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Comparison of dorsal and pectoral fin denticles for grey nurse, great white, and six whaler sharks from east Australian waters

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Abstract

Escalating demand for shark fins poses a major threat to shark populations world-wide and the monitoring of shark catches can be very difficult when only the fins are present. Identifying a species of shark using only its fins can be enhanced by using the dermal denticles as they maintain their structural integrity irrespective of freezing or drying. In this study, scanning electron microscopy was used to examine various denticle characteristics including: the number and persistence of ridges and cusps, posterior edge appearance, dispersion and size (length and width) at three positions (anterior margin, centre and posterior margin) on the dorsal fin and dorsal surface of the pectoral fin of sharks caught in the coastal waters off New South Wales, Australia. Samples were obtained from commercial and recreational fishers, and the shark meshing programme. Catches included two threatened species – *Carcharias taurus* and *Carcharodon carcharias*, and six carcharhinid whalers – *Carcharhinus brachyurus*, *C. falciformis, C. leucas, C. limbatus, C. longimanus* and *C. obscurus*. Detailed examination revealed similarities and differences among sampling positions within a fin, between fin types, and among species. Patterns of change in denticle characteristics and size across the sampling positions provided an efficacious means of unequivocally identifying eight shark species. Future studies should evaluate field-based sampling of denticle characteristics as they may provide a cost-effective alternative to genetic techniques for identifying sharks and enable much needed data on the quantity and species composition of sharks harvested for their fins in local waters.

Keywords: elasmobranch, dermal denticle, Carcharhinidae, threatened species, taxonomy.

Introduction

The protein mining (*sensu* Field et al. 2009a) of the world's oceans via recognised fisheries and illegal, unreported and unregulated fishing (FAO 2007, Pitcher et al. 2002) has lead to declining fish populations and various ecosystem changes (Stevens et al. 2000, Pauly et al. 2005, Worm et al. 2006). There is irrefutable evidence of world-wide, declining shark populations (Baum et al. 2003, Myers & Worm 2003, Baum & Myers 2004, Field et al. 2009b).

These declines are exacerbated by their relatively slow growth, late onset of sexual maturity, low fecundity and extended longevity which makes them extremely susceptible to over-fishing and many species require decades to recover (Smith et al. 1998, Cortés 2000, Mollet & Cailliet 2002). The primary causes of these declines are targeted shark fisheries (e.g. Walker 1998, Santana et al. 2009), by-catch in other fisheries (e.g. Marin et al. 1998, Campana et al. 2009) and the burgeoning shark-fin trade fuelled by booming Asian economies and increased personal wealth (Rose 1996, Clarke et al. 2007).

For many centuries shark fins have been a traditional component in Chinese banquets (Rose 1996) and the escalating demand has resulted in illegal harvesting (Shivji et al. 2005, Dulvy et al. 2008). Recent analyses of commercial fin-trade records (Clarke 2004, Clarke et al. 2006) have indicated that the shark catch documented in FAO databases has been substantially under-reported. It is likely that the wasteful shark-fining practices (comprising capture, fin removal and disposal at sea) have greatly contributed to the under-reporting of global shark catches. To redress this, some jurisdictions including Australia and parts of Central America now require that all shark carcasses are brought ashore prior to finning as this enhances species identification, the monitoring of catch, regulatory compliance and reduces waste (Dulvy et al. 2008).

The absence of the numerous morphological characteristics used with whole carcasses (e.g. Garrick 1982, Compagno 2002, Last & Stevens 2009) makes the identification of a shark from its fins alone substantially more difficult. While fin colour and shape have been used in studies of the Japanese longline fishery (Matsunaga et al. 1998, Nakano & Kitamura 1998), colouration can vary with fin size, and can be altered by post-mortem freezing and/or drying (Salini et al. 2007). The shape of fins can be similarly affected or be damaged with a subsequent loss of distinguishing characteristics. In contrast, a shark's dermal denticles maintain their structural integrity irrespective of freezing or drying and enable the species identification (Applegate 1967, Nakano & Kitamura 1998, Salini et al. 2007). For example, Tanaka et al. (2002) described the denticle characteristics of thirteen pelagic sharks from Japanese waters using scanning electron microscopy (SEM) and showed that specific characteristics could be used for species identification. Variation in denticle characteristics at different locations on a shark (e.g. fins versus the torso) has also been documented (e.g. Bagar & Thorson 1995, Salini et al. 2007) and can pose problems for species identification. Nevertheless, these are easily mitigated via rigorous experimental protocols and detailed descriptions of sampling location: information that has been notably absent in some previous studies (e.g. Matsunaga et al. 1998). In contrast, more recent studies (Shivji et al. 2002, Abercrombie et al. 2005, Clarke et al. 2007) have focussed on the development of complex genetic techniques for identifying a range of shark species from their fins and will be extremely valuable for legal proceedings. In processing the fins for human consumption, the skin is removed and the fins are soaked in bleach (Rose & McLoughlin 2001), making the use of denticles for identification impossible, and the extraction of DNA more difficult. In spite of this, the relative simplicity associated with using denticle characteristics, especially with the development of field-based techniques (e.g. digital macro-photography), will likely provide a very cost-effective, efficacious means of identifying sharks from their fins per se.

The shelf waters off New South Wales (NSW) Australia (Latitudes 28 – 37° S) support a diverse, predominantly temperate fish community that has formed the basis of numerous fisheries targeting invertebrates, bony fish and elasmobranchs. Sharks, in particular, have been commercially targeted and caught as by-catch in the Ocean Trap and Line, Ocean Trawl, Estuary General and Ocean Hauling fisheries (Pollard et al. 1996, Scandol et al. 2008, Macbeth et al. 2009) operating along the entire NSW coast. In contrast, recreational anglers have generally caught sharks when targeting bony fish (Henry & Lyle 2003), although some deliberate targeting has occurred, especially during gamefishing competitions (Stevens 1984, Pepperell 1992). Sharks have also been targeted by the shark meshing programme (SMP) operating off the bathing beaches from Newcastle to Wollongong (Reid & Krogh 1992, Krogh 1994). The commercial and recreational fishing sectors and the SMP have also inadvertently caught two threatened shark species: the "critically endangered" grey nurse shark, *Carcharias taurus* Rafinesque, 1810 and the "vulnerable" great white shark, *Carcharodon carcharias* Linnaeus, 1758 (Krogh & Reid 1996, Otway et al. 2004, Bruce et al. 2006).

Under NSW legislation (Fisheries Management Act, 1994) it is illegal to fin a shark and then discard the carcass whilst at sea, but this practice continues as fisheries compliance officers still confiscate illegally harvested shark-fins. In the absence of a field-based technique for identifying NSW sharks from their fins per se, offences relating to the two threatened sharks may be overlooked. Additionally, the catches of several other sharks included on the IUCN Red List (Cavanagh et al. 2003), but not currently recognised under Australian legislation are unlikely to be quantified. Hence, the twofold objectives of this study were to document the various characteristics of denticles sampled from first dorsal and pectoral fins of sharks and then examine whether the denticle characteristics could be used to identify and discriminate among the range of shark species caught.

Methods

Field Sampling

Sharks were sampled from the SMP and catch of commercial and recreational fishers in NSW waters from January 1995 to March 2008. Sharks were identified using standard taxonomic methods (Garrick 1982, Compagno 2002, Last & Stevens 2009) and the total length (TL) was measured to the nearest centimetre with the caudal fin in the depressed position (Francis 2006). The first dorsal and pectoral fins were then removed by cutting the torso anteriorly from under the free rear tip and inner margin, passing below the cartilaginous elements to a point 5 cm anterior to the origin of the fin. The entire, undamaged fins were then placed in labelled plastic bags and stored in a chest freezer at -200 C until processed.

