

## Teeth penetration force of the tiger shark *Galeocerdo cuvier* and sandbar shark *Carcharhinus plumbeus*

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This study examined the minimum force required of functional teeth and replacement teeth in the tiger shark *Galeocerdo cuvier* and the sandbar shark *Carcharhinus plumbeus* to penetrate the scales and muscle of sheepshead *Archosargus probatocephalus* and pigfish *Orthopristis chrysoptera*. Penetration force ranged from 7.7–41.9 and 3.2–26.3 N to penetrate *A. probatocephalus* and *O. chrysoptera*, respectively. Replacement teeth required significantly less force to penetrate *O. chrysoptera* for both shark species, most probably due to microscopic wear of the tooth surfaces supporting the theory shark teeth are replaced regularly to ensure sharp teeth that are efficient for prey capture.

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Key words: biomechanics; bite force; Elasmobranchii; teleost; tooth morphology.

### INTRODUCTION

Research on the functional morphology of feeding in sharks has typically focused on the kinematics and mechanics of cranial movement (Ferry-Graham, 1998; Wilga *et al.*, 2001; Motta, 2004; Huber *et al.*, 2005; Motta *et al.*, 2008), often neglecting to integrate the function of teeth (but see Ramsay & Wilga, 2007; Dean *et al.*, 2008; Whitenack *et al.*, 2011). While some studies have examined the design and performance of shark teeth during puncture and draw (Frazzetta, 1988; Lucifora *et al.*, 2001; Whitenack & Motta, 2010; Whitenack *et al.*, 2011; Corn *et al.*, 2016), a further exploration in the complexity of shark tooth structure is essential in elucidating the biological role that teeth play in feeding and particularly during successful prey capture.

Extant fishes have distinct tooth morphologies that appear related to their species-specific feeding behaviours (Cappetta, 1987; Wainwright & Richard, 1995; Kajiura & Tricas, 1996; Ramsay & Wilga, 2007; Whitenack & Motta, 2010). Stemming from these various tooth morphologies and accompanying diets, there has been a tendency to classify shark teeth into clutching, puncturing, slicing, crushing and grinding ecomorphological types (Moss, 1977; Cappetta, 1987; Frazzetta, 1988). For example, the broad, serrated teeth found in the white shark *Carcharodon carcharias* (L. 1758) have been suggested as a characteristic of species that gouge chunks of flesh

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from large, soft-prey items, whereas the long, pointed teeth in the sand tiger shark *Carcharias taurus* Rafinesque 1810 exemplify a piscivorous diet that pierce and grasp fast moving fishes (Frazzetta, 1988; Lucifora *et al.*, 2001; Smale, 2005; Ferrara *et al.*, 2011). Dentitions may also be suited to the stress experienced during feeding: long, pointed teeth for soft-bodied and slippery prey, contrasted to broad, serrated teeth for hard, dense prey (Frazzetta, 1988; Whitenack & Motta, 2010; Motta & Huber, 2012).

In contrast with the historical tendency to group tooth shapes into biomechanical roles, Whitenack & Motta (2010) found that puncturing performance (force to puncture) differed among different prey items, indicating that not all soft prey are alike, but there were few patterns that distinctly categorized tooth performance based on their shape. Material testing of shark teeth during puncturing and cutting (draw) on a variety of hard and soft prey established the majority of teeth were able to puncture different prey items, with broader, triangular tooth types being somewhat less effective at puncture than narrow-cusped teeth. Broad teeth, such as those of the tiger shark *Galeocerdo cuvier* (Péron & LeSueur 1822), often failed in puncture of hard and soft prey, simply crushing the prey when driven directly down into it. Their study revealed force to puncture bony fishes was remarkably low, ranging from <1 to 67 N. Force to puncture in bony fish teeth have been found in low ranges from <1 to 2 N (Galloway *et al.*, 2016).

Low forces for puncture stand in contrast to relatively high bite forces found in many sharks. For instance, the bite force of *C. carcharias*, the bull shark *Carcharhinus leucas* (Valenciennes 1839) and the great hammerhead shark *Sphyrna mokarran* (Rüppell 1837) place among the highest of all extant vertebrates and produce a considerably higher bite force than what is required for puncture (Habegger *et al.*, 2012). Durophagous species (hard-prey consumers) generate particularly high bite forces suggesting an evolutionary strategy for processing hard-prey items. Conversely, jaw closing mechanisms in piscivorous sharks may be more speed efficient rather than force efficient, as in the blacktip shark *Carcharhinus limbatus* (Valenciennes 1839) and geared towards capturing soft-prey items (Huber *et al.*, 2005; Whitenack & Motta, 2010). Consistent with the bite forces of large sharks, a finite element analysis of shark teeth indicated they are structurally strong and suited for a suite of prey capture strategies (Whitenack *et al.*, 2011).

