctuary zone, emerged as the reef with the highest index of popularity, as individuals were present almost constantly (90% of the sampling days). The findings of this study confirm the crucial role that the IWP plays in the conservation of a species that is globally Critically Endangered.

#### Introduction

The spotted ragged-tooth shark, Carcharias taurus (Rafinesque 1810), inhabits subtropical and tropical coastal waters off continental land masses in the Atlantic, Pacific and Indian Oceans (Smith and Pollard, 1999; Compagno, 2001). It favours inshore rocky reefs, where it occurs close to the seabed, often in caves and under overhangs (Pollard et al., 1996). This species reproduces by oophagous viviparity, and exhibits intrauterine cannibalism, giving birth to two embryos (one per uterus) after a gestation of 9-10 months, followed by a year of recovery (Bass, 1975; Gilmore, 1983). This biennial reproductive cycle results in leads to low reproductive output, rendering making it susceptible vulnerable to anthropogenic impacts, especially particularly overfishing. In addition to fishing threats, the species faces habitat loss and degradation in inshore coastal waters, as well as the possible impacts of climate change (Rigby et al., 2021). As a result, thise species was globally listed classified as Critically Endangered on the IUCN Red List in 2020 (Rigby et al., 2021). In South Africa, the commercial sale of C. taurus is decommercialized prohibited, meaning making it is illegal to sell any part of theis species (Marine Living Resources Act 1998 (Act 18 of 1998). A population estimate from markrecapture data over a decade ago suggested a South African adult population of C. taurus at 16700 (CV = 9%) (Dicken et al., 2008). Klein (2020) indicated that this population remains healthy, potentially representing the last stable subpopulation of *C. taurus* globally.

**Açıklamalı [a1]:** CHECK PLEASE! In the reference list (line 453), it is written as Gilmore et al (1983)...

**Açıklamalı [a2]:** CHECK PLEASE! Is this "value" given as a number of individuals from relevant literature? And also please what "cv= 9%" means.

**Açıklamalı [a3]:** CHECK PLEASE! In the reference list (line 478), it is written as Klein et al (2020)...

Aggregation behavior is common among elasmobranchs, serving various functions such as facilitating courtship (Whitney et al. 2004), reducing predation risk (Guttridge et al., 2012), enhancing foraging efficiency (Dewar et al., 2008) and providing reproductive benefits (Wearmouth et al. 2012). Similar to other shark species, C. taurus demonstrate philopatry to certain areas (Hueter et al., 2005). In Australia, aggregations have been recorded at numerous specific sites, but there is only one documented site where females congregate during their pregnancy from September to January (Bansemer and Bennett, 2009). In the southwest Atlantic, pregnant individuals aggregate in subtropical waters of Brazil (Sadowsky, 1970), and after parturition, they move south where they rest, in cooler waters (Lucifora et al., 2002). The South African population of C. taurus is regularly found at specific sites alongside the east and south coasts, from False Bay in the extreme south to northern KwaZulu-Natal (KZN), and occasionally on the west coast (Bass, 1975; Smale, 2002). The KZN Sharks Board's catch and tagging data have been used to ascertain the distribution and migratory routes of this species in South Africa (Wallet, 1974; Dudley, 2002). Adults of both sexes move northwards from the Eastern Cape into southern and central waters of KZN, where mating takes place in late October and November (Dicken et al., 2006; Olbers and Cliff, 2017). Adult females in this population are under a well-defined reproductive migration. After mating, females continue to swim north to spend their pregnancy in northern KZN and southern Mozambique (Bass, 1975; Smale, 2002), most likely to take advantage of the seasonally warm water which accelerates metabolism and development (Bass, 1975; Bansemer and Bennett, 2009; Lucifora et al., 2002). The Agulhas Current, in the western Indian Ocean, transports warm tropical waters south to the eastern South Africa's regions (Schumann, 1998). This is responsible for the warm water temperatures in the iSimangaliso Wetland Park, Marine Protected Area (IWP), which provides an optimal environment for gestating C. taurus. Then, the near-term pregnant individuals return south, around July and August, to the Eastern Cape (Wallet, 1974), where parturition takes place in specific nursery locations from September to November (Smale, 2002, (Dicken et al., 2006). To date, the known aggregationng sites of pregnant females in South Africa have not been closely monitored, highlighting a gap in our understanding of the life history of this species. Here, we assemble compiled a dataset of individuals using photo-identification of individuals in the IWP over a five-year period. We determined the sex of these identified the gender of individuals and the status of pregnancy among females. We used interannual resightings to ascertain the duration of the species' reproductive cycle-of the species. We monitored local movement and distribution at three different reefs, while seeking to understand their carrying capacity and their respective popularity indices. These objectives collectively aspire aim to address significant knowledge deficits gaps pertaining regarding to the life history of *C. taurus*, thereby providing insights for the species' conservation and management and the role played by the IWP in their life history.

Materials & Methods Study Areas.

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97 98 Açıklamalı [a4]: CHECK PLEASE! There are two Dickens et al (2006) in the reference list (line 444 and 447). Therefore, these two works from the same year should be separated as "a" and "b" and the citations in the text should be indicated as "Dickens et al (2020a)" or "b". This correction includes the following lines: 71, 81, 269, 273-274, 278, and 357.

Açıklamalı [a5]: PLEASE MOVE TO ANOTHER SECTION! It would be more meaningful to move the information given here (between lines 67 and 81) to the "Discussion" section.

