

CASE REPORT

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ANTHROPOLOGY

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Nonfatal Bites by a Sickfin Lemon Shark *Negaprion acutidens* on a Surfer in Makemo Atoll (French Polynesia)*

ABSTRACT: Identifying the species and size of sharks responsible for biting humans is essential for developing strategies to prevent these incidents. Here, we use bite wound characteristics and genetic analysis of a tooth fragment extracted from the wounds to identify a sickfin lemon shark *Negaprion acutidens* as the perpetrator of nonfatal bites on the legs of an adult male surfer at Makemo atoll (French Polynesia) in January 2018. The bite was superficial, and *N. acutidens* are fish predators not known to feed on large prey; hence, foraging is an unlikely explanation for this incident rather linked to territoriality. Lemon sharks are occasionally aggressive toward humans and are site attached with relatively small home ranges; hence, avoiding surfing in the area of a previous bite incident is recommended to decrease the risk of future injuries.

KEYWORDS: forensic science, dangerous marine wildlife, agonistic behavior, attack motivation, inter-dental distance, tooth DNA analysis

Shark bites are extremely rare events with 66 confirmed cases worldwide in 2018 (1) that receive disproportionate and sensationalized media attention leading to a distorted public perception of risk despite fatalities occurring in < 10% of these incidents (2). The slight increase in total yearly bites since 2000 is likely explained by the increasing number of people participating in ocean recreation (3) and the development of new watersports that increase the probability of shark–human interactions in areas previously unused by humans (4). For example, reef passages connecting lagoons and open ocean in tropical waters are both natural hotspots for marine predators such as sharks (5) and increasingly popular locations for surfing. Surfing is the highest risk activity for unprovoked shark bites, with surfers accounting for 59% of all nonfatal bites worldwide (1). Three shark species are responsible for most (>70%) agonistic strikes on surfers: the white shark *Carcharodon carcharias* in temperate locations, and the tiger shark *Galeocerdo cuvier* and bull shark *Carcharhinus leucas* in warm tropical waters (1). Other coastal shark species rarely bite humans (6). The motivation for shark bites is not always clear, but those resulting in massive loss of tissue are likely driven by foraging whereas those resulting in “bite and release” type behaviors or lacerations of a slashing type may stem from exploratory, competition, or defensive

behaviors (7,8). The reliable identifications of both the shark species and bite motivation of a surfer are then very relevant for the inherent risk assessment and management issues in terms of human safety.

Here, we use the case study of nonfatal bites by a single individual shark putatively identified as a *Negaprion acutidens* on a surfer in French Polynesia to identify the species involved in the incident and to discuss the implications in terms of bite motivation and the management of the risk in the presence of this specific shark species.

Material and Methods

Shark Bites Background

On January 17, 2018, a 48-year-old individual was surfing with a friend in the vicinity of the remote northwest reef passage at Makemo atoll (Fig. 1). They arrived at the surf spot at 7:00 AM and at 7:30 AM, and they were joined by three other surfers. No other human activities were occurring in the area. The waves were approximately 2 m high with an outgoing current. The victim noticed some bigger waves coming from the outside corner of the reef and decided to paddle out to try and catch one. Surfers rarely frequent this particular section of the reef. This was around 8:00 AM; the water was 2.5 m deep, and the visibility was > 15 m on the outer slope of the barrier reef. As the victim was changing from a prone (lying on his board) to a vertical (sitting) position, he felt a hard hit on his right leg above the knee. He looked back but did not see the shark and started to paddle away when he then felt a second harder hit on his left leg on the knee. The bites were three seconds apart. The shark grasped the appendage after the second bite. The victim struck the shark's snout with his left fist. The shark released the