Skin Sampling and Scanning Electron Microscopy (SEM)

Preliminary sampling of dorsal fins showed that there were no differences in denticle morphology on either side of the fin, thus the side of the fin sampled was chosen at random. In contrast, all pectoral fins were sampled on the dorsal surface. Skin samples (≈ 50 x 20 mm) were removed, from three sampling positions (anterior margin, centre and posterior margin – Fig. 1a) from a thawed fin using a scalpel and forceps.

Each skin sample was subjected to a standardised cleaning and desiccation process prior to detailed examination using SEM. Briefly, skin samples were placed in a 95% ethanol bath in a Branson ultrasonic cleaner which was run for three minutes. The ethanol was then replaced and the process repeated. Finally, each sample was rinsed with ethanol, fastened (using clips) to a rigid, plastic board to prevent buckling and placed in a desiccator to dry. After 5 days, all samples were dry and each skin sample was cut into three replicate sub-samples (≈ 15 x 15 mm and placed on 25 mm diameter aluminium stubs using carbon paint to ensure a conductive track. A 10-20 nm gold coat was then applied using a SPI sputter coating unit. Each sample was then examined on a Philips XL Series – XL30 scanning electron microscope fitted with a tungsten electron gun and images obtained at 15kv with 200x and 400x magnification. All SEM images were orientated so that the anterior-posterior axes of the denticles aligned vertically with the anterior edge at the top.

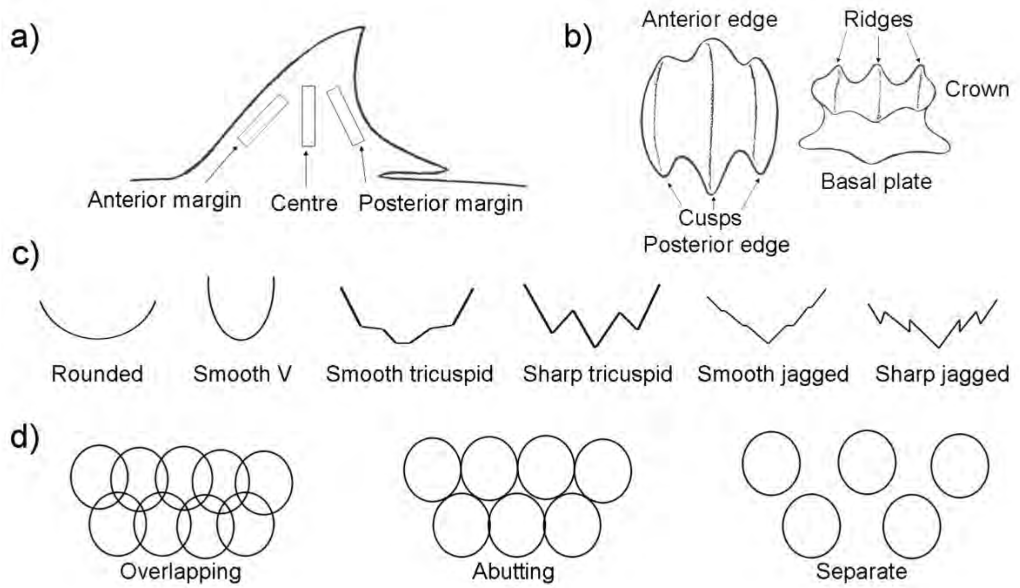

Figure 1. Sampling protocol and denticle characteristics used to describe (a) the 3 sampling locations from the first dorsal and pectoral fin anterior margin, centre and posterior margin (b) Characteristics of a typical denticle as viewed from above (left) and from the side (right) (c) Terminology used to describe the shape of the posterior edge of denticles (d) The definitions of denticle distribution types.

Denticle Descriptions

Descriptions of the denticles from the three sampling positions on each of the first dorsal and pectoral fins for each shark species were compiled using common features and consistent terminology. The crown of the denticle may be smooth, have depressions, or more commonly have a surface with one or more ridges aligned in an anterior – posterior direction (Fig. 1b). The number of ridges/denticle was recorded and the ridges described as they may run the entire length or dissipate towards the posterior edge of the denticle. Commonly, a denticle's anterior edge is rounded while the posterior edge is indented. Thus, the shape of a denticle's posterior edge was described, its appearance (distinct or indistinct) noted and number of cusps recorded (Fig. 1c). Finally, the dispersion of the denticles was classified into one of three types (Fig. 1d). Overlapping dispersion comprised very dense, regularly-spaced denticles that overlapped along the anterior, posterior and lateral edges. Abutting dispersion comprised dense, regularly-spaced denticles with contact, but no overlap along the edges. Separated dispersion comprised regularly-spaced denticles with no overlapping edges and an obvious space between surrounding denticles.

Statistical Analyses

Length and width of denticles in SEM images were measured to the nearest 0.5 µm using Grab It! XP Software. Replicate measurements were obtained from each of four, randomly-chosen denticles at the three sampling positions on the first dorsal and pectoral fins. To examine whether multivariate statistical analyses could differentiate species using either the first dorsal or pectoral fins, PRIMER version 6.1.9 was used to produce two-dimensional MDS ordination plots following square root transformation of replicate (length & width) measurements and the calculation of Euclidean distance. A one-way analysis of similarity (ANOSIM) for each fin was used to test for significant differences among species.

Results

Numerous sharks comprising nineteen species from seven families were obtained from the combined catches of the SMP, commercial and recreational fisheries along the entire NSW coast. Greater numbers of individuals and species were obtained between Coffs Harbour and Wollongong, and this was associated with increased sampling effort in this region (Fig. 2).

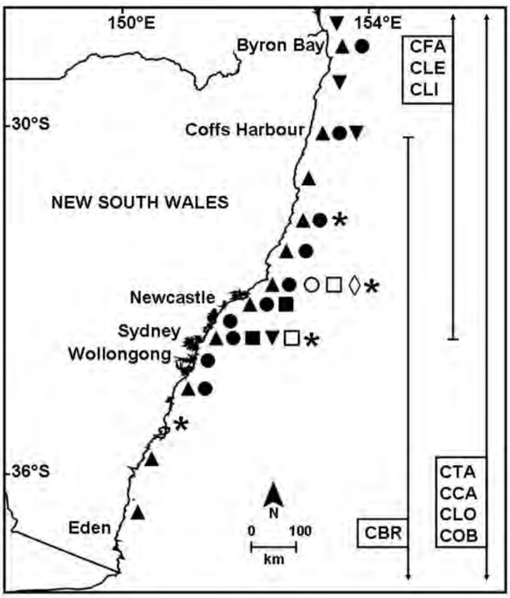

Figure 2. Map showing with geographic range (line) and capture locations (symbol) for Carcharias taurus (CTA, ▲), Carcharodon carcharias (CCA, ●), Carcharhinus brachyurus (CBR, ■), C. falciformis (CFA, ○), C. leucas (CLE, ▼), C. limbatus (CLI, □), C. longimanus (CLO, ◊), and C. obscurus (COB, *) *along the coast of New South Wales, Australia.*

While the combined catch included eleven species of carcharhinid whaler, only six species (*Carcharhinus brachyurus, C. falciformis, C. leucas, C. limbatus, C. longimanus* and *C. obscurus*, pooled n = 30) together with *Carcharias taurus* (n = 31) and

Carcharodon carcharias (n = 17) are examined in this study. Descriptions of the denticles in the remaining species will be provided in subsequent papers.

Furthermore, a sexually-mature, 272 cm TL, female *C. taurus* that had survived finning (i.e., dorsal, pectoral & lower caudal fins absent) was also examined following its accidental capture in June 2002 on a demersal setline off the NSW south coast.