Aggregation sites for *C. taurus* were defined as places where five or more individuals were consistently observed across years (Otway et al., 2003). However, this definition lacked consideration of seasonal aggregations. Recognizing this, Hoschke and Whisson (2016) refined defined the concept of aggregation sites for this species as locations where five or more sharks gather on a frequently basis each year. The research focused on three known aggregation sites Three such known areas in the IWP-were the focus of this research: Raggie Reef (RR), Quarter-Mile Reef (QM) and Mushroom Rocks (MR) located on Seven-Mile Reef (Figure 1).

Quarter Mile ReefQM is located close to the public launch site (<1 km), in the Sodwana Diving Restricted Zone (SDRZ). It is situated between 500-800 m offshore at a depth of 10-12 m.

Raggie ReefRR is approximately 43 km south of the public launch site. It is situated in the iSimangaliso Offshore Wilderness Zone (IOWZ). No angling, scuba diving or spearfishing are permitted within this sanctuary. In comparison to QM, it is a far larger reef, approximately 250x180 m, at a depth of 10-14 m. Seven-Mile Reef is approximately 11 km to the north of the launch site, 800 m offshore, at a depth of 14-25 m. It is bigger than QM and RR, being 1400x390 m. However, only a small portion of this reef was monitored, designated Mushroom Rocks

Açıklamalı [a6]: PLEASE DELETE: It might be more appropriate to make such a statement in the "Discussion" section.

Açıklamalı [a7]: PLEASE DELETE OR CHECK! This abbreviation is not referenced in this way anywhere else in the text. In several places the words "Sodwana" or "Sodwana Bay" (lines: 270, 330, 332 etc.) are mentioned.

#### Sampling.

 Sampling was conducted carried out over five consecutive seasons from 2018/2019 to 2022/2023 during the months of from September to March, which coincided coinciding with the peak abundance of *C. taurus*. The first sampling season, which began in December 2018, was an exception One exception was the first sampling season, which commenced in December 2018. In this study, summer months were considered as December, January and February.

(MR), as this is by far the most common congregating site on 7Seven-Mile Reef.

Açıklamalı [a8]: PLEASE EXPLAIN! Was the system (RUP) described and established here based on previous studies or is it a method used for the first time in this study? It would be good to make an explanation on this subject.

#### Remote Underwater Photography (RUP).

GoPro Hero3 or 4 cameras were used for the Remote Underwater Photography (RUP). Each photographic unit (Figure 2) consisted of two cameras, positioned at 90° to one another to broaden the field of view and set to take images in wide angle at 30–second intervals. Each camera was connected to an external battery (50 Ah or 100 Ah), through a USB cable. The two cameras and batteries were placed inside a sealed Plexiglass tube housing (20 cm in diameter and 25 cm in height). The camera housing was mounted 1 m above the seabed on a vertical pole which was secured to a 45 kg base. Freedivers deployed units and retrieved them for servicing and cleaning every 6–12 days. Each pair of cameras was manually synchronized to capture images at the same time.

The RUPs were installed at three sampling sites, for different sampling durations. The RUP at QM was first deployed in 2019/2020 season, and every sampling season thereafter. At RR, the RUP was first deployed in 2020/2021 and for the subsequent two sampling seasons, while the RUP was only installed at MR, during the fourth sampling season, 2021/2022.

Diver observations.

During the Underwater Visual Census (UVC), freedivers used handheld GoPro Hero5 or 6 cameras to capture images of *C. taurus* whenever visibility and circumstances allowed. They also counted the maximum number (MaxN) of sharks present at the reefs. An Underwater Visual Census (UVC) was undertaken by freedivers when visibility and circumstances permitted, with the purpose of capturing images of *C. taurus* with handheld GoPro Hero5 or 6 cameras, while also counting the maximum number (MaxN) of sharks that were present at those reefs. Weather conditions were a very important factor in dive observations. QM reef was surveyed almost every day during the summer months, weather conditions permitting, since it was the most accessible reef. It was first surveyed in the first sampling season, 2018/2019. Surveys at RR were undertaken every 10 days, weather permitting, with the longest gap being 12 days. This reef was first surveyed during the second sampling season, 2019/2020. As MR was a deeper reef, snorkel surveys were not suitable, so scuba surveys were conducted instead of snorkel surveys in 2019/2020 and in 2021/2022.

Photo-identification.

All the images were downloaded, with and only those the ones suitable for photo-identification being were selected. The selected images show, such that the shark's entire right side of the trunk was with visible, with spots present on its body, as well as and both dorsal fins and the pelvic fin present and visible in the frame. Only the images on the right-side images were used, to avoid double counting. The images were then analyzed with the I3S Classic version 4.02 software as described by van Tienhoven et al., 2007 (Interactive Individual Identification System, https://reijns.com/i3s/) to identify each individual based on using their natural spot patterns requiring with a minimum of 12 identifiers and a maximum of 30. This necessitated selecting three reference points on the body in each image: at the base of the first dorsal fin, the base of the second dorsal fin and the base of the pelvic fin (Figure 3).