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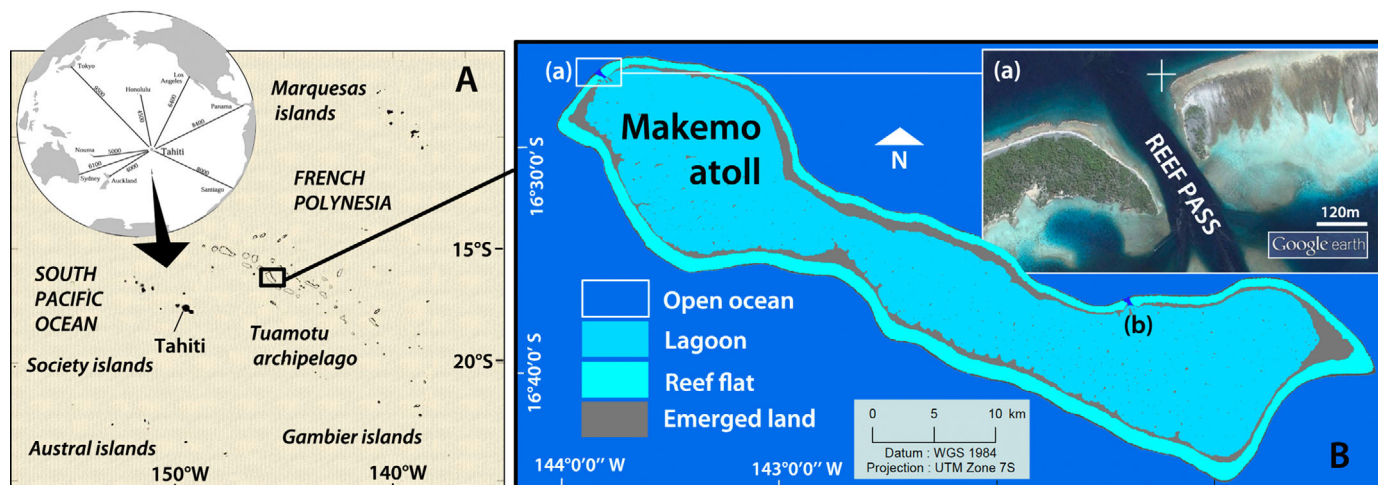


FIG. 1—(A) Location of the Makemo atoll belonging to the central atolls of the Tuamotu archipelago northeast to the Society islands including Tahiti in French Polynesia (Central-Eastern Pacific). (B) This large atoll holds two reef passages: (a) to the northwest—see close-up on the top right that indicates the spot of the bites on the surfer (white cross)—and (b) to the northeast. Those reef passages offer an open mouth to the ocean with two reef outer slopes in both sides with rapid drop-offs.

leg and appeared to reorient itself under the victim's surfboard. After initially swimming away from the victim, the shark returned and circled until a second surfer, approximately 30 feet away began to shout and swim toward the victim. At this point, the victim was able to catch a wave into shore. Within three minutes, all surfers had exited the water and were rendering aid to the victim. Based on the victim's visual assessment, the shark was bigger than him at 2.5 m long with a blunt round snout that was about 14 inches wide and dark gray in color. It was a fast and aggressive shark when it attacked and slow-moving shark when it was swimming away from him and circling the area. During the medical care, a piece of tooth came out of a wound on the outside of the left knee. This would have come from the lower jaw, because of the way he was positioned on his board.

The wounds on his right thigh would have come from the upper jaw (Fig. 2).

IDD Calculation

For many species of sharks, the inter-dental distance (IDD) varies directly with the total length (TL) of an individual through a log-linear relationship and can therefore give a reliable estimate of the TL (9). The IDD is measured between the tip of a functional tooth and the tip of the functional tooth of the adjacent file. On a wound, the center of each tooth imprint is used as a proxy for the tip of the tooth and gives a reliable estimate of the IDD (9). A relationship also exists between the bite circumference (BC) and the length of a shark. The BC is the