Comparisons Between Fins and Among Fin Positions

Denticle samples used in this study were obtained from individuals 111 to 450 cm TL. Detailed examination of the eight species clearly showed that the various denticle characteristics did not differ between the sexes. The SEM images of the denticles (Figs. 3–5) and their various characteristics (Tables 1–3) highlight the similarities and differences among sampling positions within a fin, between fin types, and among species.

The denticles from the first dorsal and pectoral fins of *Carcharias taurus* (Fig. 3) were shield-shaped and very similar. Denticle size (length & width) and dispersion differed among the three sampling positions (anterior margin, central and posterior margin) with smaller, separated denticles at the posterior margin of the dorsal fin (Tables 1 & 3). In contrast, the pectoral fin denticles were larger and abutting at the anterior margin compared to the remaining positions on the fin (Tables 1 & 3).

The denticles from the first dorsal and pectoral fins of *Carcharodon carcharias* were ellipsoidal in shape and had similar features (Fig. 3). Denticles on the dorsal fin differed among sampling positions with those at the anterior margin exhibiting no cusps, a smooth posterior edge and larger size (Tables 1 & 3). A similar pattern of difference was evident on the pectoral fin denticles which had no cusps, a rounded posterior edge and larger size at the anterior margin (Tables 1 & 3).

The denticles from the first dorsal and pectoral fins of *Carcharhinus brachyurus* (Figs. 4 & 5) were diamond-shaped, similar in some features and differed among sampling positions (Tables 2 & 3). Denticles at the posterior margin of the dorsal fin had continuous ridges, five cusps and a smooth, jagged posterior edge. In contrast, denticles at the anterior margin of the pectoral fin exhibited the greatest difference with dissipating ridges, no cusps, and a smooth V posterior edge.

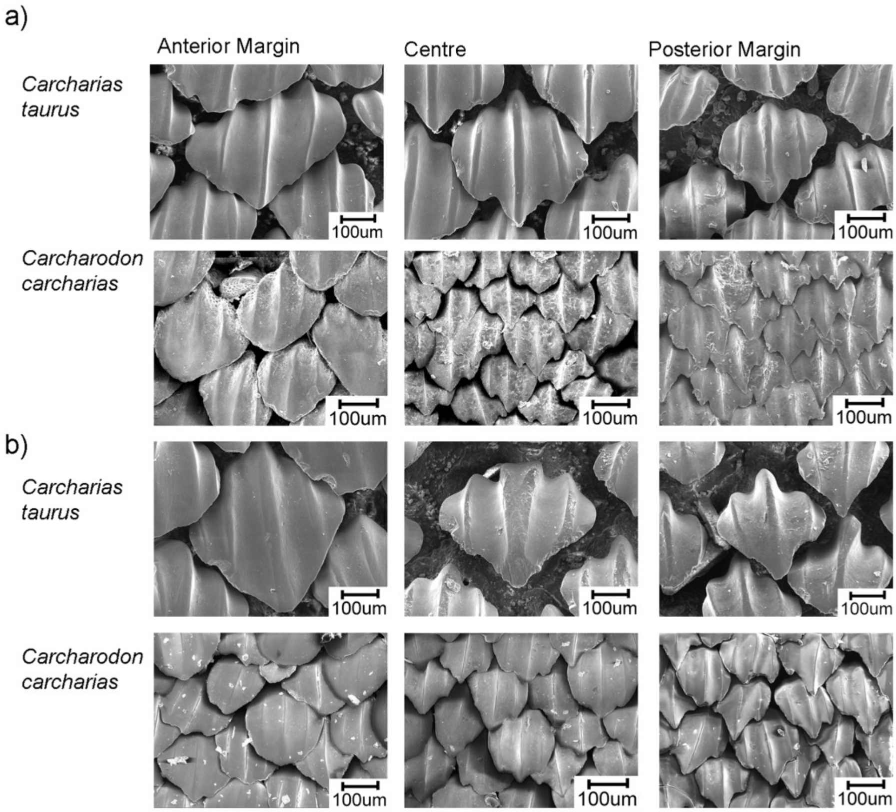

Figure 3. Scanning electron micrographs of denticles from the anterior margin, centre and posterior margin of (a) the first dorsal fin and (b) the dorsal surface of the pectoral fin of Carcharias taurus *(251 cm TL) and* Carcharodon carcharias *(209 cm TL).*

Denticles from the first dorsal and pectoral fins of *C. falciformis* (Figs. 4 & 5) were diamond-shaped with differences on the dorsal fin arising via a smooth V posterior edge and abutting denticles at the anterior margin (Table 2). Differences on the pectoral fin were evident through the varying shapes of the posterior edge across the three sampling positions and overlapping denticles at the posterior margin (Table 2).

Denticles from the first dorsal and pectoral fins of *C. leucas* (Figs. 4 & 5) were diamond-shaped, large and showed various similarities (Tables 2 & 3). Denticles on the dorsal fins differed among sampling positions with those at the anterior margin exhibiting dissipating ridges, no cusps and a smooth V posterior edge. Smaller denticles were also present at the posterior margin. A similar pattern of difference was evident on the pectoral fins except that abutting denticles occurred at the anterior margin and their size did not differ among sampling positions.

Denticles from the first dorsal and pectoral fins of *C. limbatus* (Figs. 4 & 5) were diamond-shaped and very similar overall (Tables 2 & 3). Smaller denticles at anterior margin were the only difference apparent on the first dorsal fin. Differences on the pectoral fin were manifest via larger denticles at the anterior margin together with distinct cusps and a sharp jagged posterior edge at the posterior margin.

Denticles from the first dorsal and pectoral fins of *C. longimanus* (Figs. 4 & 5) were diamond-shaped and exhibited many similarities. Differences on the dorsal fin were confined to the anterior margin where denticles were abutting and had dissipating ridges (Table 2). In contrast, differences on the pectoral fin occurred at the posterior margin with denticles possessing distinct cusps and a sharp, jagged posterior edge (Table 2).

Denticles from the first dorsal and pectoral fins of *C. obscurus* (Figs. 4 & 5) were diamond-shaped with some variation among fin positions (Tables 2 & 3). The denticles at the anterior margin of the dorsal fin had no ridges and cusps, and a smooth V posterior edge and differed from those at the other sampling positions. Similarly, the denticles from the anterior margin of the pectoral fin were distinctive because of dissipating ridges, no cusps and a smooth V posterior edge.

Species	**Position**	**Ridges** (No., Persistence)	**Cusps** (No., Form)	**Posterior edge**	**Dispersion**
First Dorsal Fin					
Carcharias	AM	3, Continuous	3, Indistinct	Smooth tricuspid	Abutting
taurus	C	3, Continuous	3, Indistinct	Smooth tricuspid	Abutting
	PM	3, Continuous	3, Indistinct	Smooth tricuspid	Separated
Carcharodon	AM	3, Continuous	0, Absent	Smooth V	Overlapping
carcharias	C	3, Continuous	3, Distinct	Sharp tricuspid	Overlapping
	PM	3, Continuous	3, Distinct	Sharp tricuspid	Overlapping
Pectoral Fin					
Carcharias	AM	3, Continuous	3, Indistinct	Smooth tricuspid	Abutting
taurus	C	3, Continuous	3, Indistinct	Smooth tricuspid	Separated
	PM	3, Continuous	3, Indistinct	Smooth tricuspid	Separated
Carcharodon	AM	3, Continuous	0, Absent	Rounded	Overlapping
carcharias	C	3, Continuous	3, Distinct	Sharp tricuspid	Overlapping
	PM	3, Continuous	3, Distinct	Sharp tricuspid	Overlapping

Table 1. Denticle characteristics for the anterior margin (AM), centre (C) and posterior margin (PM) of the first dorsal fin and dorsal surface of the pectoral fin of Carcharias taurus (251 cm TL) and Carcharodon carcharias (209 cm TL).