The darkest spots were chosen first, as they were most likely to be seen in all conditions. Once every point was selected, the image of that individual was compared with the individuals already in the database. The matches were then revealed, each with a score, with the lowest score representing the closest match to the unknown individual. Typically, scores above 20-30 indicated different individuals, though this could vary considerably based on lighting conditions and other image factors. Scores below 10 were almost always accurate, with a score of 5 being highly likely to indicate the same individual. Photo-identification methods can lead to two potential errors: 1) false positive, incorrectly matching two unique individuals as the same one or; 2) false negative, where a known individual is not identified, thus classifying it as a new one. Therefore, a visual comparison was always necessary, to compare specific spots on the two sharks, and the individual was either matched positively or recorded as a new individual. To minimize errors of subjectivity, the combination of the I3S automated system and a manual pattern—matching exercise was undertaken by the same observer. After the identification was complete, the images of all females were examined, in order to detect any evidence of

Açıklamalı [a9]: PLEASE EXPLAIN! What standards were adopted as criteria when using the UVC technique? Were the observation hours the same and carried out in a standard manner (for example, between 09:00 and 12:00 in the morning)? Were transects/transects determined for the observations (for example, 100 m long) and were the observations/counts always made on the same line?

Açıklamalı [a10]: PLEASE CHECK AND EXPLAIN! The text says "the lowest score representing the closest match to the unknown individual" (lines 156-157). The next section explains the high scores (above 20-30). At this point, confusion arises: "scores below 10 were almost always accurate, with a score of 5 being highly likely to indicate the same individual". Now, what should be explained by the "lowest score"? If a score of "5" represents the "same individual", what does "the lowest score represented the closest match to the unknown individual" mean?

pregnancy. If the female had mating scars and/or a noticeably distended abdomen, with lateral bulges, it was considered to be pregnant (Bansemer and Bennett, 2009).

#### Index of popularity and density of each site.

The index of popularity reflects the relative frequency of <a href="sharks">shark</a> occurrences</a>—of sharks at the aggregation site, <a href="irrespective-regardless">irrespective-regardless</a> of the number of sharks. <a href="In order to To assess determine">In order to To assess determine</a> the density or carrying capacity of each site, <a href="which is">which is</a> based on the MaxN, the analysis <a href="exclusively relied on\_only used">exclusively relied on\_only used</a> UVC data. <a href="Ithis was necessary because-due to the inherent limitation of RUP imagery has limitations and only">has limitations and only</a>, <a href="which which provides a field of view of up to 180° from a fixed point.">fixed point</a>.

#### Results

#### Sampling effort.

Sampling effort from both RUPs and UVCs varied throughout the study. QM experienced the highest effort, followed by RR and lastly, MR ([Table 1]). Over the entire survey period, UVC sampling effort per reef revealed notable differences, with QM emerging as the most extensively sampled (n = 482 sampling days), as opposed to RR (n = 76 sampling days) and MR (n = 36 sampling days). Sampling effort per month was uneven, with the summer months (December, January and February) being the most heavily sampled between 17 to 31 days.

#### Identification of individuals with photo-identification.

In this study, 574 individuals were identified between 2018 and 2023 (Figure 4). The discovery curve shown has not yet reached an asymptote, with a steady increase in the number of newly identified individuals over the five-year study. A total of 1200 sightings of these 574 identified individuals were recorded. It is important to note that a sighting is counted each time an individual shark is identified, even if it has been identified before. For example, if an individual shark is identified on one day and then identified again on the next day, this counts as two sightings for that one individual. Most (62.9%, n = 358) individuals were only sighted once, but 37.1% (n = 213) were resighted at least once, predominantly in the same sampling season (Figure 5), with a single individual sighted 19 times across the 5 year-long study period. Of the resighted individuals, only 2.1% (n = 12) were resighted in a subsequent sampling season. The sex ratio of the population was 99% (n = 569) females and 1% (n = 5) males.

#### Pregnancies.

During the five-year survey period, 97.0% of females were classified as pregnant, based on mating scars and/or a distended belly (n = 550), while the status of 3.0% was uncertain (n = 19). In some pregnant individuals who were seen several times in a particular season, it was possible to see the belly enlarging with time (Figure 6). Of the 550 females identified over the entire survey period, 12 were resighted in different sampling seasons, and were always considered to be

**Açıklamalı [a11]:** Please review the additional file (peerj-102837-Table\_1) for table editing and suggestions.

Açıklamalı [a12]: PLEASE CHECK AND EXPLAIN! The section under the heading "Sampling effort" and the data presented in Table 1 are quite confusing. What should the reader understand by the term "Sampling effort" here? Are we considering the number of sightings as 1200 (data in lines 28 and 189)? Or the number of observations as 898 (total in Table 1 as 359+484+55)? Or the number of identified sharks as 574? Or the number of sampling days as 594 (482+76+36 days)?

In addition, in order to make Table 1 more explanatory and to enable a more accurate comparison of the study data, the following is suggested. Researchers should standardize their data as "number of sharks" per "observation hour" or "number of observation days". Thus, the actual effort per unit effort can be seen (as CPUE). A CPUE pre-calculation was made by me by adding to Table 1 (please see additional word file; peerj-102837-Table\_1). In these calculations, the highest effort was in RR.. Researchers should use such an approach.

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pregnant (Table 2). Three females were resighted with a one-year interval between sightings and nine females were resighted with a two-year interval.

#### Description of the sharks' movements within the IWP.

Over the entire study period, 29 of the 213 individuals that were resighted were found at a different reef from where they were first detected. There was a total of 33 movements, of which 19 were heading north and 14 south. There are no significant differences between northward and southward movements ( $\chi^2$ : 2, N = 33, p = .34). Considering the northward movements, 11 were from RR to QM, three from QM to MR and five from RR to MR. Of the 14 southward movements, there were 11 from QM to RR, two from MR to QM and one from MR to RR. The majority of northward movements (n = 15) were between December and January, as opposed to those which moved south, which were mostly between February and March (n = 9).

