

Predator declines and morphological changes in prey: evidence from coral reefs depleted of sharks

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ABSTRACT: Evidence from the wild as to the ecological and evolutionary consequences of top predator depletions remains limited, especially in marine systems. Given the pace and extent of predator loss, an understanding of these processes is important. Two sets of adjacent coral reef systems off north-western Australia have similar biological, physical and environmental conditions, but one of the reef systems has been exposed to nearly exclusive commercial fishing of sharks. Across reefs where sharks have been depleted, prey fishes had significantly smaller caudal fins and eyes compared to the reefs with intact shark populations (up to 40 and 46% relative difference in standardized means). These patterns were consistent across 7 teleost prey species (N = 611 individuals) that vary in behavior, diet and trophic guild. We hypothesize that these morphological patterns were primarily driven by differences in shark predation. Morphological differences were not consistent with plausible alternative explanations (habitat complexity, temperature, light, current, food availability, prey targets, competition) as primary drivers. These results provide field evidence of morphological changes in prey potentially due to predator depletions consistent with ecological predictions; specifically, predator loss caused a reduction in the size of prey morphological traits associated with predator detection and evasion. While our analysis cannot differentiate between rapid evolutionary change versus morphological plasticity due to shark depletions, either possible outcome would indicate that predator removals may have profound effects on body shapes of prey communities. This is particularly significant in the case of sharks, given that the consequences of their widespread removal have been a topic of significant speculation, debate and concern.

KEY WORDS: Fishing · Morphology · Predation risk · Predator–prey interactions · Shark · Trophic cascades

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INTRODUCTION

Understanding the impact of upper-level predators on community structure and function is central to ecology, evolutionary biology and wildlife conservation. Predators can impact prey through consumptive effects (predators eating prey) or via risk effects, such

as eliciting anti-predator behavior in prey (e.g. increased vigilance, shifts in habitat use) (Werner & Peacor 2003) or inducing physiological stress responses (Clinchy et al. 2013). These direct effects on prey may initiate trophic cascades that can indirectly impact ecosystem biodiversity, stability, structure and function (Ritchie & Johnson 2009, Estes et al.

2011). To date, studies of predator-induced trophic cascades have typically focused on density-mediated effects on prey, mostly from laboratory experiments, in which removal of predators results in increased abundance of prey due to reduced consumption (Peckarsky et al. 2008). Relatively less attention has been directed to risk effects in wild populations, although experimental measurements are revealing that the strength of risk effects and emergent indirect impacts can equal or exceed those caused through consumption (Creel & Christianson 2008).

Studies in controlled environments (microcosms and mesocosms) and field data show that selection by predators can elicit changes in morphological traits of prey such as body shape and depth (Brönmark & Milner 1992, Hoverman et al. 2005, Oufiero et al. 2011). Some of these predator-induced changes in morphology may represent genetically based differences that have evolved via natural selection (Reznick et al. 1990). In other cases, these changes represent adaptive plasticity in response to the presence of predators, such as the induction of relatively deep tail fins and shorter bodies in tadpoles (McCollum & VanBuskirk 1996). However, examples of how the presence or absence of predators influences the morphology of prey species in wild populations remain rare, especially for top-order predators and larger prey of various species. This gap in knowledge is primarily due to the logistic challenges of working with large predators and prey in natural systems. It also remains poorly understood whether humans can indirectly affect the morphology of other species by altering the abundance of predators in nature. Given the increasing loss of top predators in the wild (Estes et al. 2011) and growing predator recoveries (Marshall et al. 2016), development of an understanding of these processes is timely.

The ecological importance of sharks and their declines, in particular, are active areas of research and debate (Roff et al. 2016, Ruppert et al. 2016). Prior work comparing coral reefs with varying abundances of predators (sharks and other large teleosts) have provided data suggesting that differences in predator abundance associated with varying levels of human activity and fishing intensity can cause demographic shifts in communities of prey fishes (e.g. Ruttenberg et al. 2011) and alterations in behaviors of prey fishes (e.g. Madin et al. 2010, McCauley et al. 2012). However, a potential issue with trying to specifically isolate and assess the potential impacts of shark removals on coral reefs is that other economically important fishes are also usually exploited at the locations where sharks have been heavily fished.