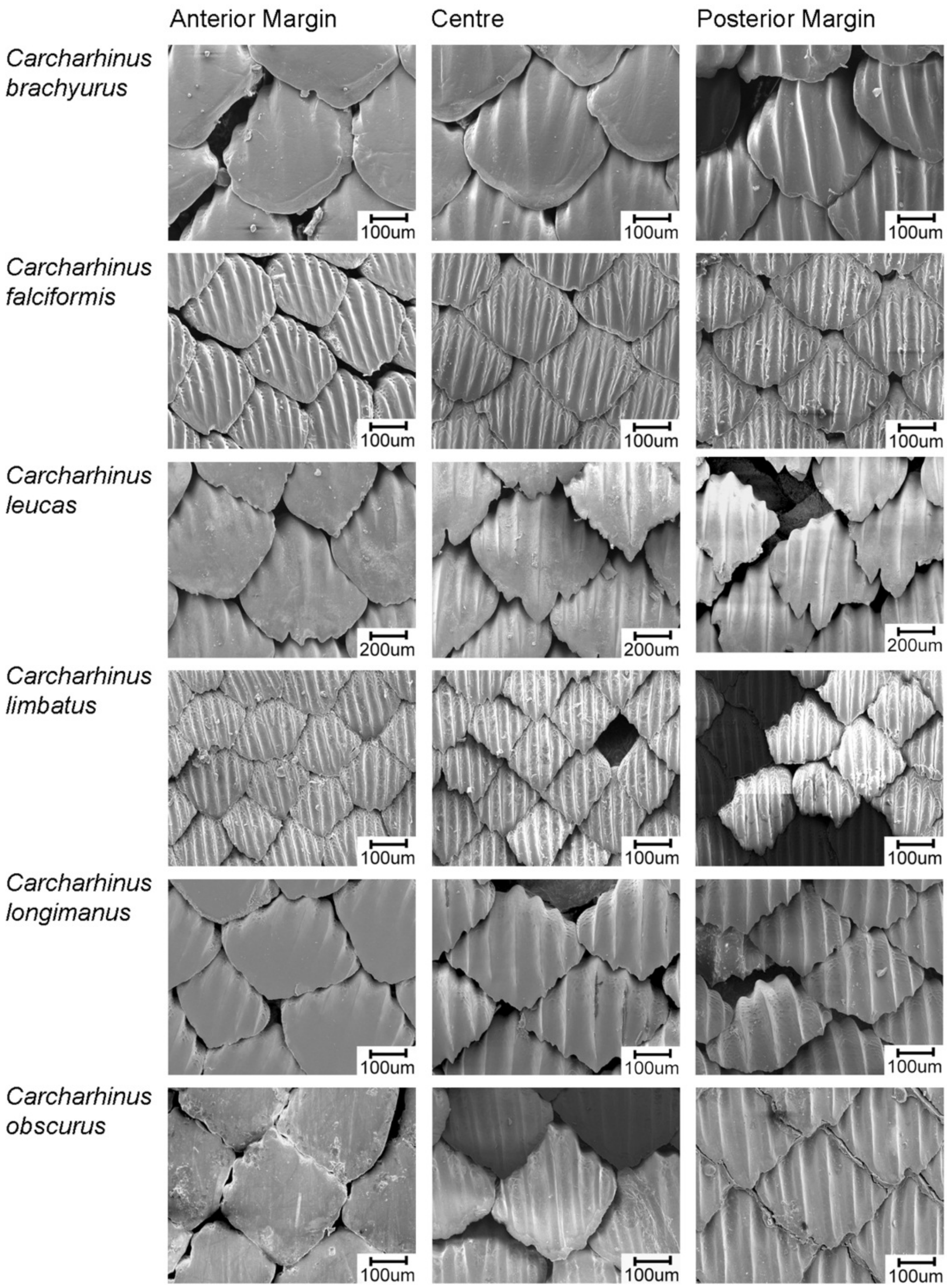

Figure 4. Scanning electron micrographs of denticles from the anterior margin, centre and posterior margin of the first dorsal fin from Carcharhinus brachyurus *(271 cm TL),* C. falciformis *(264 cm TL),* C. leucas *(239 cm TL),* C. limbatus *(265 cm TL),* C. longimanus *(178 cm TL) and* C. obscurus *(265 cm TL).*

Species	**Position**	**Ridges** (No., Persistence)	**Cusps** (No., Form)	**Posterior edge**	**Dispersion**
First Dorsal Fin					
Carcharhinus	AM	5, Dissipate	0, Absent	Rounded	Overlapping
brachyurus	C	5, Dissipate	0, Absent	Rounded	Overlapping
	PM	5, Continuous	5, Indistinct	Smooth jagged	Overlapping
Carcharhinus	AM	5-7, Continuous	0, Absent	Smooth V	Abutting
falciformis	C	5-7, Continuous	0, Absent	Rounded	Overlapping
	PM	5-7, Continuous	0, Absent	Rounded	Overlapping
Carcharhinus	AM	5-7, Dissipate	0, Absent	Smooth V	Overlapping
leucas	C	5-7, Continuous	5, Distinct	Sharp jagged	Overlapping
	PM	5-7, Continuous	5, Distinct	Sharp jagged	Overlapping
Carcharhinus	AM	5, Continuous	5, Indistinct	Smooth jagged	Overlapping
limbatus	C	5, Continuous	5, Indistinct	Smooth jagged	Overlapping
	PM	5, Continuous	5, Indistinct	Smooth jagged	Overlapping
Carcharhinus	AM	5, Dissipate	5, Indistinct	Smooth jagged	Abutting
longimanus	C	5, Continuous	5, Indistinct	Smooth jagged	Overlapping
	PM	5, Continuous	5, Indistinct	Smooth jagged	Overlapping
Carcharhinus	AM	0, Absent	0, Absent	Smooth V	Abutting
obscurus	C	5, Continuous	5, Indistinct	Smooth jagged	Abutting
	PM	5, Continuous	5, Indistinct	Smooth jagged	Abutting
Pectoral Fin					
Carcharhinus	AM	5, Dissipate	0, Absent	Smooth V	Abutting
brachyurus	C	5, Continuous	5, Indistinct	Smooth jagged	Abutting
	PM	5, Continuous	5, Indistinct	Smooth jagged	Overlapping
Carcharhinus	AM	5-7, Continuous	0, Absent	Rounded	Abutting
falciformis	C	5-7, Continuous	0, Absent	Smooth jagged	Abutting
	PM	5-7, Continuous	0, Absent	Smooth V	Overlapping
Carcharhinus	AM	5-7, Dissipate	0, Absent	Smooth V	Abutting
leucas	C	5-7, Continuous	5, Distinct	Sharp jagged	Overlapping
	PM	5-7, Continuous	5, Distinct	Sharp jagged	Overlapping
Carcharhinus	AM	5-6, Continuous	5-6, Indistinct	Smooth jagged	Overlapping
limbatus	C	5-6, Continuous	5-6, Indistinct	Smooth jagged	Overlapping
	PM	5-6, Continuous	5-6, Distinct	Sharp jagged	Overlapping
Carcharhinus	AM	5, Continuous	5, Indistinct	Smooth jagged	Abutting
longimanus	C	5, Continuous	5, Indistinct	Smooth jagged	Abutting
	PM	5, Continuous	5, Distinct	Sharp jagged	Abutting
Carcharhinus	AM	5, Dissipate	0, Absent	Smooth V	Abutting
obscurus	C	5, Continuous	5, Indistinct	Smooth jagged	Abutting
	PM	5, Continuous	5, Indistinct	Smooth jagged	Abutting