#### Index of popularity and shark abundance of each site.

Throughout the survey period, *C. taurus* were observed on 223 out of 484 (46.0%) sampling days at QM. In contrast, sharks were recorded on 325 out of 359 sampling days at RR, constituting a 90.0% presence. Insufficient sampling was conducted at MR to determine an index of popularity.

Sightings of *C. taurus* fluctuated each year, with 2022/2023 having higher numbers on mean and 2021/2022 having lower numbers on mean (Table 3). The mean number of *C. taurus* aggregating at once was constantly observed at RR. Raggie Reef consistently exhibits higher values of mean MaxN per day compared to QM, notably in December of 2021 (n = 18 for RR vs. 9 for QM) and January of 2022 (n = 33 for RR vs. 1 for QM). Regarding shark abundance, RR had the highest numbers, with 114 individuals of *C. taurus* observed in a single day, followed by QM (n = 55) and lastly MR (n = 14) (Table 3). Significant differences of MaxN values were observed among the reef sites, according to the Kruskal-Wallis test (H = 61.52, p < .0001). The mean MaxN per day showcases some monthly fluctuations, but with consistent peaks observed during the summer months, highlighting a recurring trend in shark abundance. December, January, and February consistently emerge as months with higher MaxN values across sampling seasons (Figure 7).

#### **Discussion**

The results of this study indicate that the three surveyed reefs in the iSimangaliso Wetland Park (IWP) meet the criteria for *C. taurus* aggregation sites, as proposed by Hoschke and Whisson (2016). Aggregation behavior is common among elasmobranchs, and this species is no exception, being well-known for its gregarious nature (Olbers and Cliff, 2017; Haulsee et al., 2016). Pregnant females of *C. taurus* are known to form groups, a behavior also observed in other various parts of the world (Hoschke and Whisson, 2016; Bansemer and Bennett, 2009). Courtship is a common cause of aggregation in many species (Whitney et al., 2004), however, these aggregations occurred long after mating. Wearmouth et al. (2012) demonstrated how

Açıklamalı [a15]: It was written as 482 sampling days in line 182. Confusion arose again regarding the numbers... What is the data taken into account? If it is "Sampling days" then for example it should be 482 for QM. The 484 data here should be the "total number" in Table 1, and no explanation is given as to what this data is

Açıklamalı [a16]: It was written as 76 sampling days in line 182. Confusion arose here again regarding the numbers...

aggregations can provide reproductive benefits by minimizing sexual harassment from males, but, again, these aggregations occurred long after mating. Reducing predation risk is another potential benefit of such aggregations (Guttridge et al., 2012). Considering the large size of these sharks (±190 cm PCL) and the scarcity of natural predators, it seems improbable that they congregate to reduce risk of predation. Foraging efficiency is frequently improved by group behavior (Dewar et al., 2008) yet the presence of hydroid growth on the teeth of these sharks suggests that they may not feed much during these aggregations (Pollard et al., 1996).

#### Population assessment.

In this study, 574 distinct individuals were identified from just three sites in the IWP, over a five-year sampling period, with no sign of the discovery curve attaining an asymptote. Our study showed 569 mature females, which contrasts with research from the east coast of Australia, where only 271 mature females were identified across 19 aggregation sites (Bansemer and Bennett, 2011).

Throughout the five years of survey, the summer months of December, January and February had the highest numbers of identified individuals and average MaxN per day. Local water temperatures are highest in February (Staiger, 2020), which enhances embryo development and reduces gestation time, especially in ectothermic species (Bass, 1975; Bansemer and Bennett, 2009). This trend is evident in other coastal shark species, where pregnant females also aggregate in warm waters to accelerate gestation, displaying seasonal fidelity to such sites, such as the leopard shark, *Triakis semifasciata*, in southern Carolina, USA (Nosal et al., 2014) and the tiger shark, *Galeocerdo cuvier* in the Bahamas (Sulikowski et al., 2016).

#### Reproductive cycle.

The population in the IWP between September and March is predominantly female (n = 99.0%). This aligns with the findings of Dicken et al. (2006) who observed that *C. taurus* catches by anglers from Sodwana Bay to Richards Bay between November and February, were dominated by large females. The clear dominance of females is because most of the mating takes place to the south of the IWP and only the females continue northwards, while the whereabouts and movement patterns of mature males outside of the mating season remains uncertain (Dicken et al., 2006). Over the course of five years, 97.0% females appeared to be pregnant. Those not visibly pregnant at the time were probably in the early stages of pregnancy, where signs were not yet apparent. Non-pregnant females are unlikely to be in IWP. The high incidence of adult females in summer in the Eastern Cape was hypothesized as being individuals who were in resting between pregnancies (Dicken et al., 2006).