Furthermore, sharks can be absent or rare from some reefs due to variations in habitat structure, coastal development and prey density (e.g. Valdivia et al. 2017).

Two remote sets of adjacent coral reef systems off the coast of north-western Australia provide a unique opportunity to examine the potential effects of targeted shark removals on teleost fishes (Ruppert et al. 2013, Barley et al. 2017a,b). The Rowley Shoals (composed of the Mermaid, Clerke and Imperieuse Reefs) and the Scott Reefs (composed of Seringapatam, North and South Scott Reefs) are atoll-like reefs that are of similar size and share the same fish and benthic communities, habitat structure, coral cover, productivity, shelf position, reef size and temperature regimes (Ruppert et al. 2013, Barley et al. 2017a,b). The primary difference between these reef systems is that the Scott Reefs have been subjected to almost exclusive targeted shark fishing by Indonesian fishermen for centuries, whereas the Rowley Shoals is a marine reserve protected from nearly all forms of fishing (Russell & Vail 1988). As a result, shark populations at the Scott Reefs are now 4–17 times lower than at the Rowley Shoals (Ruppert et al. 2013) and are also composed of shark species that occupy comparatively lower trophic positions (Barley et al. 2017a). Previous research has revealed evidence of mesopredator release at the Scott Reefs due to these shark declines. Specifically, the abundances of prey fishes were higher on the reefs depleted of sharks (Ruppert et al. 2013), as were the body conditions of prey fishes, as measured by body weight, height and width for a given length (Barley et al. 2017b). These measures of condition were correlated with changes in diet that were argued to be based on behavioral adjustments due to reduction in predation risk (Barley et al. 2017b). Thus, the contrast between these 2 reef systems provides a large-scale (hundreds of kms, multiple reefs) comparative study approach for investigating the potential effects of predators and their removals on the biology and ecology of prey fishes in the wild (Barley & Meeuwig 2017).

Here, we sampled across the reefs of the Rowley Shoals and the Scott Reefs to test for potential changes in fish morphological traits as a consequence of targeted shark removals. Based on ecological theory (see 'Materials and methods'), we predicted that morphological traits associated with predator escape and detection performance in fishes would be affected by shark depletions. Specifically, we predicted that on reefs with intact shark populations, the caudal fins of teleost prey would be relatively larger in size to enable bursts of high speed swimming and rapid escape from predators, com-

pared to the same fish species from reefs where sharks have been depleted (Daniel et al. 1992, Rouleau et al. 2010, Oufiero et al. 2011). Additionally, we predicted that teleost prey eye sizes would be larger under high versus low risk of shark predation, since this should enable enhanced vigilance, early predator detection or the ability to better assess and appropriately respond to risk of attack given a predator encounter (Kiltie 2000, Brown & Kotler 2004, Cronin 2005). Larger eyes should also permit increased light sensitivity during twilight and night (Thomas et al. 2006, Land & Nilsson 2012), times when reef shark species are known to actively hunt (e.g. Papastamatiou et al. 2015). Accordingly, large eyes may be most important for reducing predation risk under low-light conditions. In addition to our central hypotheses, we also considered plausible alternative explanations (habitat complexity, temperature, light, current, food availability, prey targets, competition) as primary drivers of potential differences in fish caudal fin and eye sizes between the study reef systems.

We note upfront that in our investigation for potential system differences in the morphology of fishes, our study design cannot distinguish between morphological differences arising from either rapid evolution or plasticity due to shark removals, as this would require additional genetic and possibly experimental data (Irschick & Reznick 2009, Irschick & Higham 2016). However, we argue that any changes in the body shape of prey found arising from predator loss, whether driven by a plastic response, or through evolutionary change, is of interest, as both have ecological consequences and species can also display adaptive plasticity, for example (Denver et al. 1998).