Fossil records suggest the earliest shark teeth evolved from dermal denticles that lined the mouth and with an increase in dietary diversity, elasmobranchs encompassed a variety of tooth sizes and shapes to accommodate more heterogeneous prey options (Zangerl, 1981; Motta & Huber, 2012). As diet progressed to include increasingly larger prey, the ancestral slow tooth replacement condition was lost and evolved to synchronous, rapid tooth replacement by moving newer teeth lingually in waves to serve as functional teeth (Motta & Huber, 2012; Rasch *et al.*, 2016). The evolutionary combination of structurally strong, polyphyodont dentition combined with high bite force suggests the frequency of tooth replacement in sharks is driven by tooth wear, not tooth failure (Whitenack *et al.*, 2011; Motta & Huber, 2012; Corn *et al.*, 2016), an idea also supported by dynamic testing of shark teeth (Corn *et al.*, 2016). Consequently, replacement teeth should be sharper and require less force to penetrate prey than worn, functional teeth.

To define feeding processes and behaviour in sharks, determining how different tooth morphotypes function, including force to penetration, is critical in understanding prey capture and processing. Therefore, this study investigated the minimum penetration force of two shark species with dissimilar tooth morphologies [the sandbar shark

*Carcharhinus plumbeus* (Nardo 1827) and *G. cuvier*], during penetration of teleost prey with large and small elasmoid scales [sheepshead *Archosargus probatocephalus* (Walbaum 1792) and pigfish *Orthopristis chrysoptera* (L. 1766)]. This study also seeks to determine if older, functional teeth require more force to penetrate prey than unused and presumably sharper replacement teeth. For analysis purposes, functional and replacement teeth have been categorized as tooth types. Penetration is defined here as the initial piercing of the prey by the tooth tip, whereas puncture might involve further excursion of the tooth into the prey requiring more force as the surface area of the tooth increases.

## MATERIALS AND METHODS

### SPECIMENS FOR PENETRATION TESTING

Teeth from the functional row and first replacement row of *C. plumbeus* and *G. cuvier* were glued to a cantilever beam and driven into the epaxial muscle below the dorsal fin of two species of bony fishes, *A. probatocephalus* [ $n=6$ , mean range standard length ( $L_S$ ) 25–30 mm], with larger elasmoid scales ( $n=20$ , mean  $\pm$  s.d. thickness and diameter  $0.2 \pm 0.0$  mm and  $10.2 \pm 0.7$  mm, respectively) and *O. chrysoptera* ( $n=6$ , mean range  $L_S$  15–20 mm), with smaller elasmoid scales ( $n=20$ , mean  $\pm$  s.d. thickness and diameter  $0.1 \pm 0.02$  mm and  $3.4 \pm 0.3$  mm, respectively). Additionally, scale overlap was determined from two specimens of each species by marking and removing scales from the test area and measuring the overlap with a digital micrometre and expressed as per cent overlap. For *A. probatocephalus* and *O. chrysoptera* the scales of the epaxial region overlapped by 50 and 52%, with 50 and 48% of the posterior edge of the scale protruding beyond the scale immediately anterior to it, respectively. One jaw from each shark species was used in this study ( $n=2$ ). Three functional and three immediately posterior replacement teeth were removed from the lower jaw (Lowry *et al.*, 2009). Lower-jaw teeth were utilized as these teeth generally are more pointed in carcharhiniform sharks and impale the prey first, whereas the triangular upper teeth saw through the prey during lateral head shaking (Frazzetta & Prange, 1987; Compagno, 1988; Frazzetta, 1988; Smale *et al.*, 1995; Motta & Huber, 2012). Each tooth underwent three penetration trials first on *O. chrysoptera* and then on *A. probatocephalus* to reduce any possible dulling of teeth, for a total of 72 trials. Each teleost endured a maximum of nine penetrations per epaxial region, such that penetration holes did not overlap. Teleost specimens were frozen within 1 h of landing and thawed in cool water for 1 h prior to testing. Trials were conducted on each teleost for no  $>1$  h after thawing. The shark jaws and attached teeth were provided from Gulf of Mexico waters by recreational and commercial fishers. The fishers, however, neglected to include total length ( $L_T$ ), so applying inter-tooth distance methods according to Lowry *et al.* (2009), the estimated  $L_T$  range of the sharks *C. plumbeus* and *G. cuvier* were 2027–2074 and 1998–2048 mm, respectively.