Research has demonstrated a biennial reproductive cycle for this species in both the NW Atlantic (Branstetter and Musick, 1994, Henningsen et al., 2004), SW Atlantic (Lucifora et al., 2002); and Australia (Hoschke and Whisson, 2016). The present study adds an additional perspective, as there is evidence suggesting both biennial and triennial reproductive cycles. Three pregnant females were resighted after a one-year interval, which is indicative of the well-documented

biennial cycle, with a one-year resting period. Yet, nine females were resighted only after a two-year interval, which is suggestive of a triennial cycle, with a two-year resting period. Although this appears surprising, it should be noted the triennial cycle was also described for 9 females in the Australian east coast (Bansemer and Bennett, 2009). These findings have important repercussions for a species which, based on a two-year cycle, already has a low reproductive output but now it may be reduced by 33.0%. This could result in reduced population recruitment and increased vulnerability to threats, such as habitat loss and overfishing. As shown in other studies, elasmobranchs are particularly vulnerable to overfishing and present a limited capacity for population recovery, compared to most teleosts (Myers and Worm, 2005; Stevens et al., 2000). Future research should focus on explaining the drivers of this variability in such cycles and its implications for the population. Additionally, it would be valuable to investigate whether the triennial cycle is associated with smaller females, as our study did not assess female size. Smaller individuals might potentially outgrow this pattern and revert to a biennial cycle with age.

#### Description of movements.

Throughout the study period, the observed movements almost exclusively involved mature females presumed to be in a pregnant state. Since there are movements between among sites, there is no site fidelity to a specific reef. This is in contrast to the findings of Klein et al. (2019) who demonstrated the existence of genetically differentiated nursery areas, with female C. taurus returning to specific sites, thereby exhibiting reproductive philopatry. To date, no studies have explored whether C. taurus consistently travels with the same group of individuals. Most of the northward movements were between December and January, while southward movements were predominantly observed between February and March. These migration patterns align with literature documenting the coastwise movements of pregnant females in South Africa (Bass, 1975; Dicken et al., 2007) and in the southwest Atlantic, where the pregnant individuals are found aggregating in subtropical waters (Sadowsky, 1970), and after parturition, they move south where they rest, in cool waters (Lucifora et al., 2002). On a broader scale, knowledge of coastal migration patterns is essential for efficient management (Bonfil, 1997), but unfortunately, available data is frequently lacking. For coastal resident sharks that migrate seasonally, like C. taurus, this problem is particularly important (Speed et al., 2010). Marine Protected Areas (MPAs) are a useful and effective strategy for protecting coastal sharks, especially for species that rely on specific areas for nursery, reproduction or feeding (Speed et

#### Spatial distribution.

Raggie Reef had higher sighting rates than the other two sites throughout the study period, with a remarkable 90.0% presence of *C. taurus*. These results can be attributed to its geographical location (Figure 1). Being the southernmost and by far the largest of the three study sites, it is possibly the first reef in the IWP to receive pregnant females coming from the south, and also the

gestation period, highlighting the relevance of such areas and the species' reproductive success.

al., 2010). For instance, C. taurus emphasizes the value of MPAs by using IWP during its

last known stop before heading back south to pup. Most importantly though, it is zoned as a sanctuary area, i.e., no fishing, or diving activities are permitted in the IOWZ (South Africa, 2019), with the exception of bona fide research, as was the case for this project. This sanctuary status ensures that pregnant females are very rarely disturbed at RR. On the other hand, diver disturbance is a problem at Quarter Mile Reef, which has long been recognized as an important site for *C. taurus*. Because of its extremely close proximity from the launch site, scuba diving with *C. taurus* at QM has become a popular dive attraction in Sodwana (Dicken, 2014). This proximity to the launch site can also be a source of disturbance, (Koper and Plön, 2012). The small vessels used at Sodwana Bay are all equipped with twin outboard engines and are required to negotiate the surf zone at high speed, generating noise that falls within the peak sensitivity range of sharks (Casper, 2006). These two factors combined could adversely influence the numbers of sharks at QM. This resulted in the introduction of a park management initiative to close the reef when the sharks first arrive at QM and to only open the reef to scuba diving when the sharks appear to have settled (Olbers and Cliff, 2017).

The three aggregating sites are the only ones known in an MPA which spans some 180 km of

The three aggregating sites are the only ones known in an MPA which spans some 180 km of coastline. It is also uncertain why these reefs are preferred over others, despite extensive surveying of other reefs. One new site was discovered in January 2022, where an aggregation of close to 50 individuals was found at a new location, aptly named Raggie Garden, a short distance offshore from RR. This reef is at 22 m, far deeper than the three sites used in this study, but well within the depth range (< 40 m) of this species (Bennett and Bansemer, 2004; Otway and Ellis, 2011; Hoschke and Whisson, 2016). Although the newly discovered reef was not included in this study, the search for more aggregation sites is ongoing.

#### Conservation.

Since our discovery curve has not reached an asymptote after 574 identifications, it supports the conclusion that the South African population seems to be the last remaining stable subpopulation of *C. taurus* globally (Klein, 2020). Furthermore, the latter genetics-based study suggested that the species has shown no decline in the region. By contrast, Otway et al. (2004) indicated that Australia's south-eastern coast population has been decreasing severely (minimum of 300, most likely < 1000 individuals), with quasi-extinction predicted to be less than 45 years away, unless all anthropogenic, largely fishing-related, mortalities are eliminated. Currently, recreational shore angling for this species is permitted in the IWP, with the stipulation that all the catches are returned to the water alive, but the possibility of post-release mortality cannot be eliminated (Dicken et al., 2006; Otway and Burke, 2004; Bansemer and Bennett, 2010). Therefore, angler education is important to ensure correct handling. Even if survival rates of released sharks are high, females may still lose their pups through abortion (Adams et al., 2018).

Further, Olbers and Cliff (2017) proposed an improved code of conduct for divers to minimize disturbance or stress, which may cause sharks to relocate from aggregation sites like QM or MR.