MATERIALS AND METHODS

Study sites

The Rowley Shoals and the Scott Reefs are located on the edge of the continental shelf in north-western Australia, 300 km from the mainland (Fig. 1). Both reef systems have similar areas (~180 km²), physiochemical conditions, productivity levels and disturbance histories (Ruppert et al. 2013). Moreover, despite geographic separation among the 2 reef systems, monthly mean temperatures between the Rowley Shoals and Scott Reefs are not statistically different (Barley et al. 2017a). Similarly, recent habitat surveys have also revealed no significant differences in habitat complexity between the reef systems (Bar-

ley et al. 2017a). However, the distance between these reef systems (~400 km) appears to be large enough to prevent mixing of reef shark and fish populations (Field et al. 2011). While the Rowley Shoals has been a marine protected area since 1990 with both no-take zones and areas with minimal levels of charter fishing, targeted shark fishing at the Scott Reefs by Indonesian fishers has been occurring for centuries, a practice that has commercially intensified in recent decades due to the legalization of shark fishing in the Scott Reefs in 1974 (Russell & Vail 1988), coupled with the growing demand for shark fin in Asian markets (Worm et al. 2013). Accordingly, shark abundance at the Rowley Shoals is 4–17 times higher than at the Scott Reefs (Ruppert et al. 2013), whereas teleost populations are still relatively intact at the Scott Reefs as fishers primarily target teleosts for subsistence and also lack the capacity to transport large quantities of teleosts compared to shark fins (Nowara & Newman 2001).

Shark composition

Barley et al. (2017a) tested for differences in shark species diversity, abundance, length and biomass between the Rowley Shoals and Scott Reefs using stereo-baited remote underwater video stations (Stereo-BRUVS). The study revealed that sharks at the Rowley Shoals were almost twice as diverse, over twice as abundant, over 20% longer, and had almost 13 times as much biomass per BRUVS sample as compared to the Scott Reefs. In terms of species composition, 15% of the Rowley Shoals shark assemblage was composed of large-bodied, apex predators (tiger sharks *Galeocerdo cuvier*, scalloped and great hammerhead sharks *Sphyrna lewini*, *S. mokarran*), 81% were medium-bodied species, occupying relatively high trophic positions (silvertip *Carcharhinus albimarginatus*, grey reef *C. amblyrhynchus*), and only 4% were small-bodied species, occupying relatively lower trophic positions (zebra *Stegostoma fasciatum*, tawny nurse *Nebrius ferrugineus* and whitetip reef *Triaenodon obesus*). In contrast, only 5% of sharks observed at the Scott Reefs were large-bodied, apex predators, whereas the shark assemblage was primarily composed of medium-bodied, relatively higher trophic position species (55%) and small-bodied, lower trophic position species (40%). As a consequence, predation pressure in the form of both consumptive (i.e. shark-induced mortality) and non-consumptive effects (i.e. predation risk) likely differ between the reef systems. Although we cannot