### PENETRATION TESTING

Each tooth was fixed with epoxy glue to a removable 27.3 mm diameter PVC coupling (Fig. 1). The coupling was fitted over one end of the 1.25 cm diameter PVC cantilever beam of 60 cm total length. The beam was pivoted at its centre and mounted to a concrete block for stability. Upon completion of each trial, the coupling and attached tooth were removed and replaced for the next tooth's trial. Thus, the six replacement teeth and six functional teeth were glued onto 12 associated PVC couplings. At the end opposite the tooth, a digital scale (RGSDS 15/50 LB., Rapala; www.rapala.com) determined the minimum force at the time of tooth penetration. The Rapala scale was subsequently calibrated against two digital scales: a SILTEC electronic weighing scale (siltec.balances.com) and a Brainweigh B1500 OHAUS (OHAUS Scale Corporation; www.ohaus.com/en-US). A least-squares linear regression of the Rapala force *v.* the SILTEC and OHAUS scales resulted in  $r^2$  values of 0.98 and 0.96, respectively. In both cases the Rapala scale measurement was on average 1.7 N greater than the two calibration scales, so this value

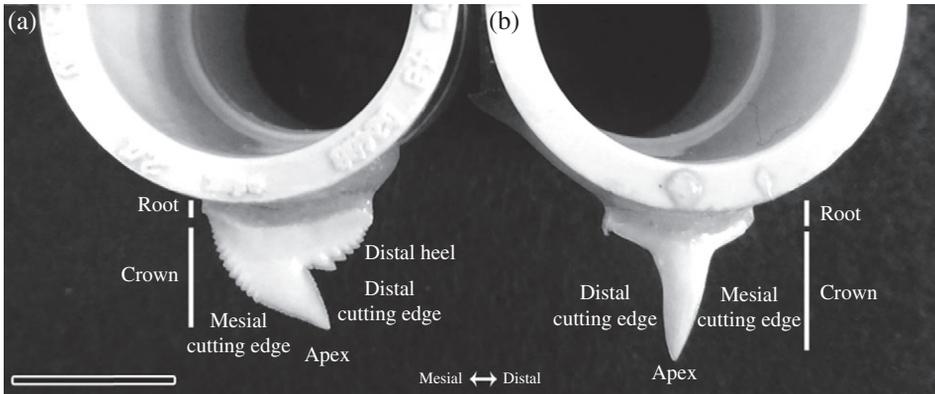


FIG. 1. Teeth from shark species mounted on removable PVC coupling. (a) Replacement tooth position 3 from the right side of the lower jaw of *Galercerdo cuvier* showing: crown, root, serrated cutting edges, apex of crown and the serrated distal heel. (b) Replacement tooth position 4 from the left side of the lower jaw of *Carcharhinus plumbeus* showing: crown, root, serrated cutting edges and apex of crown. Scale bar = 1 cm.

was subtracted off all penetration forces. Teleost specimens were placed directly beneath and touching the tooth tip at the same height as the cantilever beam, such that the beam was horizontal at the initiation of the trial. The tooth was positioned on the centre of an elasmoid scale in the epaxial region of the specimen, but ventral to the bony pterygiophores of the dorsal fin. The digital scale was pulled up vertically by hand at the other end of the beam as consistently as possible to ensure a set rate of tooth movement. For each trial, the minimum force at penetration and rate of tooth movement was recorded.

A Fastec imaging high-speed camera model IN500M512 (Fastec Imaging Corporation; [www.fastecimaging.com](http://www.fastecimaging.com)) recorded the penetration trial at 250 fields (f)  $s^{-1}$ , documenting when initial penetration occurred and the rate of tooth movement determined as the distance moved by the tooth apparatus from the start of loading (with the tip just touching the fish) until the moment the tooth first penetrated the fish and then divided by elapsed time. The Rapala scale was simultaneously used to record and store the force at penetration. The force at penetration was visually observed and recorded the instant the tooth first penetrated the fish (standardized as the moment the tooth broke through the scale or flesh). Depth of tooth penetration at this moment was measured from the video sequences and averaged; mean  $\pm$  s.d. =  $3.3 \pm 0.2$  mm. Video sequences (Fig. 2) were analysed with Redlake Motionscope software 2.30.0 (DEL Imaging Systems; [www.delimaging.com/high-speed-cameras](http://www.delimaging.com/high-speed-cameras)).

## TOOTH MEASUREMENTS

To quantify tooth morphology, the following tooth measurements were collected: base-cusp width ( $W_{BC}$ ; maximum cusp width), cusp height ( $H_C$ ; perpendicular from cusp apex to base-cusp width), mesial cutting edge length ( $L_{MCE}$ ; distance between cusp apex and most mesial point of base-cusp width) and distal cutting edge length ( $L_{DCE}$ ; distance between cusp apex and most distal point of base-cusp width) (Fig. 1). From these measurements, cusp inclination ( $A_C$ ; *i.e.*  $L_{MCE}L_{DCE}^{-1}$ ) was calculated. Serrations were observed under a stereomicroscope and enumerated using methods from Moyer & Bemis (2016). The number of primary serrations  $mm^{-1}$  midway along the tooth's edge were counted on both the mesial and distal cutting edges. On the distal cutting edge of *G. cuvier* teeth, primary serrations were measured along the distal heel. While secondary serrations (effectively serrations within serrations) can be found on *G. cuvier* teeth (Moyer & Bemis, 2016), only primary serrations were enumerated because *C. plumbeus* primary serrations are almost indistinguishable on the mesial cutting edge and do not have secondary serrations on either cutting edge. Although *G. cuvier* teeth are similar in shape on the