Other studies have shown that scuba diving affects the behavior and distribution of C. taurus

**Açıklamalı [a17]:** CHECK PLEASE! In the reference list (line 518), it is written as Republic of South Africa (2019)....

**Açıklamalı [a18]:** CHECK PLEASE! In the reference list (line 478, it is written as Klein et al (2020)...

(Barker and Williamson, 2010; Barker et al., 2011). In this regard, smaller groups of recreational divers, governed by a clear code of conduct around the sharks, would be highly beneficial.

#### Conclusions

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402 403 Through the establishment of a comprehensive five-year database, it was possible to monitor the use by pregnant C. taurus of the iSimangaliso Wetland Park, where they spend much of their gestation. This study has contributed to a better understanding of localized movements and the distribution of pregnant females at only three known aggregation sites within this Marine Protected AreaMPA spanning 180 km of coastline. A suggestion for future studies is to capitalize on the species' tendency to aggregate at specific, known sites. By combining this understanding with increased sampling efforts across diverse sites, it may be possible to reduce periods of absence between sites, leading to a more accurate understanding of individual movement patterns. It would be highly beneficial to continue the monitoring at these locations, with a view to quantifying the size of the mature female component of the population and the extent of a triennial reproductive cycle. Another recommendation is extending the sampling to year-round at all three sites - a practice that has recently been implemented at RR (G. Smith, pers. comm). The search for more sites should continue to improve our understanding of finescale habitat use within the MPA, along with a need to understand the factors that drive movement between sites. The identification of 569 mature females strongly supports the contention of a stable population, which is not Critically Endangered, like others elsewhere.

#### **Acknowledgements**

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**Açıklamalı [a19]:** CHECK PLEASE! It was observed that this study was not cited in the text.

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	Reefs		Moni	toring Per	riod(s)	Total	Sampling Days	<u>CPUE</u>	
		18/19	19/20	20/21	21/22	22/23			
Sampling effort	RR	-	9	100	142	108	359	<u>76</u>	<u>4.7</u>
	QM	69	108	132	86	89	484	<u>482</u>	1.0
	MR	-	23	-	32	-	55	<u>36</u>	<u>1.5</u>

For example, if we consider only MaxN (as N) and sampling days (as SN):

 $\underline{\mathsf{CPUE}} = [\underline{\Sigma_{\mathsf{N}}} / \underline{\Sigma_{\mathsf{SN}}}]$ 

 $\Sigma_{N}$  = Total number of sharks observed on the Reef

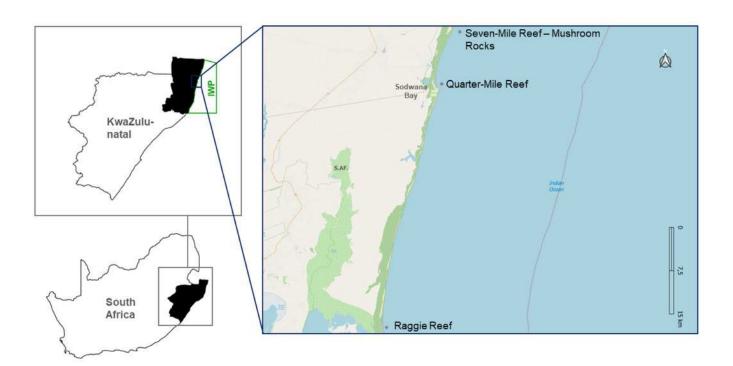
 $\Sigma_{SN}$  = Total number of observation days on the Reef

**Açıklamalı [a1]:** 359+484+55= 898. A number not existed in the text!!!



Map depicting the study sites, in a Marine Protected Area.

Map of South Africa highlighting KwaZulu-Natal province and the specific locations of the three study sites, within the iSimangaliso Wetland Park (IWP). The study sites include Raggie Reef (RR), Quarter-Mile Reef (QM) and Seven-Mile Reef (7M), with a focus on the Mushroom Rocks (MR) area.



Components and deployment of the Remote Underwater Photographic (RUP) system.

(A) RUP housing equipped with GoPro cameras and batteries, specifically designed for underwater imaging. (B) Unit without underwater housing. (C) The RUP system installed at the study site, actively capturing images.