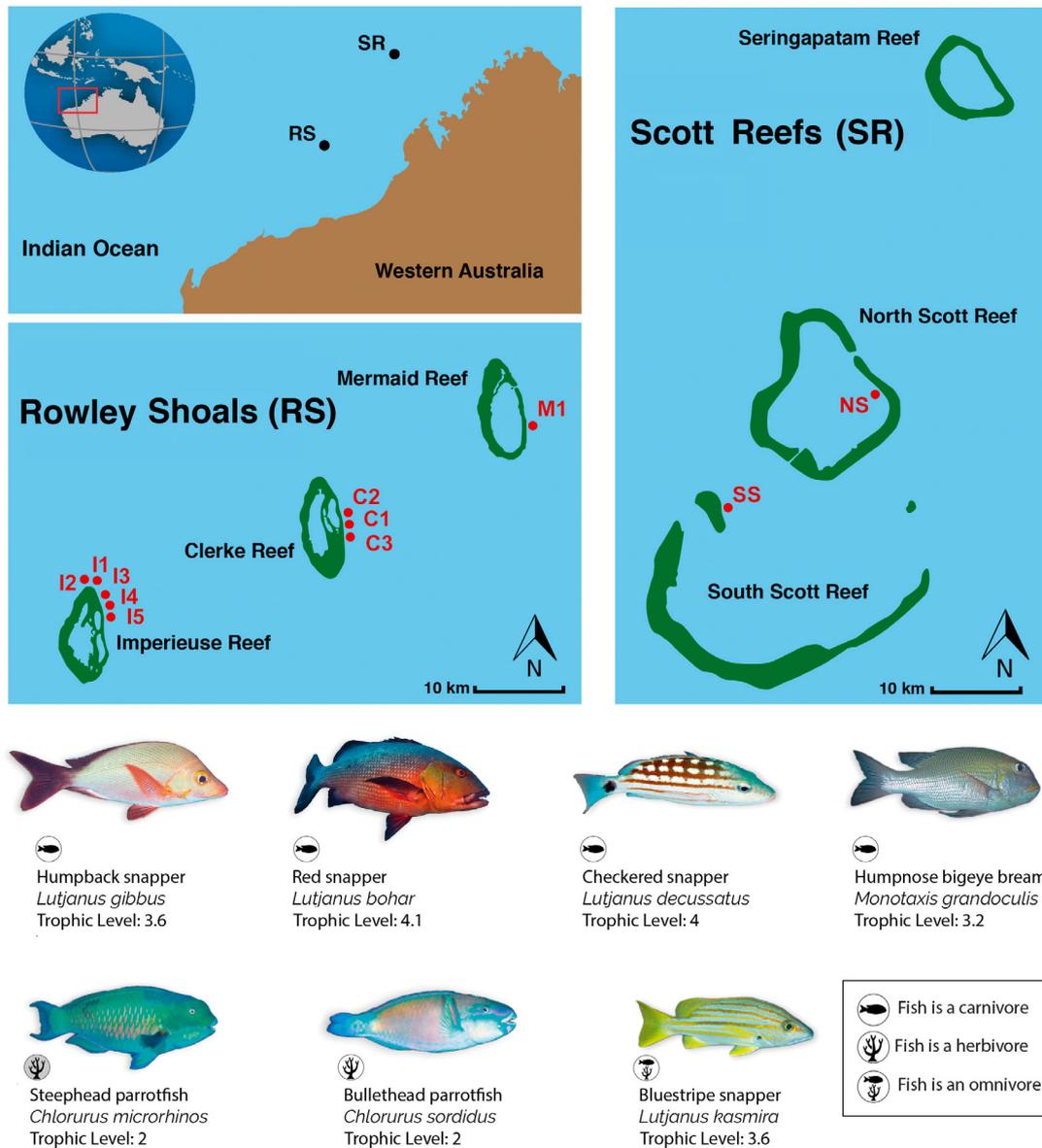


Fig. 1. Study species and location of the Rowley Shoals and the Scott Reefs in north-western Australia. Fish collection sites are indicated in red; corresponding positional information and associated sample sizes are provided in Table S1 in the Supplement at www.int-res.com/articles/suppl/m586p127_supp.pdf. Fish photos: Paddy Ryan; trophic levels from www.fishbase.org

tease apart the individual contribution of consumptive versus risk effects on the fishes, the significant differences in shark abundance and trophic composition between the reef systems are likely to influence both (Creel & Christianson 2008).

Fish morphological traits and ecological predictions

Active predatory fishes that consume prey in open water, such as some sharks, often ambush their prey over short distances. As successful predator avoid-

ance by prey typically involves bursts of high-speed swimming (Oufiero et al. 2011), morphological traits that minimize hydrodynamic drag and maximize thrust, such as caudal fin shape, are expected to be important for the process of escape performance (Rouleau et al. 2010). Propulsion in most fishes is primarily generated from the caudal fin. Larger fin spans for a given body length generate enhanced lift (Daniel et al. 1992), although excessively large caudal fins could result in reduced burst speeds in some species. Thus, we predicted that on the reefs with healthy shark populations (Rowley Shoals), the cau-