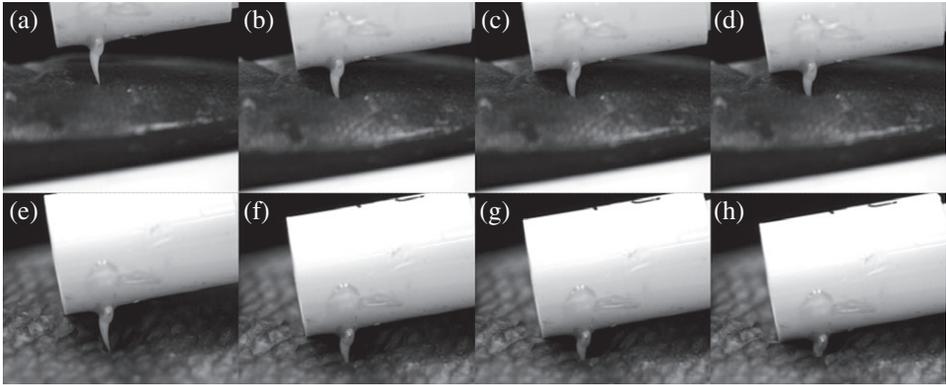


FIG. 2. Sequence showing *Carcharhinus plumbeus* replacement tooth position 4 penetration event on (a)–(d) *Orthopristis chrysoptera* and (e)–(h) *Archosargus probatocephalus*. Sequence order follows from left to right: (a) and (e) initial resting, (b) and (f) initial penetration, (c) and (g) increasing force, and (d) and (h) penetration achieved. Rate of tooth movement ( $\text{mm s}^{-1}$ ) is calculated as distance travelled from initial penetration [(b) and (f)] to penetration achieved [(d) and (h)] over time.

upper and lower jaws, all teeth extracted from both shark species were from the lower jaw to account for any fluctuating asymmetry in tooth shape. Teeth were extracted from the side of the lower jaw that appeared to be in the best condition. From the lower jaw of *G. cuvier*, two teeth were extracted from the left side (replacement tooth position 1 and functional tooth position 1) and four teeth were extracted from the right side (replacement teeth positions 2 and 3 and functional teeth positions 2 and 3). All replacement and functional teeth extracted from the lower jaw of *C. plumbeus* were from the left side positions 3, 4 and 5 (Lowry *et al.*, 2009). No teeth showed any visible differences in tooth-tip damage or damage to the edges, though it should be noted that wear on the tooth edge is microscopic and would be extremely difficult to quantify or even see (Corn *et al.*, 2016; Moyer & Bemis, 2016). The tooth measurements are based on Whitenack & Motta (2010) and Shimada (2002) and were taken with a Leica M80 stereomicroscope ([www.leicabiosystems.com](http://www.leicabiosystems.com)) in conjunction with digital callipers.

## STATISTICAL ANALYSIS

Differences among shark species (*G. cuvier* and *C. plumbeus*), tooth type (functional and replacement) and prey species (*A. probatocephalus* and *O. chrysoptera*) in penetration force ( $N_p$ ) and rate of tooth movement ( $R_p$ ) were evaluated by fitting separate linear mixed models in R ([www.r-project.org](http://www.r-project.org)) using the lme function of the nlme package (Pinheiro *et al.*, 2014) and least-square means of these models using the lsmeans R package (Lenth, 2016). These models were estimated *via* maximum likelihood using the general purpose optimizer (optim) in R. Fixed effects included in each model were shark species, tooth type, prey species and all their potential interaction effects. Random effects structures here aimed to model repeated measures of individual teeth on each prey species by shark species, this was completed by including individual teeth nested by shark species as random effects and prey species nested among individual teeth as secondary random effect structure among shark species. Inclusion of the importance of all random effects were assessed using likelihood ratio tests (LRT which uses a  $\chi^2$  test statistic) contrasting models with and without fixed effect (predicting) structures. These LRT tests were also corrected following Stram & Lee (1994). Random effects that were non-significant, however, were still kept within models to avoid over interpreting main effects due to small sample sizes (Barr *et al.*, 2013). To improve the d.f. of the main hypothesis tests (*e.g.* differences between tooth types), non-significant interaction terms were dropped; results from analyses are described as reduced models (see regression equations in Tables I and II). Finally, both  $R_p$  and  $N_p$  were  $\log_e$  transformed prior to regression modelling.