Table 2. Denticle characteristics of the anterior margin (AM), centre (C) and posterior margin (PM) from first dorsal fin and dorsal surface of the pectoral fins of Carcharhinus brachyurus *(271 cm TL), C. falciformis (264 cm TL),* C. leucas *(239 cm TL),* C. limbatus *(265 cm TL),* C. longimanus *(178 cm TL) and* C. obscurus *(265 cm TL).*

Species	**Position**	**Dorsal fin**		**Pectoral fin**	
		Length (µm)	Width (µm)	Length (µm)	Width (µm)
Carcharias taurus	AM	301 (± 27)	298 (± 30)	389 (± 10)	386 (± 38)
	C	302 (± 12)	310 (± 12)	274 (± 7)	306 (± 9)
	PM	246 (± 7)	252 (± 13)	284 (± 13)	262 (± 12)
Carcharodon carcharias	AM	314 (± 18)	293 (± 14)	242 (± 12)	236 (± 10)
	C	216 (± 5)	192 (± 6)	233 (± 18)	191 (± 8)
	PM	230 (± 8)	205 (± 4)	225 (± 4)	195 (± 5)
Carcharhinus brachyurus	AM	403 (± 23)	439 (± 22)	473 (± 16)	427 (± 22)
	C	453 (± 7)	410 (± 13)	446 (± 17)	391 (± 18)
	PM	411 (± 24)	403 (± 8)	463 (± 14)	354 (± 15)
Carcharhinus falciformis	AM	252 (± 21)	270 (± 31)	342 (± 5)	391 (± 9)
	C	277 (± 4)	340 (± 8)	318 (± 8)	395 (± 13)
	PM	274 (± 4)	327 (± 14)	337 (± 13)	382 (± 10)
Carcharhinus leucas	AM	752 (± 12)	762 (± 15)	746 (± 13)	751 (± 22)
	C	738 (± 20)	783 (± 28)	695 (± 14)	749 (± 20)
	PM	654 (± 25)	672 (± 14)	721 (± 7)	652 (± 21)
Carcharhinus limbatus	AM	189 (± 7)	194 (± 7)	303 (± 21)	278 (± 10)
	C	225 (± 8)	204 (± 6)	252 (± 5)	255 (± 16)
	PM	222 (± 6)	237 (± 5)	225 (± 4)	256 (± 13)
Carcharhinus longimanus	AM	330 (± 7)	385 (± 13)	307 (± 9)	377 (± 5)
	C	333 (± 11)	423 (± 6)	358 (± 8)	422 (± 7)
	PM	270 (± 10)	371 (± 21)	262 (± 10)	348 (± 11)
Carcharhinus obscurus	AM	401 (± 21)	398 (± 16)	477 (± 18)	437 (± 16)
	C	399 (± 19)	451 (± 23)	453 (± 16)	463 (± 19)
	PM	360 (± 18)	386 (± 17)	485 (± 17)	466 (± 18)

Table 3. Mean (± SE) length and width of denticles sampled from the anterior margin (AM), centre (C) and posterior margin (PM) of the first dorsal fin and dorsal surface of the pectoral fin of Carcharias taurus, Carcharodon carcharias, Carcharhinus brachyurus, C. falciformis, C. leucas, C. limbatus, C. longimanus, *and* C. obscurus.

Comparisons Among Species

The two threatened species, *Carcharias taurus* and *Carcharodon carcharias*, had three continuous ridges on the denticles from all sampling positions on the first dorsal and pectoral fins (Table 1). This single characteristic distinguished both species from the six carcharhinid whalers which had at least five ridges on the denticles from the centre and posterior margin of the first dorsal and pectoral fins (Table 2). *C. taurus* had three cusps on the denticles from all sampling positions on the first dorsal and pectoral fins, whereas *C. carcharias* had no cusps present on the denticles at the anterior margin of both fins (Table 1). This pattern of difference could be used to distinguish the species.

The pattern of difference in the denticle characteristics across the sampling positions could also be used to distinguish the six carcharhinid whalers. For example, the denticles on the dorsal fins of *Carcharhinus brachyurus* and *C. falciformis* exhibited distinct

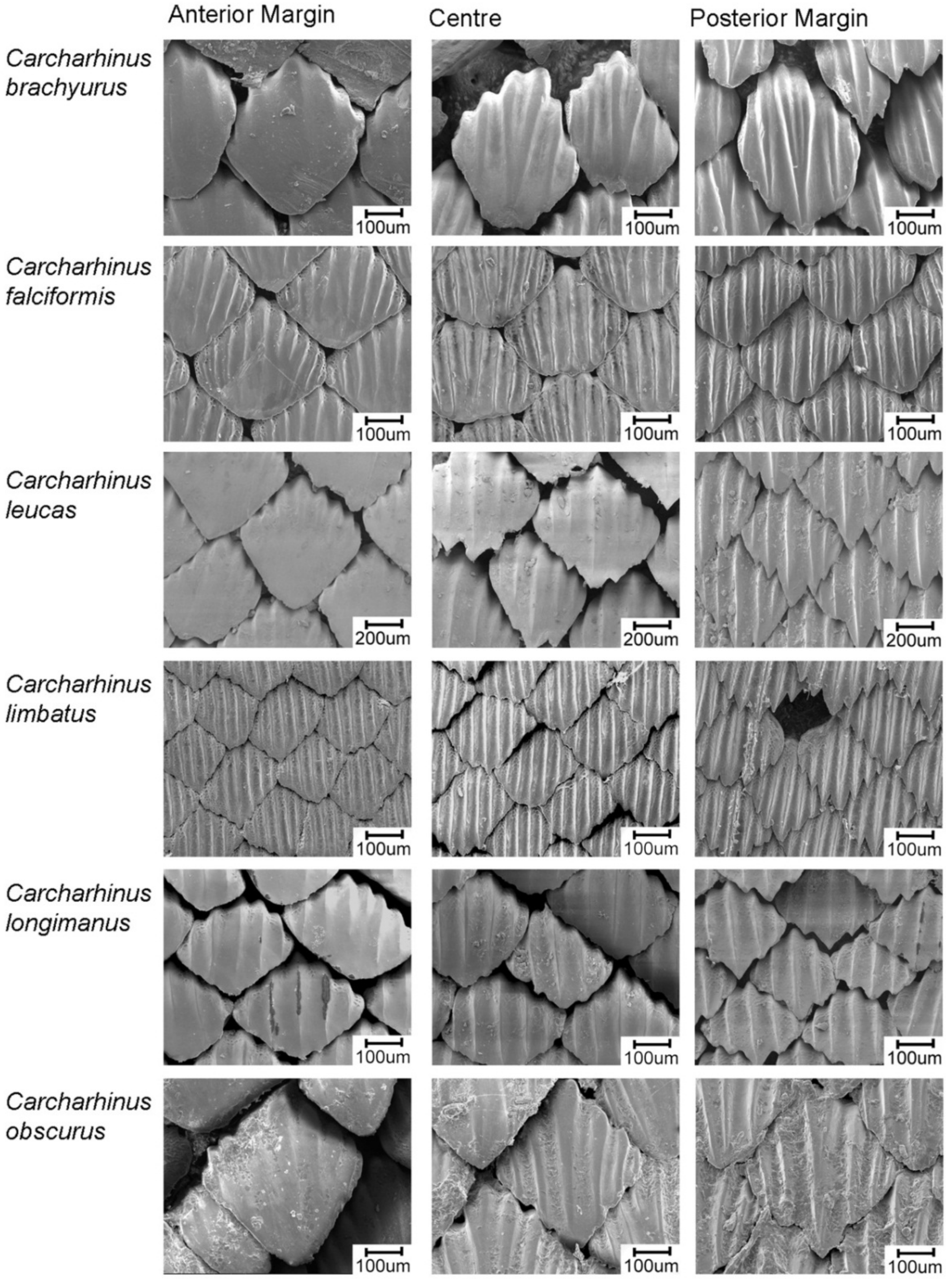

Figure 5. Scanning electron micrographs of denticles from the anterior margin, centre and posterior margin of the dorsal surface of the pectoral fin from Carcharhinus brachyurus *(271 cm TL),* C. falciformis *(264 cm TL),* C. leucas *(239 cm TL),* C. limbatus *(265 cm TL)*, C. longimanus *(178 cm TL), and* C. obscurus *(265 cm TL).*

patterns of difference across the fin. No ridges were present on denticles from the anterior margin in *C. obscurus*, whereas no cusps were present on denticles from the central region in *C. brachyurus* (Table 2). The pectoral fins also showed distinct patterns of difference among species. For example, there were no cusps on denticles across the fins of *C. falciformis*, whereas five to six cusps were present at all sampling positions in *C. limbatus* (Table 2).