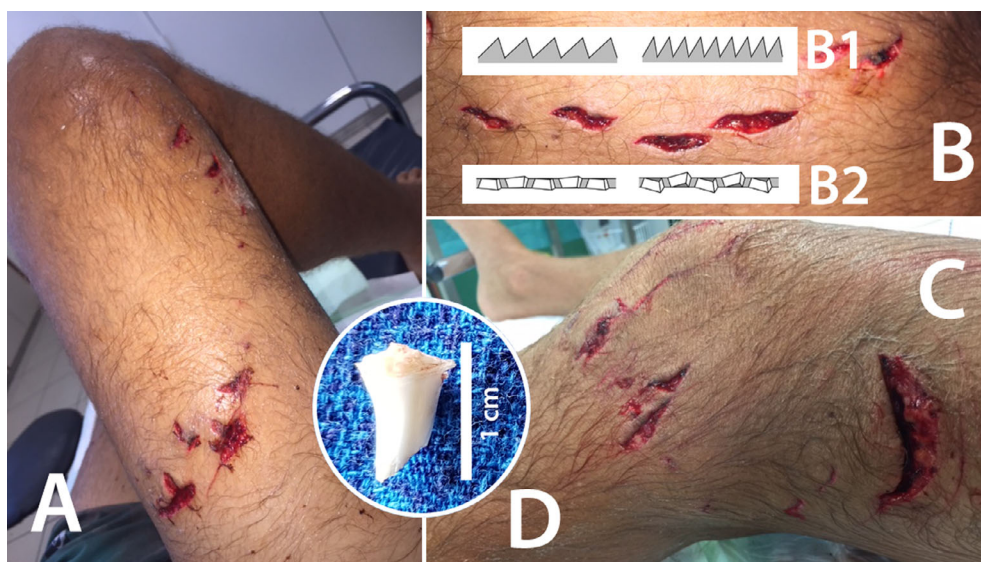


FIG. 2—General features of the bite wounds inflicted on the surfer, showing (A) the circular imprints of the upper jaw of the shark on the right thigh, (B) imprints of teeth from the upper jaw above the left knee, and (C) imprints of teeth from the lower jaw on the left side of the knee and the lower left part of the left thigh. (D) close-up of a piece of tooth that was found in the wound close to the knee displayed in C (Photo courtesy Dominick Dibartolomeo). B1 and B2 represent general scheme of the set (left to right: slight to strong) and the pitch (left to right: coarse to fine) of the teeth (adapted from [12]).

distance measured between the two most posterior teeth of the functional row of each extreme side of a jaw, following the base of the consecutive tooth. As the imprints of the last teeth are not always present on a wound, the assessment of the BC provides a minimal estimate of a shark TL (9). Both IDD (and their average) and BC were assessed by the first author based on the high-quality photographs taken prior to surgery and a metric marker (to the millimeter) provided by the victim, a few days after the incident.

DNA Analysis

The cytochrome oxidase subunit I (COI) is a standardized mitochondrial DNA region that has demonstrated accurate and reliable species identification of fishes with high resolution (10,11). The tooth fragment was preserved in 70% ethanol. The fragment was placed in 360 μ L of Qiagen ATL buffer, and DNA was extracted using Qiagen DNeasy Tissue kit following the manufacturer's instructions. A DNA extraction was also performed on two shark's skin samples conserved in 70% ethanol, reef blacktip shark *Carcharhinus melanopterus* and gray reef shark *Carcharhinus amblyrhynchos* (positive controls). A negative control extraction was performed without tissue. A total of 100 μ L of genomic DNA was obtained for each sample, and 2 μ L were used for amplification of the cytochrome c oxidase subunit I (COI) barcode region.

A fragment of the mitochondrial gene *COI* was amplified using a set of fish universal primers (FishF1 5'-TCAACCAAC-CACAAAGACATTGGCAC-3' and FishR1 5'-TAGACTTCTGGGTGGCCAAAGAATCA-3') (6). Polymerase chain reaction (PCR) was conducted in a 25 μ L final reaction volume containing 1.5 mM $MgCl_2$, 400 μ M of each nucleotide (Promega dNTP mix), 0.5 μ M of each primer and 2 U of GoTaqG2 DNA Polymerase (Promega), and 2 μ L of previously extracted DNA. The amplification was performed using an Eppendorf Master cycler 96-Well Thermal Cycler with the following cycle conditions: denaturation at 95°C for 4 min; followed by 35 cycles at 95°C for 30 sec, 52°C for 30 sec, and 72°C for 45 sec; and a final elongation step at 72°C for 10 min. PCR amplification success and specificity were checked on agarose 1% gel electrophoresis.

The negative control showed no amplification. PCR products that were successfully amplified were sequenced using reverse primer FishR1 (Macrogen sequencing service, The Netherlands). The sequences were viewed and manually trimmed using CodonCode Aligner. A 640 bp sequence was used for nucleotide blast (<https://blast.ncbi.nlm.nih.gov>).

Results

Assessment of the Species and Size of the Shark

The general appearance of the teeth imprints with a coarse pitch (based on the lack of alignment of the imprints) and a strong set (large tooth spacing) (12) (see Fig. 2A,B) does not fit with a jaw holding more aligned and linguo-labially flattened teeth such as those found for most *Carcharhinidae*, including the bull shark and the gray reef shark *C. amblyrhynchos* and the blacktip shark *Carcharhinus limbatus*, or those of a tiger shark that would leave more continuous and straight cuts (13). Furthermore, the piece of tooth (see Fig. 2D) showed that the shark's teeth were rather elongated and round, and allowed (through a computerized drawing) the virtual fitting with a

loose tooth from the lower jaw of a sicklefin lemon shark *N. acutidens* (Fig. 3A).

The measurements of the several IDD between teeth imprints from the partial bite on the left knee gave an average IDD of 18 mm, and the BC measured on the partial bite on the right thigh was 36 cm (Fig. 3B,C). Based on the algorithms mentioned in the methodology for IDD (Figure S1) and BC (Figure S2), respectively (9), the TL of the shark would range between approx. 265 and 250 cm.