TABLE I. Linear mixed-model regression testing for differences in penetration force ( $N_p$ ) among shark species, tooth type and prey species (see Fig. 3)

Regression model	$F$	$\chi^2$	d.f.	$P$
Full model*				
Fixed effects variables				
Shark species	2.81		1.8	>0.05
Tooth type	5.31		1.8	<0.05
Prey species	112.19		1.8	<0.001
Shark species $\times$ tooth type	0.02		1.8	>0.05
Shark species $\times$ prey species	0.01		1.8	>0.05
Tooth type $\times$ prey species	10.81		1.8	<0.05
Shark species $\times$ tooth type $\times$ prey species	0.06		1.8	>0.05
Random effects (LRT test)		45.4	1	<0.001
Reduced model†				
Fixed effects variables				
Shark species	2.93		1.9	>0.05
Tooth type	5.56		1.9	<0.05
Prey species	117.54		1.10	<0.001
Tooth type $\times$ prey species	11.32		1.10	<0.01
Random effects (LRT test)		45.3	1	<0.001

Full model includes all non-significant interactions; reduced model excludes non-significant interaction effects. The significance of inclusion of random effects in model was assessed using a corrected likelihood ratio test (LRT; Stram & Lee, 1994).

Mixed-model equations have random effects described parentheses with vertical bars (|) emphasizing the nestedness of random factors aimed at modelling repeated measures of teeth among prey samples.

\*Model:  $N_p \sim$  shark species  $\times$  tooth type  $\times$  prey species + (shark species | tooth type) + (shark species | tooth type  $\times$  prey species).

†Model:  $N_p \sim$  shark species + tooth type  $\times$  prey species + (shark species | tooth type) + (shark species | tooth type  $\times$  prey species).

## RESULTS

Overall, penetration force in successful tooth performance trials was low and ranged from 3.2 to 41.9 N. While both teleost species, *A. probatocephalus* and *O. chrysoptera*, are considered soft prey, the force required to penetrate the robust scales of *A. probatocephalus* was significantly higher ( $P < 0.001$ ; Table I and Fig. 3) compared with the thinner scales of *O. chrysoptera* for both functional and replacement tooth types (Table I). There was no evidence for differences in penetration force between teeth of the two shark species ( $P > 0.1$ ; Tables I, III and Fig. 3). Often, teeth from both shark species were incapable of penetrating *A. probatocephalus* scales (Table III).

The present results confirmed an interesting significant interaction between functional and replacement teeth during penetration trials on *O. chrysoptera* ( $P < 0.01$ ; Table I and Fig. 3). Replacement teeth in both *C. plumbeus* and *G. cuvier* required less force to penetrate the epaxial region of *O. chrysoptera* (Fig. 3). No significant difference, however, was found between replacement and functional teeth during penetration trials on *A. probatocephalus* in either shark species, most probably because of a lack of successful penetration trials.

TABLE II. Linear mixed-model regression testing for differences in rate of tooth movement ( $R_p$ ) among shark species, tooth type and prey species (see Fig. 4)

Regression model	$F$	d.f.	$\chi^2$	d.f.	$P$
Full model*					
Fixed effects					
Shark species	13.31			1.9	<0.01
Tooth type	0.01			1.9	>0.05
Prey species	0.72			1.6	>0.05
Tooth type $\times$ prey species	1.15			1.6	>0.05
Random effects (LRT test)			10.1	1	<0.01
Reduced model†					
Fixed effects					
Shark species	12.50			1.9	<0.01
Tooth type	0.01			1.9	>0.05
Prey species	0.72			1.7	>0.05
Random effects (LRT test)			8.9	1	<0.01

Mixed-model equations have random effects described parentheses with vertical bars (|) emphasizing the nestedness of random factors aimed at modelling repeated measures of teeth among prey samples.

\*Model:  $R_p \sim$  shark species + tooth type  $\times$  prey species + (shark species | tooth type) + (shark species | tooth type  $\times$  prey species).

†Model:  $R_p \sim$  shark species + tooth type + prey species + (shark species | tooth type) + (shark species | tooth type  $\times$  prey species).