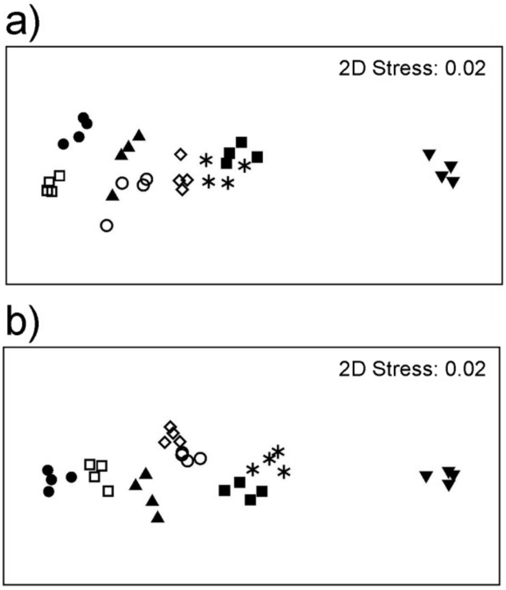

Figure 6. MDS ordination plots of denticle lengths and widths from the anterior margin, centre and posterior margin (n = 4 replicates) for (a) the first dorsal fin and (b) the dorsal surface of the pectoral fin from Carcharias taurus (▲), Carcharodon carcharias (●), Carcharhinus brachyurus (■), C. falciformis (○), C. leucas (▼), C. limbatus (□), C. longimanus (◊), *and* C. obscurus (*).

The size (length & width) of denticles from the first dorsal and pectoral fins also showed distinct patterns of difference across the three sampling positions (Table 3) and suggested that size of denticles could be used to distinguish among species. The size of denticles from the first dorsal fin (Fig. 6a) differed among species (ANOSIM, Global R = 0.944, P < 0.001) and, all except two, pairwise tests were significant (pairwise R = 0.771 – 1.000, P = 0.029). The pairwise tests could not distinguish the differences in denticle-size between *Carcharias taurus* and *Carcharhinus falciformis* (pairwise R = 0.188, P = 0.117), and *C. brachyurus* and *C. obscurus* (pairwise R = 0.365, P = 0.057). The size of denticles from the pectoral fin (Fig. 6b) also differed among species (ANOSIM, Global R = 0.970, P < 0.001) and all pairwise tests were significant (pairwise R = 0.802 – 1.000, P ≤ 0.029) indicating clear separation of the eight sharks species.

Discussion

Varying numbers of sharks, comprising the eight species provided representative samples for detailed examination of the denticles from the first dorsal and pectoral fins. The sharks were caught at various locations along the entire NSW coast and this reflected their established geographic ranges (Stevens 1984, Last & Stevens 2009, Otway & Ellis 2011). The incidental capture of *Carcharias taurus* and *Carcharodon carcharias* occurred along the entire NSW coast and was consistent with other studies of the SMP (Krogh & Reid 1996, Reid et al. 2011), commercial fisheries (Pollard et al. 1996, Macbeth et al. 2009) and the recreational fishing sector (Pepperell 1992, Otway et al. 2004).

Comparisons Among Species

The overall appearances of the denticles, irrespective of sampling position, were in general agreement with earlier descriptions (Garrick 1982, Bargar & Thorson 1995, Matsunaga et al. 1998, Nakano & Kitamura 1998). While some studies (e.g. Garrick 1960, Applegate 1967) have documented size-related differences in denticle characteristics particularly with small juveniles, there were no marked differences in denticle size and characteristics between the sexes and among individuals >150 cm TL. This result was in agreement with Salini et al. (2007) who showed that there were limited changes in denticle characteristics after a species-specific TL was attained (e.g. *C. limbatus* >150 cm TL). However, the reduced numbers of neonates and juveniles (0–2 years) examined in this study (particularly with whalers) prevented an extensive evaluation of possible size-related differences in denticle characteristics in very small sharks. Quantifying the degree of variation and its implications for species identification in young individuals will require additional sampling in the future.

Previous studies (e.g. Bagar & Thorson 1995, Garrick 1982 , Salini et al. 2007) have shown denticle characteristics can vary with location on the fins and/or torso. Variation in denticle characteristics across the fins was also clearly evident in this study. Despite this, it was still possible to distinguish species through either clear differences in denticle characteristics from one or all of the sampling positions. The number of denticle ridges was the most obvious character that distinguished groups of similar species. For example, the carcharhinid whalers had five to seven ridges on denticles from at least one sampling position, whereas *Carcharias taurus* and *Carcharodon carcharias* had only three ridges on the denticles from each sampling position.

Patterns of change in denticle characteristics across the sampling positions could also be used to discriminate species or groups of similar species. For example, the dispersion of dorsal fin denticles across the three sampling positions separated three distinct groups within the carcharhinid whalers. Three species (*C. brachyurus*, *C. leucas* and *C. limbatus*) displayed no change in the overlapping denticle dispersion across sampling positions. In contrast, the dispersion of denticles changed from abutting at the anterior margin to overlapping at the centre and posterior margin in *C. falciformis* and *C. longimanus*. Finally, *C. obscurus* had abutting denticle dispersion which did not change across sampling positions. With this in mind, future studies should quantify denticle characteristics from several sampling positions on a fin as this will enable patterns of change (or no change) in denticle characteristics for augmenting the identification of shark species.

The use of denticle size (i.e., length and width) as a method for differentiating species has not been explored in previous studies. Similar to the denticle characteristics, there were distinct patterns of change in denticle size across the three sampling positions. The size of denticles from the first dorsal fin separated all species except *Carcharias taurus* and Carcharhinus falciformis, and *Carcharhinus brachyurus* and Carcharhinus obscurus, respectively. However, the size of denticles from the pectoral fin unequivocally separated all eight species. Combining denticle size with the other denticle characteristics provides an efficacious approach to species identification, and a method for enhancing taxonomic keys developed previously (e.g. Garrick 1982, Compagno 2002, Last & Stevens 2009). For example, if only denticle size from the dorsal fins was used, the additional denticle characteristics and dispersion provided a method of unequivocally separating species.

Large-Scale Geographic Comparisons

The denticle characteristics at the anterior margin and centre of the first dorsal fin of *C. leucas* from NSW waters were entirely consistent with those described from Costa Rica (Bagar & Thorson 1995). Similarly, the denticle characteristics from the centre of the first dorsal fins of *Carcharhinus longimanus*, *C. falciformis* and *C. obscurus* from NSW waters were consistent with those observed from these species in Japanese waters (Tanaka et al. 2002). While the precise sampling details have been provided in several studies (e.g. Dingerkus & Koestler 1986, Raschi & Tabit 1992, Mojetta 1997), others have not described exactly where, on the fins or torso, the denticles were obtained. For example, the specific sampling location on the first dorsal fin of Carcharhinus longimanus was not provided by Matsunaga et al. (1998) and while the denticle characteristics were similar to those described here, at least two different conclusions could be drawn. If Matsunaga et al. (1998) sampled the denticles from the centre or posterior margin of the fin, the denticles would be similar suggesting no large-scale geographic variation. Alternatively, if Matsunaga et al. (1998) sampled the denticles from the anterior margin of the first dorsal fin, then differences would have been evident and attributed to large-scale geographic variation.