Confirmation of the Shark Species Through DNA

The sequence from the sampled shark tooth was assigned to *N. acutidens* with 100% identity over 630 bp (*N. acutidens* voucher BIOUG < CAN >: BW-A3012 cytochrome oxidase subunit 1 COI gene, partial cds; mitochondrial accession number gil166837539|EU398939.1). The two positive controls were successfully assigned to their respective species, the reef blacktip shark *C. melanopterus* and the gray reef shark *C. amblyrhynchos*, with 99% and 98% identity (*C. melanopterus* voucher SOSSRC: *C. melanopterus* OC-69 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial accession number FJ519121.1 and *C. amblyrhynchos* isolate DFD 165 cytochrome c oxidase subunit 1 (COI) gene, partial cds; mitochondrial accession number KX063633.1).

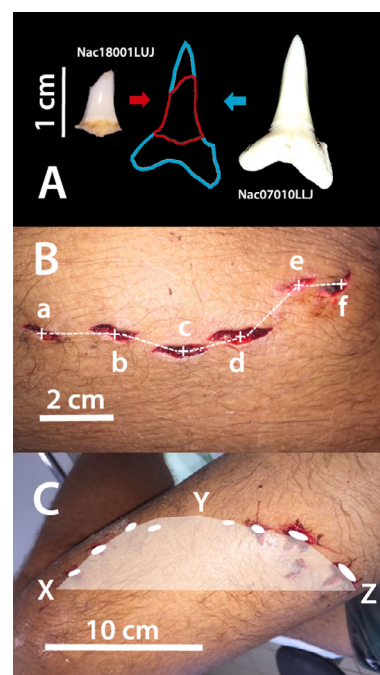


FIG. 3—(A) Virtual reconstruction (in red and blue) of the shape of a tooth (ID Nac07010LLJ) from the lower jaw of a sicklefin lemon shark, showing the fitting of the tooth piece (ID Nac18001LUJ) found in the surfer's left knee wound (references of the teeth stored at the CRIOBE collection are indicated). (B) Close-up of five imprints made by the teeth from the upper jaw of the shark on the top of the left knee, that allowed an assessment of the shark size through the calculation of an average IDD of 18.43 mm to be made (a-b = 20.31 mm; b-c = 17.18 mm; c-d = 15.62 mm; d-e = 26.56 mm; e-f = 12.5 mm). (C) Top of the surfer's right thigh showing the imprints of the teeth from the upper jaw of the shark that allowed an assessment of the shark length through the measurement of the BC (XYZ = 36 cm et XZ = 19 cm) (Photo courtesy Frederick Dibartolomeo).

The cytochrome C oxidase subunit I (COI) gene fragment barcode was successfully amplified and identified using BLAST public databases. Positive controls were also successfully assigned to their respective species. The tooth fragment comes from a sicklefin lemon shark.

Discussion

Most case studies documenting bites on humans by sharks are not reported with reliably detailed accounts of the occurrence, its location, or genetic samples left by the shark. For this study, the quality of bite wound photographs taken prior to surgery and a tooth fragment suitable for genetic analysis left in the victim indicates that the species involved in this occurrence was a sicklefin lemon shark. Such identification was confirmed with certainty through the genetic analysis of the tooth fragment. The scarcity of confirmed strikes of this species along with its sister species from the Atlantic ocean *Negaprion brevirostris* on people, with only 13 bites on humans over the past 10 years though none of them fatal nor analyzed from a forensic point of view (14), makes this case study particularly interesting.

The motivations for shark bites may include foraging (6,15) but many other motivations exist such as exploration, defense, territoriality, competition (15–18), or confusion (19). From a forensic point of view, foraging aggressions would involve a loss of ingested tissues whereas other motivations would result in more superficial wounds (7). In this case, the wounds were superficial and thus the bite was unlikely motivated by foraging. The presence of two bites suggests that motivation was neither exploratory in nature, as such a behavior usually relies on a single bite (20). In addition, the sicklefin lemon shark is a piscivore and, as such, it would be highly unusual that this species would have intentionally fed on a human prey. Defense (by fear or following a human aggression, [15]), competition (for a food source, [16,17]), and confusion (19) may also lead to superficial wounds but rather involve spearfishes and scuba divers in underwater interactions with sharks carrying out provoked bites that hardly concern the surfing activity.