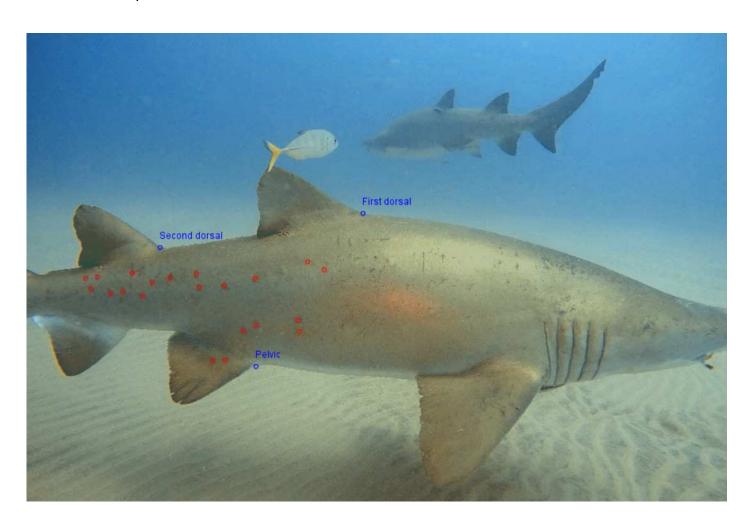






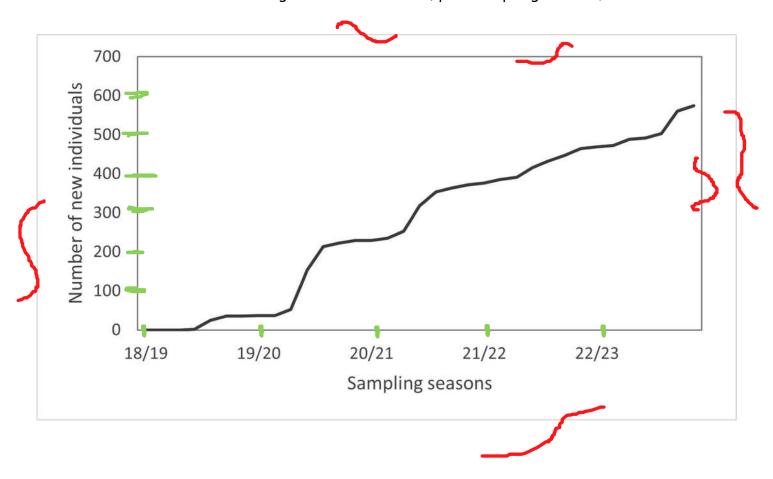
Reference and identification points for photo-identification of Carcharias taurus.

The three blue points are used by the I3S as reference points for the photo identification process: the first dorsal fin near the top, the second dorsal fin midway along the back, and the pelvic fin on the underside. The red points indicate distinctive spots on the shark's body, which are unique to each individual.



Cumulative discovery curve of newly identified Carcharias taurus individuals.

The cumulative discovery curve depicts the number of newly identified individuals of *Carcharias taurus* in the iSimangaliso Wetland Park, *per* sampling season, from 2018 to 2023.



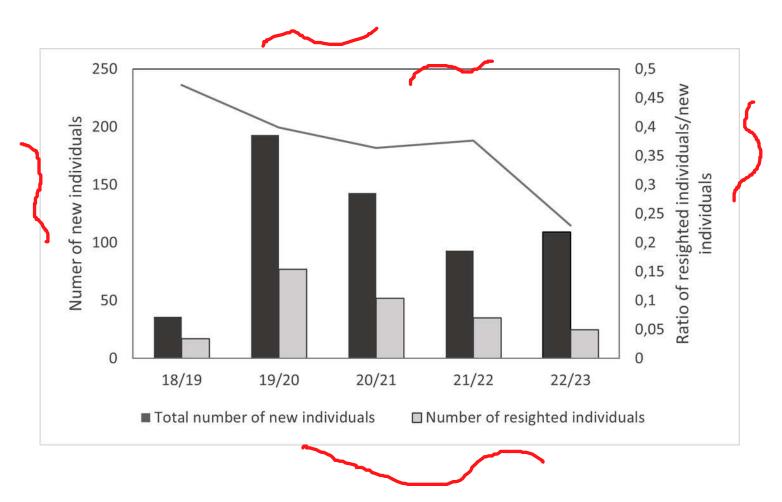
PLEASE DELETE: Romove the frame (drawn in red) seen around the graphic by deleting it

CONSIDER SUGGESTION: Show the "interceps" on the "x" and "y" axes of the graph (shown in green). Also, shiw "the number of individuals" intersecting with "year" on the graph line.

THESE CHANGES AND SUGGESTIONS SHOULD BE TAKEN INTO ACCOUNT FOR FIGURE 5 & 7



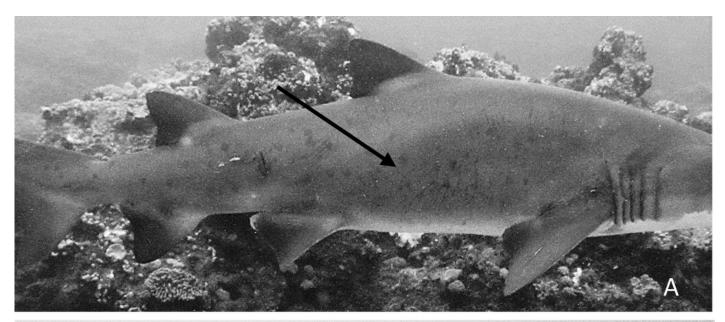
Total number of newly identified individuals of *Carcharias taurus*, alongside the number of individuals resighted during that season and respective ratio.

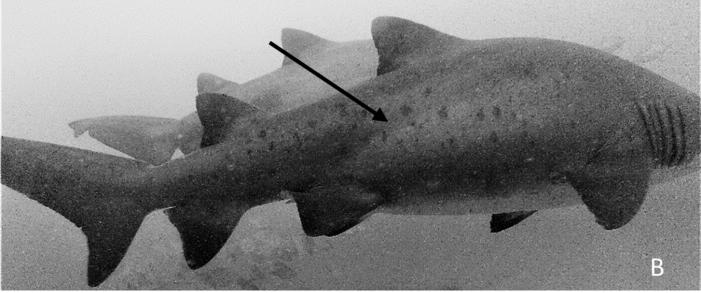


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Carcharias taurus: pregnancy observation.

(A) Female individual seen at RR on 21<sup>st</sup> November 2021, with no visible signs of pregnancy, and B) the same individual, seen at RR on 24<sup>th</sup> March 2022, with clear signs of pregnancy.