More generally, the absence of detailed sampling information has lessened the number of large-scale geographic comparisons and reduced the efficacy of using denticle characteristics in shark taxonomy (e.g. Garrick 1982). To redress this, future studies should describe precisely where the denticles are sampled as this will enable unconfounded comparisons of denticle characteristics within and among species, and between geographic regions.

Management Implications for the Shark-Fin Trade

With the ever-increasing demand for shark fins, the denticle characteristics documented in this study provide an alternative to genetic techniques (Shivji et al. 2002, Abercrombie et al. 2005, Clarke et al. 2007) for quantifying the species composition and quantities of sharks harvested for their fins in NSW waters.

Moreover, with the continuing incidental capture of *Carcharias taurus* and *Carcharodon carcharias* by the SMP, commercial and recreational fishers documented in this and other recent studies (Bruce et al. 2006, Macbeth et al. 2009, Otway & Ellis 2011, Reid et al. 2011), it is likely that the fins of these threatened species will find their way, albeit illegally, into the shark-fin trade. The denticle characteristics of *Carcharias taurus* and *Carcharodon carcharias* will permit an unequivocal, cost-effective method for detecting the presence of both species within the domestic (Rose & McLoughlin 2001, Lack & Sant 2006) and international (Rose 1996, Clarke 2004, Clarke et al. 2006) shark-fin trade and enable their illegal catch in NSW waters and elsewhere to be quantified.

Additionally, the populations of *Carcharhinus brachyurus*, *C. leucas*, *C. limbatus*, and *C. obscurus* are recognised as near threatened globally, whereas *C. longimanus* is recognised as globally threatened (i.e., Vulnerable) on the IUCN Red List (Cavanagh et al. 2003, Dulvy et al. 2008). The denticle characteristics of these species will also permit their identification and catches to be quantified.

Conclusion

Replicated sampling of various denticle characteristics from the three sampling positions on the first dorsal and pectoral fins of sharks of varying TL provided an efficacious means of unequivocally identifying eight shark species including the critically endangered *Carcharias taurus* and vulnerable *Carcharodon carcharias*. When sampling denticles, it is imperative that future studies ensure that the sampling locations (on the fins or torso) are adequately described to permit unconfounded comparisons of denticle characteristics within and among species, and over large geographic scales.

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References

Abercrombie, D.L., Clarke, S.C. & Shivji, M.S. (2005) Global-scale genetic identification of hammerhead sharks: Application to assessment of the international fin trade and law enforcement; *Conservation Genetics*; **6**, 775-788.

Applegate, S.P. 1967, "A survey of shark hard parts"; in: *Sharks, Skates, and Rays*. P.W. Gilbert, R.F. Mathewson and D.P. Rall, eds. John Hopkins Press, Baltimore, 37-67.

Bagar, T.W. & Thorson, T.B. (1995) A scanning electron microscope study of the dermal denticles of the bull shark, *Carcharhinus leucas*; *Journal of Aquariculture & Aquatic Sciences*, **7**, 120-137.

Baum, J.K. & Myers, R.A. (2004) Shifting baselines and the decline of pelagic shark populations in the Gulf of Mexico. *Ecology Letters*; **7**, 135-145.

Baum, J.K., Myers, R.A., Kelher, D.G., Worm, B. & Harley, S.J. 2003, Collapse and conservation of shark populations in the northwest Atlantic. *Science*, **299**, 389-392.

Bruce, B,D., Stevens, J.D. & Malcolm, H. 2006, Movements and swimming behaviour of white sharks (*Carcharodon carcharias*) in Australian waters. *Marine Biology*, **150**, 161-172.

Campana, S.E., Joyce, E. & Manning, M.J. 2009, Bycatch and discard mortality in commercially caught blue sharks Prionace glauca assessed using archival satellite pop-up tags. *Marine Ecology Progress Series*, **387**, 241-253.

Cavanagh, R., Kyne, P., Fowler, S.L., Musick, J.A. & Bennett, M.B. 2003, The conservation status of Australasian chondrichthyans. Report of the IUCN Shark Specialist Group Australia and Oceania Regional Red List workshop, 7-9 March 2003, University of Queensland, Brisbane, Queensland.

Compagno, L.V.J. 2002, Sharks of the world. An annotated and illustrated catalogue of shark species known to date. 2, Bullhead, Mackerel and Carpet Sharks (Heterodontiformes, Lamniformes and Orectolobiformes). Food and Agriculture Organization of the United Nations, Rome.

Clarke, S. 2004, Understanding pressures on fishery resources through trade statistics: a pilot study of four products in the Chinese dried seafood market. *Fish & Fisheries*, **5**, 53-74.

Clarke, S.C. Magnussen, J.E., Abercrombie, D.L., Mcallister, M.K. & Shivji, M.S. 2006, Identification of shark species composition and proportion in the Hong Kong shark fin market based on molecular genetics and trade records. *Conservation Biology*, **20**, 1, 201-211.

Clarke S.C., McAllister, M.K., Milner-Gulland, E.J., Kirkwood, G.P., Michielsens, C.G.J., Agnew, D.J., Pikitich, E.K., Nakano, H. & Shivji, S. 2006, Global estimates of shark catches using trade records from commercial markets. *Ecology Letters*, **9**, 1115-1126.

Clarke, S., Milner-Gulland, E.J. & BjØrndal, T. 2007, Social, economic, and regulatory drivers of the shark fin trade. *Marine Resource Economics*, **22**, 305-327.

Cortés, E. 2000, Life-history patterns and correlation in sharks. *Reviews in Fisheries Science*, **8**, 299-344.

Dingerkus, G. & Koestler, R.J. 1986, Applications of scanning electron microscopy to the study of shark dermal denticles. In: *Scanning Electron Microscopy, II*. O. Johari, ed. SEM Inc, Chicago, 513-519.

Dulvy, N.K., Baum, J.K., Clarke, S., Compagno, L.J.V., Cortes, E., Domingo, A., Fordham, S., Fowler, S., Francis, M.P., Gibson, C., Martinez, J., Musick, J.A., Soldo, A., Stevens, J.D. & Valenti, S. 2008, You can swim but you cant hide: the global status and conservation of oceanic pelagic sharks and rays. *Aquatic Conservation: Marine & Freshwater Ecosystems*, **18**, Issue 5, 459-482.

FAO (Food and Agriculture Organization). 2007, FishSTAT Capture Production (1950-2005) Database. Food and Agriculture Organization of the United Nations, Rome.

Field, I.C., Meekan, M.G., Buckworth, R.C. & Bradshaw, C.J.A. 2009a, Protein mining the worlds oceans: Australasia as an example of illegal expansion-and-displacement fishing. *Fish & Fisheries*, **10**, 323-328.

Field, I.C., Meekan, M.G., Buckworth, R.C. & Bradshaw, C.J.A. 2009b, Susceptibility of sharks, rays and chimaeras to global extinction. *Advances in Marine Biology*, **56**, 275-363.

Francis, M. 2006, Morphometric minefields – towards a measurement standard for chondrichthyan fishes. *Environmental Biology of Fishes*, **77**, 407-421.

Garrick, J.A. 1960, Studies on New Zealand Elasmobranchii. Part XII. The species of *Squalus* from New Zealand and Australia; and a general account and key to the New Zealand *Squaloidea*. *Transactions of the Royal Society of New Zealand*, **88**, 519-557.