For the functional and replacement teeth of *G. cuvier*, tooth measurements (mean  $\pm$  s.d.) were:  $W_{BC} = 12.7 \pm 1.1$  and  $13.4 \pm 1.6$  mm;  $H_C = 7.8 \pm 0.4$  and  $8.5 \pm 0.8$  mm;  $L_{MCE} = 12.2 \pm 1.0$  and  $14.1 \pm 1.7$  mm;  $L_{DCE} = 8.4 \pm 0.3$  and  $9.4 \pm 0.2$  mm;  $A_C = 1.5 \pm 0.1$  and  $1.5 \pm 0.1$  mm, respectively. For the functional and replacement teeth of *C. plumbeus*, tooth measurements (mean  $\pm$  s.d.) were as

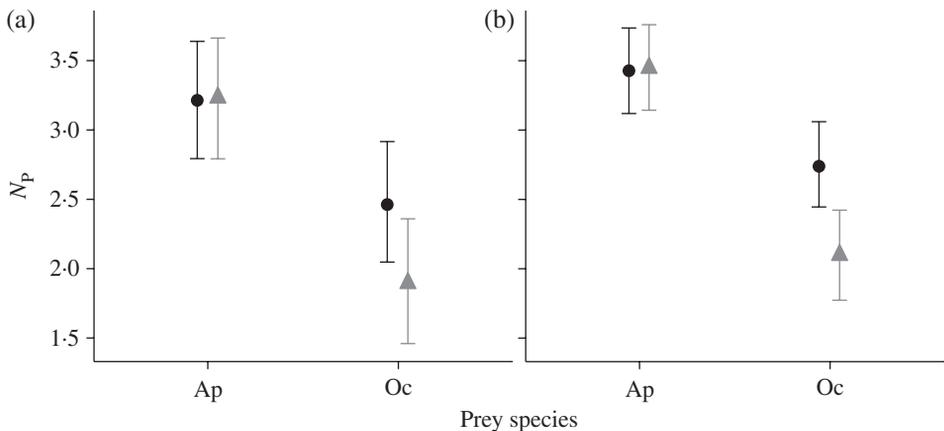


FIG. 3. Full mixed model (Table I) least-square mean  $\pm$  95% c.i. differences in tooth penetration force ( $N_p$ ) among (a) *Carcharhinus plumbeus* and (b) *Galercerdo cuvier* using functional (●) or replacement teeth (▲) on two prey species: *Archosargus probatocephalus* (Ap) and *Orthopristis chrysoptera* (Oc).

TABLE III. Force to penetration ( $N_p$ ) for functional and replacement teeth of two shark species

Shark species	Tooth type	Prey species				
		<i>Archosargus probatocephalus</i>		<i>Orthopristis chrysoptera</i>		Species mean
		$n$	$N_p$ (mean $\pm$ S.D.)	$n$	$N_p$ (mean $\pm$ S.D.)	
<i>Galeocerdo cuvier</i>	Functional	0	–	9	16.5 $\pm$ 5.5	16.5 $\pm$ 0.0
	Replacement	2	35.7 $\pm$ 0.6	9	8.7 $\pm$ 3.5	22.2 $\pm$ 19.1
<i>Carcharhinus plumbeus</i>	Functional	3	12.2 $\pm$ 4.5	9	12.1 $\pm$ 2.7	12.1 $\pm$ 0.0
	Replacement	5	30.5 $\pm$ 13.0	9	7.8 $\pm$ 3.4	19.0 $\pm$ 16.2
Prey mean		10	26.1 $\pm$ 6.1	36	11.2 $\pm$ 3.8	

–, unsuccessful penetrations;  $n$  = number of successful penetration trials per tooth.

follows:  $W_{BC}$  = 11.1  $\pm$  0.0 and 11.0  $\pm$  0.8 mm;  $H_C$  = 9.0  $\pm$  0.8 and 8.7  $\pm$  0.2 mm;  $L_{MCE}$  = 10.5  $\pm$  0.6 and 10.4  $\pm$  0.7 mm;  $L_{DCE}$  = 10.1  $\pm$  0.8 and 10.2  $\pm$  0.6 mm;  $A_C$  = 1.0  $\pm$  0.0 and 1.0  $\pm$  0.0 mm, respectively. The number of serrations  $\text{mm}^{-1}$  midway along the tooth edge on the mesial cutting edge (mean  $\pm$  S.D.) for *G. cuvier* and *C. plumbeus* were 2.0  $\pm$  0.0 and 7.0  $\pm$  0.0 for functional teeth and 2.0  $\pm$  0.0 and 7.0  $\pm$  1.0 for replacement teeth and on the distal cutting edge were 2.0  $\pm$  0.0 and 7.5  $\pm$  2.1 for functional teeth and 2.0  $\pm$  0.0 and 7.5  $\pm$  0.7 for replacement teeth, respectively.