dal fin sizes of prey fishes would be relatively larger compared to conspecifics from the reefs where sharks have been depleted (Scott Reefs). We also predicted that aspects related to vision in teleost prey should be influenced by predation pressure. Foraging and vigilance are often mutually exclusive activities, and studies have shown that prey will mitigate risk by increasing levels of vigilance (Brown & Kotler 2004). In many fishes, vision is the primary sensory modality for early detections of predators, assessing predator intent and for making behavioral decisions appropriate to predation risk (Cronin 2005). Although not always the case, larger eyes generally have higher increased visual resolution and sensitivity to low light (Thomas et al. 2006, Land & Nilsson 2012). Accordingly, large eyes may be most important for reducing predation risk during twilight and night, i.e. times when some species of sharks hunt actively (reviewed by Hammerschlag et al. 2017). Thus, we predicted that eye size would be relatively larger for prey fishes on the reefs with healthy shark populations (Rowley Shoals) as compared to conspecifics from the reefs where sharks have been depleted (Scott Reefs).

Fish species and sampling

Fishes of 7 focal species were collected at the Scott Reefs in November 2012 and February 2014 and at the Rowley Shoals in April 2013 and November 2013. These species included *Lutjanus gibbus*, *L. decussatus*, *L. bohar*, *L. kasmira*, *Monotaxis grandoculis* and 2 species of parrotfish, *Chlorurus sordidus* and *C. microrhinos* (Fig. 1).

At the Rowley Shoals, fishes were collected from 9 sites across 3 reefs (Mermaid, Clerke and Imperieuse Reefs). At the Scott Reefs, fishes were collected from 2 sites across 2 reefs (North and South Scott Reefs). Between 34 and 52 individuals of each species were sampled from each reef system by free divers using spear guns (Fig. 1, Table 1; Table S1 in the Supplement at www.int-res.com/articles/suppl/m586p127_supp.pdf). Fishes were collected at sites and at depths of up to 20 m where complementary shark surveys had been conducted by Barley et al. (2017a). Collected fishes were stored in an ice slurry on tenders for

a maximum of 3 h, before being taken to the research vessel, the RV 'Solander,' for subsequent measuring and photo imaging. Photos were taken of the fish on a 1 m long measuring board and stored for later digital analysis of morphological features. Body length in mm (mouth to fork length) was recorded.

The lutjanid (*L. gibbus*, *L. decussatus*, *L. bohar*, *L. kasmira*) and lethrinid fishes (*M. grandoculis*) are mesopredatory (secondary consumer) species, feeding on a range of benthic invertebrates such as gastropods, worms, echinoderms and crustaceans and prey from the water column including smaller fishes and squid (Barley et al. 2017b). Lethrinids tend to prefer slow-moving prey, such as mollusks compared to lutjanids, and forage over a range of substrates, whereas lutjanids primarily forage on the reef (Kulbicki et al. 2005). Average trophic levels for these species are 3.6, 4.0, 4.1, 3.6 and 3.2 for *L. gibbus*, *L. decussatus*, *L. bohar*, *L. kasmira* and *M. grandoculis*, respectively (www.fishbase.org). The 2 parrotfishes (*C. sordidus* and *C. microrhinos*) are herbivorous (primary consumers), feeding primarily on endolithic algae found on the reefs, and both species occupy an average trophic level of 2.0 (www.fishbase.org). *C. sordidus* and *C. microrhinos* reach maximum lengths of 40 and 80 cm, respectively, while *L. gibbus*, *L. decussatus*, *L. kasmira*, *M. grandoculis* and *L. bohar* attain maximum lengths of 50, 35, 40, 60 and 90 cm, respectively.