Garrick, J.A.F. 1982, *Sharks of the genus* Carcharhinus. National Oceanic & Atmospheric Administration Technical Report, National Marine Fisheries Service, Circular 445, Washington, DC.

Henry, G.W. & Lyle, J.M. 2003, *The National Recreational and Indigenous Fishing Survey.* Final Report to the Fisheries Research & Development Corporation and Fisheries Action Program Project FRDC 1999/158. Fisheries Final Report Series No. 48. Cronulla, New South Wales.

Krogh, M. 1994, Spatial, seasonal and biological analysis of sharks caught in the New South Wales protective beach meshing programme. *Australian Journal of Marine & Freshwater Research*, **454**, 1087-1106.

Krogh, M. & Reid, D. 1996, Bycatch in the protective shark meshing programme off south eastern New South Wales, Australia. *Biological Conservation*, **77**, 219-226.

Lack, M. & Sant, G. 2006, *World shark catch, production and trade, 1990-2003.* Australian Government Department of Environment and Heritage and TRAFFIC Oceania.

Last, P.R. & Stevens, J.D. 2009, *Sharks and Rays of Australia.* CSIRO Publishing, Melbourne, Victoria.

Macbeth, W.G., Geraghty, P.T., Peddemors, V.M. & Gray, C.A. 2009, Observer-based study of targeted commercial fishing for large shark species in waters off northern New South Wales. Industry & Investment NSW. Fisheries Final Report Series No. 114. Cronulla, New South Wales.

Marin, Y.H., Brum, F., Barea, L.C. & Chocca, J.F. 1998, Incidental catch associated with swordfish longline fisheries in the south-west Atlantic Ocean. *Marine & Freshwater Research*, **49**, 633–640.

Matsunaga, H., Kitamura, T. & Mizoguchi, M. 1998, *Preliminary results of species identification methods of the shark fin.* International Commission for the Conservation of Atlantic Tunas report 48, 90-95.

Mojetta, A. 1997, *Sharks: History and biology of the lords of the sea.* Thunder Bay Press, ISBN 1571451781.

Mollet, H. F. & Cailliet, G.M. 2002, Comparative population demography of elasmobranchs using life history tables, Leslie matrices and stage based matrix models. *Marine & Freshwater Research*, **53**, 503-516.

Myers, R.A. & Worm, B. 2003, Rapid world-wide depletion of predatory fish communities. *Nature*, **423**, 280-283.

Nakano, H. & Kitamura, T. 1998, *Identification of eleven sharks caught by tuna longline using morphological characters of their fins.* Food and Agricultural Organization Technical Working Group for Conservation and Management of Sharks. Food and Agricultural Organization, Rome.

Otway, N.M., Bradshaw, C.J.A. & Harcourt, R.G. 2004, Estimating the rate of quasi-extinction of the Australian grey nurse shark (*Carcharias taurus*) population using deterministic age- and stage-classified models. *Biological Conservation*, **119**, No. 3, 341-50.

Otway, N.M. & Ellis, M.T. 2011. Pop-up archival satellite tagging of *Carcharias taurus*: movements and depth/temperature-related use of south-eastern Australian waters. *Marine & Freshwater Research*, **62**, 607-620.

Pauly, D., Watson, R. & Alder, J. 2005, Global trends in world fisheries: impacts on marine ecosystems and food security. *Philosophical Transactions of the Royal Society B-Biological Sciences*, **360**, 5-12.

Pepperell, J.G. 1992, Trends in the distribution, species composition and size of sharks caught by gamefish anglers off south-eastern Australia. *Australian Journal of Marine & Freshwater Research*, **43**, 213-215.

Pitcher, T.J., Watson, R., Forrest, R., Valtysson, H.P. & Guenette, S. 2002, Estimating illegal and unreported catches from marine ecosystems: a basis for change. *Fish & Fisheries*, **3**, 317-339.

Pollard, D.A., Lincoln Smith, M.P. & Smith, A.K. 1996, The biology and conservation status of the grey nurse shark (*Carcharias taurus* Rafinesque 1810) in New South Wales, Australia. *Aquatic Conservation: Marine & Freshwater Ecosystems*, **6**, 1-20.

Raschi, W. & Tabit, C. 1992, Functional aspects of placoid scales: a review and update. *Australian Journal of Marine & Freshwater Research*, **43**, 123-147.

Reid, D.D. & Krogh, M. 1992, Assessment of catches from protective meshing off New South Wales between 1950 and 1990. *Australian Journal of Marine & Freshwater Research*, **43**, 283-296.

Reid, D., Robbins, W.D. & Peddemors, V.M. 2011, Decadal trends in shark catches and effort from the New South Wales, Australia, Shark Meshing Program 1950–2010. *Marine & Freshwater Research*, **62**, 676–693.

Rose, D.A. 1996, An overview of world trade in sharks and other cartilaginous fishes. TRAFFIC International, Cambridge.

Rose, C. & McLoughlin, K. 2001, *Review of shark finning in Australian fisheries*. Bureau of Rural Sciences, Canberra, Australian Capital Territory.

Salini, J., Giles, J., Holmes, B., Last, P., Marshall, L., Meekan, M., Ovenden, J., Pillans, R., Stevens, J. & Ward, B. 2007, *Species identification from shark fins*. Australian Fisheries Management Authority, R05/0538, Canberra, Australian Capital Territory.

Santana, F.M., Duarte-Neto, P. & Lessa, R. 2009, Demographic analysis of the night shark (Carcharhinus signatus, Poey, 1868) in the equatorial Southwestern Atlantic Ocean. *Fisheries Research*, **100**, 210-214.

Scandol, J., Rowling, K. & Graham, K. (Eds.) 2008, *Status of fisheries resources in NSW 2006/07*. NSW Department of Primary Industries, Cronulla, New South Wales.

Shivji, M., Clarke, S., Pank, M., Natanson, L., Kohler, N. & Stanhopes, M. 2002, Genetic identification of pelagic shark body parts for conservation and trade monitoring. *Conservation Biology*, **16**, No. 4, 1036-1047.

Shivji, M.S., Chapman, D.D., Pikitich, E.K. & Raymond, P.W. 2005, Genetic profiling reveals illegal international trade in fins of the great white shark, *Carcharodon carcharias*. *Conservation Genetics*, **6**, 1035-1039.

Smith, S.E., Au, D.W. & Show, C. 1998. Intrinsic rebound potentials of 26 species of Pacific sharks. Marine & Freshwater Research, 49, 663-678.

Stevens, J.D. 1984, Biological observation on sharks caught by sport fisherman off New South Wales. *Australian Journal of Marine & Freshwater Research*, **35**, 573-590.

Stevens, J.D., Bonfil, R., Dulvy, N.K. & Walker, P. 2000, The effects of fishing on sharks, rays, and chimaeras (chondrichthyans), and the implications for marine ecosystems. ICES *Journal of Marine Science*, **57**, 476-494.

Tanaka, S., Kitamura, T. & Nakano, H. 2002, *Identification of shark species by SEM observation of denticle of shark fins*. International Commission for the Conservation of Atlantic Tuna, **54**, 1386-1394.

Walker, T.J. 1998, Can shark resources be harvested sustainably? A question revisited with a review of shark fisheries. *Marine & Freshwater Research*, **49**, 553-572.

Worm, B., Barbier, E.B., Beaumont, N., Duffy, J.E., Folke, C., Halpern, B.S., Jackson, J.B.C., Lotze, H.K., Micheli, F., Palumbi, S.R., Sala, E., Selkoe, K.A., Stachowicz, J.J. & Watson, R. 2006, Impacts of biodiversity loss on ocean ecosystem services. *Science*, **314**, No. 5800, 787-790.

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