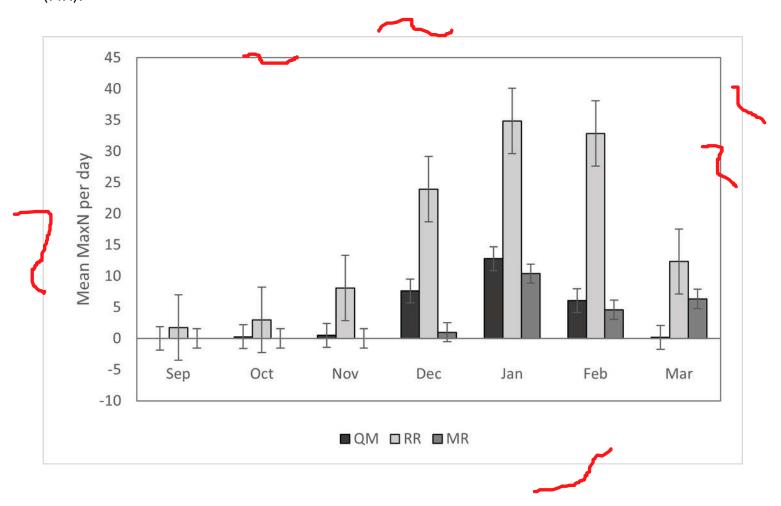




Seasonal variation in Carcharias taurus abundance at three sites (QM, RR and MR).

Mean MaxN per day across sampling months, over five consecutive years (2018 to 2023).

Data is shown for study sites Quarter-Mile Reef (QM), Raggie Reef (RR) and Mushroom Rocks (MR).



PLEASE DELETE: Romove the frame (drawn in red) seen around the graphic by deleting it CONSIDER SUGGESTION: Show the "interceps" on the "x" and "y" axes of the graph.



### Table 1(on next page)

Sampling effort (number of sampling days), for both Remote Underwater Photography (RUP) and Underwater Visual Census (UVC) methods.

This table presents the sampling effort for both RUP and UVC methods, across study sites over the five-year study period. It includes the number of days each site was sampled per sampling season, along with the total sampling effort for each site. Raggie Reef (RR), Quarter-Mile Reef (QM), Mushroom Rocks (MR).



	Reefs	18/19	19/20	20/21	21/22	22/23	Total
Sampling	RR	-	9	100	142	108	359
effort	QM	69	108	132	86	89	484
	MR	-	23	-	32	-	55

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PLEASE CHECK: Please check the word file of Table 3 (peerj-102837-Table\_1) for correction and suggestion.

IMPORTANT CORRECTION: The most important point in this article is that it does not have a clear statement about what should understand by term "sampling effort". In different sections in the text, different numbers (sometimes MaxN, sometimes "sampling days") are used as "effort". Then, suddenly we are faced with completely different number ("Total" and data in the table). What is meant by "effort" and which data is taken into account should be clarified "definitively" and "numerically".



### Table 2(on next page)

Pregnancy history of resighted Carcharias taurus' females.

Reproductive patterns of females, including the season of their first pregnancy recorded in this study, years in between sightings, and second pregnancies observed, over the five-year sampling period.  $1^{st}$  = first pregnancy observed; \* = years without sighting such individual;  $2^{nd}$  = second pregnancy observed.

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ID	Sampling seasons							
ID	18/19	19/20	20/21	21/22	22/23			
27_Ct_F	1 <sup>st</sup>	;	k	2 <sup>nd</sup>	-			
37_Ct_F	1 <sup>st</sup>	;	k	2 <sup>nd</sup>	-			
49_Ct_F	1 <sup>st</sup>	:	k	2 <sup>nd</sup>	-			
99_Ct_F	-	1 <sup>st</sup>	resting years		2 <sup>nd</sup>			
106_Ct_F	-	1 st	*		2 <sup>nd</sup>			
122_Ct_F	-	1 <sup>st</sup>	*		2 <sup>nd</sup>			
281_Ct_F	-	1 st	* 2 <sup>nd</sup>		-			
334_Ct_F	-	1 <sup>st</sup>	*		2 <sup>nd</sup>			
375_Ct_F	-	1 st	*		2 <sup>nd</sup>			
403_Ct_F	-	1 <sup>st</sup>	*		2 <sup>nd</sup>			
489_Ct_F	-	-	1 <sup>st</sup>	*	2 <sup>nd</sup>			
511_Ct_F	-	-	1 <sup>st</sup>	*	2 <sup>nd</sup>			



### Table 3(on next page)

Variation in Carcharias taurus' density at different reefs over five sampling seasons.

The table displays the maximum (Max) MaxN and the mean of MaxN per day recorded at Quarter-Mile Reef (QM), Raggie Reef (RR) and Mushroom Rocks (MR) across five consecutive sampling seasons.



1				1				
2	Sampling	M			RR	MR		
	season	Max	Mean	Max	Mean	Max	Mean	
	18/19	33	4,9	1	-	۱ (	-	
	19/20	48	8,5	41	24,6	11	6,4	
	20/21	44	4,2	45	17,8	ı	-	
	21/22	48	2,6	62	11,2	14	6,1	
	22/23	55	10,2	114	23,1	-	-	

MaxN

If the average values in Table 3 were prapered based on data between lines 219 and 220, it is recommended that they be checked again. Because the data presented here and the data presented Table 1 seem to be different. Also, in lines 182 and 183 of the article, different data are shown (as sampling days). A confusing situation..