In general, teeth from both shark species showed difficulty penetrating the tough scales of *A. probatocephalus*. Of the 18 penetration-force trials conducted on *A. probatocephalus*, *G. cuvier* teeth had only two successful penetrations. More often, *G. cuvier* teeth left only indentations in the skin, a response seen in past research (Whitenack *et al.*, 2011; Motta & Huber, 2012). The differences seen in rate of tooth movement ( $P < 0.01$ ; Table II and Fig. 4) between teeth of the two shark species for successful penetration trials (*G. cuvier*  $n = 19$ , mean  $\pm$  S.D. 7.44  $\pm$  4.67  $\text{mm s}^{-1}$ ; *C. plumbeus*  $n = 25$ , mean  $\pm$  S.D. 53.96  $\pm$  94.35  $\text{mm s}^{-1}$ ) suggests the larger surface area of *G. cuvier* teeth in contact during penetration may increase the time required for penetration, if they penetrate at all (Fig. 4).

## DISCUSSION

### BIOMECHANICS OF PREY ITEMS

Whilst the performance of teeth is recognized as a key component in survival and fitness, previous elasmobranch feeding research has largely focused on cutting mechanics, performance and general morphology, but overlooked how the teeth themselves function during feeding. In research examining tooth function during feeding, performance tests have been primarily conducted on different prey items grouped into hard and soft, with an intent on searching for possible significance between the two categories of prey, but not within the categories (Motta, 2004; Huber *et al.*, 2005; Ramsay & Wilga, 2007; Motta *et al.*, 2008; but see Whitenack & Motta, 2010; Enax *et al.*, 2012). Traditionally, prey items have been organized into hard prey, such as crustaceans,

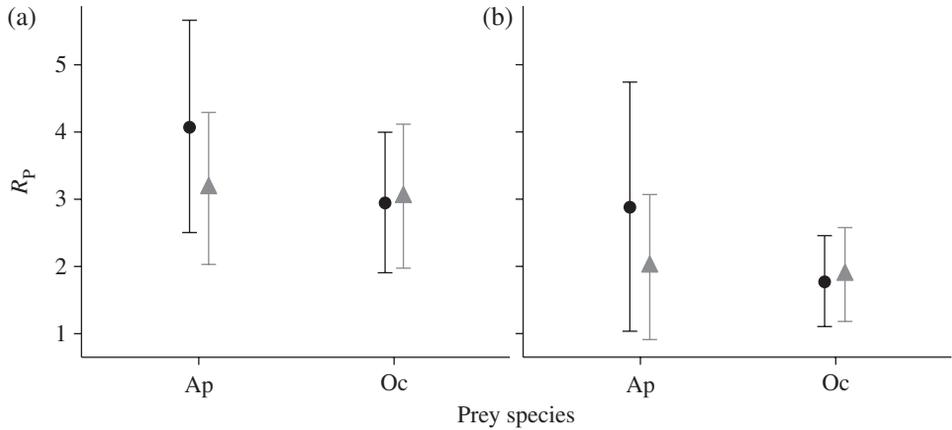


FIG. 4. Full mixed model (Table II) least-square mean  $\pm$  95% c.i. differences in rate of tooth movement ( $R_p$ ) among (a) *Carcharhinus plumbeus* and (b) *Galercerdo cuvier* using functional (●) or replacement teeth (▲) on two prey species: *Archosargus probatocephalus* (Ap) and *Orthopristis chrysoptera* (Oc).

echinoderms and molluscs and soft prey, which encompasses everything else (Wainwright & Richard, 1995; Ramsay & Wilga, 2007; Huber *et al.*, 2009; Whitenack & Motta, 2010). From a functional perspective of shark teeth, however, results from this study question the broad segregation of prey items. Among the two teleosts utilized for performance testing, the thinner scaled *O. chrysoptera* required significantly less force to penetrate and of particular importance, replacement teeth in both shark species required less force to penetrate *O. chrysoptera* compared with functional teeth. No significance was found between tooth types for performance testing on the robust-scaled *A. probatocephalus* probably because of the low number of successful penetrations, with *G. cuvier* successfully penetrating in only two trials.

Though no evidence was found to support functional differences between the two tooth morphologies, the difference observed in penetration force between prey species indicates perhaps the type of soft-prey item being targeted may determine the chance of successful capture more than tooth morphology. Past research has shown shark teeth to quickly dull with use (Corn *et al.*, 2016; Moyer & Bemis, 2016), though prey items with thinner scales should wear teeth at a slower rate. Nevertheless, many sharks are known to opportunistically feed on crustaceans and other hard-prey items (Huber *et al.*, 2005; Mara *et al.*, 2009; Whitenack *et al.*, 2011). The strategy of shedding and replacing dull teeth to enable effective prey capture, regardless of whether the prey item is hard or soft, supports the concept shark teeth are replaced at a species-specific frequency stimulated by tooth wear, not tooth failure (Whitenack *et al.*, 2011; Motta & Huber, 2012; Corn *et al.*, 2016).