The sicklefin lemon shark is very common in French Polynesia and is one of the most common shark species involved in underwater shark-based ecotourism activities, mainly those located in the Society islands such as Tahiti, Moorea or Bora-Bora (19). This species is present in the Tuamotu archipelago, including Makemo atoll as indicated by local fishermen. One of the surfers involved in the incident reported that he routinely spearfishes in this area and often sees adult lemon sharks. The fact that the victim did not identify this species based on its typical yellowish color is not surprising, as for older animals, darker skin will often replace the yellow that is more visible in younger individuals (EC, Pers. Obsv.).

With total lengths that can reach more than 330 cm, implying large and powerful jaws holding from 80 to 100 functional tearing-type teeth (12) well-suited for fish consumption, displaying hunting strategies that include quick and powerful ramming, the sicklefin lemon shark constitutes a potential threat to humans (21). This species responds promptly and aggressively when touched, poked, or speared and will attack people or boats when accosted. Unprovoked attacks by this shark were reported on divers, possibly after the divers approached too close or otherwise alarmed the shark (21). In French Polynesia, the sicklefin lemon shark was involved in 7% ($n = 4$) of the bites on humans between 1997 and 2005, far behind the gray reef shark ($n = 21$) (22). Over the past century, the sicklefin lemon shark was

responsible for one of the two human fatalities recorded in French Polynesia. A fisherman was bitten in the late 1970s in the atoll of North Tepoto, after accidentally spearing a sicklefin lemon shark that he confused with a tawny nurse shark *Nebrius ferrugineus* (EC, Unpublished data). Since the late 2000s, most of the bites by lemon sharks were perpetrated in the context of diving, in particular within the context of artificial food provisioning for ecotourism (23). Bites happen mainly in the presence of risky practices such as hand-feeding when sharks confuse a diver's appendage with bait (24) and also other poor diver practices (although not well documented). For these cases, including the fatality, the bites were most likely accidental in nature, rather than a deliberate attempt by the shark to feed on human prey (7,8). These accidental bites that do occur, whether related to spearfishing or shark feeding, either motivated by confusion, defensiveness, or competitiveness, are facilitated by the high territoriality and natural aggression of the sicklefin lemon shark (21,23). Interestingly, the degree of aggression and territoriality is not evenly distributed across the species as individuals from the Tuamotu archipelago (where both this incident and the fatality occurred), as well as in places such as New Caledonia (Western Pacific) (EC, Pers. Obs.), show much more agonistic behaviors toward humans (1) than in the Society islands where most of the individuals demonstrate sluggish and nonaggressive behaviors in the context of uncontrolled diving ecotourism (19). Therefore, the location of the incident in the Makemo atoll, among the Tuamotu archipelago, further supports a territorial motivation for the nonfatal bites. Another piece of circumstantial evidence comes from statements made by the other three surfers who had visited the atoll over the past 12 years, who when questioned, stated that they had never paddled out as far as the victim did that morning. As such, the victim likely entered into a new area and committed an inadvertent provocation as a trespass of a shark's territorial limits in terms of space (15). The instantaneous trigger of the strike might be linked to the violation of the lemon shark's personal ideosphere—the volume of water immediately surrounding an individual animal (of any species) that it will defend if broached without appropriate signaling or permission (25).

Regarding other potential motivations, the hypothesis of the “mistaken identity” (18) suggesting that the shark would confuse—in this case—the surfer with the shape of a turtle (as seals do not exist in French Polynesia) is highly unlikely for two reasons. First, the position of the surfer (sitting on his board) just before the strike does not come close to approximating the shape of a turtle, and this is particularly true in an abiotic environment with high water clarity. Second, none of the extensive, heavily researched field guides mention any member of the genus *Negaprion* feeding on sea turtles of any size or locality around the world (21,26). The territoriality (15,21) remains then the most probable hypothesis for explaining these nonfatal bites on a surfer.

Conclusion

Besides the sicklefin lemon shark, other species such as the gray reef shark also show a strong territoriality (17) that can trigger agonistic behaviors toward sea users, regardless of their activity. In areas where humans regularly practice watersports, we recommend that scuba divers with expertise in shark identification and behavior conduct basic underwater assessments to assess the presence of potential territorial problem individuals that could pose problems for humans. With baseline data

collected, adequate management measures through a spatial approach could then be implemented in order to decrease the risk of accidental shark bites.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Figure S1. Algorithms representing the relationship between the Inter-dental Distance (IDD) of the upper (A) and lower (B) jaws, and the Total Length (TL) that can be used for *Negaprion* spp. (based [10]).

Figure S2. Algorithms representing the relationship between the Bite Circumference (BC) of the lower (C) and upper (D) jaws respectively, and the Total Length (TL) that can be used for *Negaprion* spp. (based [10]).