## BIOMECHANICS OF TOOTH TYPES

There has been an apparent desire to organize tooth shapes into functionally relevant groups based on performance (Moss, 1977; Cappetta, 1987; Frazzetta, 1988; Motta, 2004) as the key method for classifying the biological role of teeth, since they appear related to feeding behaviours and prey selection (Wainwright & Richard, 1995; Kajiura & Tricas, 1996; Ramsay & Wilga, 2007; Galloway *et al.*, 2016). The lack of support for

morphotypes found in this study is probably because teeth must perform a combination of functions during prey capture and processing. The majority of large, predatory sharks feed with an open-mouthed strike, followed by vigorous and rapid lateral head-shaking motion to cut through tissues with their teeth (Moss, 1972; Tricas & McCosker, 1984; Corn *et al.*, 2016). Often, teeth undergo both puncture and draw-loading regimes within a single predation event (Frazzetta & Prange, 1987; Motta & Wilga, 2001; Whitenack *et al.*, 2011).

Additional to the thick scales of *A. probatocephalus* requiring more force to penetrate, teeth from both shark species often had lower success during force trials on this hard-scaled teleost, suggesting scale thickness among teleosts affects the function of teeth. Notched and multi-cusped teeth such as those found in *G. cuvier* and the sixgill shark *Hexanchus griseus* (Bonnaterre 1788) present a larger surface area upon contact with prey items and when directed orthogonally to the prey, they simply crush or leave indentations instead of penetrate (Whitenack & Motta, 2010; Corn *et al.*, 2016). It should be noted that the low number of successful penetration trials by *G. cuvier* teeth on *A. probatocephalus* is not due to the maximum forces applied being too low. Whitenack & Motta (2010) used a material testing system with a 5 kN load cell and likewise found *G. cuvier* teeth failed to penetrate prey in almost all trials, though when penetration was successful on *A. probatocephalus*, it took a force of 46 N. Highly comparable, the current study found *G. cuvier* replacement teeth to require 36 N to penetrate *A. probatocephalus* (Table III). Although no support was found for grouping tooth morphologies based on performance during prey penetration, it is interesting to further consider the lower number of successful penetrations by *G. cuvier* teeth ( $n = 2$ ). The combination of lower rate of tooth movement in *G. cuvier* teeth while penetrating the scales and muscle in both *O. chrysoptera* and *A. probatocephalus*, along with the general absence of successful penetrations, could further signify this tooth morphology is designed and better suited for cutting (*i.e.* unidirectional draw; Witzell, 1987; Whitenack & Motta, 2010; Motta & Huber, 2012; Corn *et al.*, 2016).

Research examining the force required of teeth to penetrate and puncture prey raises the question of why high bite force is generated in sharks, when force to penetrate is generally low (Huber *et al.*, 2006, 2009; Whitenack & Motta, 2010; Galloway *et al.*, 2016). The excessively high bite forces of shark species suggests an over-development of the feeding mechanism in that they generate forces higher than necessary for puncture (Habegger *et al.*, 2012), with some carcharhinid species generating kN bite forces greater than those required by the prey tissues (Huber *et al.*, 2006, 2009; Wroe *et al.*, 2008; Mara *et al.*, 2009). This study focused on investigating the minimum force required to pierce two species of teleosts in the fleshy epaxial region and did not incorporate additional force required to catch and consume shelled prey, hold struggling prey, or sever tough integument and skeletal elements of larger prey items like turtles, large teleosts, elasmobranchs and marine mammals. It is possible that sharks possess high bite force as a secondary result of having large body size, though there is strong potential that it is tied to initial prey capture (Habegger *et al.*, 2012; Motta & Huber, 2012). Furthermore, high bite force in males may be related to their ability to hold on to females during copulation (Compagno, 1970, 1988; Kajiura & Tricas, 1996; Carrier *et al.*, 2004; Huber *et al.*, 2006; Motta & Huber, 2012).

Overall, the teeth of *C. plumbeus* and *G. cuvier* require low force to penetrate teleost epaxial muscle on both small and large-scaled fishes, agreeing with previous studies (Whitenack & Motta, 2010; Galloway *et al.*, 2016). These findings support the

hypotheses that high bite forces in sharks may in part be linked to prey holding and manipulation, as well as mating (Habegger *et al.*, 2012; Motta & Huber, 2012), as they appear to be unnecessarily high for initial prey penetration during capture of most prey. The results also indicate that replacement teeth are sharper than functional teeth and require significantly less force to penetrate prey items suggesting shark teeth are replaced regularly to ensure sharpness and preserve efficient prey capture. Additionally, perhaps not all soft-prey items can be considered equal, as the thinner, more gracile scales of *O. chrysoptera* required significantly less force to penetrate than the robust, sturdy scales of *A. probatocephalus*. The findings also lend little support to the notion that the shape of shark teeth imply a biological role. To further understand the mechanics and function of shark teeth during prey capture, studies should seek to include a wider range of tooth morphologies and hard and soft-prey items assisted by a material testing system.

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