Image analysis

ImageJ64 processing software was used to measure morphological variables from digital images of fishes (<http://rsbweb.nih.gov/ij/download.html>). Morphological features in the images were scaled using a metric ruler present in the photos. The following morphological measurements were made (Fig. S1 in the Supplement): (1) fork length (FL, horizontal length in cm

Table 1. Mean \pm SE and minimum and maximum values for fork length (cm) for focal fishes; N is the sample size of focal fish for each reef system

Species	Rowley Shoals (sharks present)				Scott Reefs (sharks depleted)			
	N	Mean \pm SE	Min	Max	N	Mean \pm SE	Min	Max
<i>Lutjanus kasmira</i>	35	20.61 \pm 0.25	15.95	23.32	52	19.71 \pm 0.20	14.53	22.53
<i>Chlorurus sordidus</i>	51	22.64 \pm 0.36	14.87	27.87	48	20.83 \pm 0.24	15.45	24.42
<i>L. decussatus</i>	48	23.22 \pm 0.32	17.90	27.47	41	21.01 \pm 0.38	14.82	28.43
<i>Monotaxis grandoculis</i>	47	25.59 \pm 1.02	17.77	43.16	51	25.61 \pm 1.02	16.99	42.52
<i>L. gibbus</i>	41	30.97 \pm 0.54	24.01	39.56	39	29.47 \pm 0.47	24.11	35.51
<i>C. microrhinos</i>	34	43.96 \pm 1.03	34.30	57.65	50	39.83 \pm 0.87	25.38	52.07
<i>L. bohar</i>	35	47.45 \pm 1.92	20.77	68.81	39	43.99 \pm 2.20	13.62	65.43

from the base of the mouth to fork of the tail), (2) caudal fin height (CFH, longest vertical distance in cm length from the highest to lowest point on caudal fin), (3) caudal fin length (CFL, length in cm from the center of the caudal fin base to the tip of the upper caudal lobe), (4) caudal fin area (CFA, total area in cm² encompassing the caudal fin), (5) eye diameter (ED, longest horizontal line through the center of the eye) and (6) eye area (EA, total area encompassing the eye). Measurements 1, 2 and 5 were recorded using the 'straight' function in ImageJ64 software, while measurements 4 and 6 were recorded by tracing the area using the 'freehand' function. After measurements were made, the data were plotted and visually inspected for outliers. Any outliers were re-measured for precision and accuracy.

Data analysis

For each of the morphological variables, we calculated the mean and standard error for each species within each reef system. Pearson's correlation was used to test for independence among response caudal fin and eye variables, respectively. We found that for each species, both CFL and CFH had a significant positive linear correlation with CFA (Table S2). Similarly, ED had a significant positive linear correlation with EA (Table S2). Accordingly, we focused subsequent statistical analysis only on caudal fin area (CFA) and eye area (EA) to maintain independence among response morphological variables being tested. CFA and EA are also the metrics directly related to overall eye and caudal fin size, which has been found to impact predator evasion/detection performance.

Fishes at the Scott Reefs are wider at a given length than conspecifics at the Rowley Shoals (Barley et al. 2017a), and FL is positively correlated with body width in the focal species (Table S2). Accordingly, to eliminate potential confounding effects of fish width on morphological traits, we normalized CFA and EA values by body width; hereafter, referred to as size-adjusted caudal fin area (sCFA) and eye area (sEA).

We calculated the percent relative difference (% ΔV) in mean sCFA and sEA at the Rowley Shoals (RS) compared to the Scott Reefs (SR) using the following equation:

$$\% \Delta V = [(V_{RS} - V_{SR}) / V_{SR}] \times 100 \quad (1)$$

where V is either sCFA or sEA.

Multivariate analysis of variance (MANOVA), with reef nested within each reef system (Rowley Shoals

vs. Scott Reefs), was used to test for significant differences in sCFA and sEA between systems and reefs, separately for each species. This multivariate approach allowed for testing potential system effects on both response morphological variables in combination and independently. Moreover, given that the Rowley Shoals and the Scott Reefs, as well as the individual reefs making up each system, differ spatially on a roughly latitudinal gradient, the nested approach also permitted testing for possible differences in fish morphologies from spatially segregated reefs within each individual system. Accordingly, this would provide insights as to whether unmeasured or unknown spatially differing variables could be influencing any morphological patterns found. Specifically, if spatial gradients in unmeasured variables were driving differences among systems, then tests for differences among reefs nested within systems may also be significant and in a consistent direction if and when differences among systems were found to be significant.

To investigate potential linear relationships between length and morphological traits, we regressed CFA and EA against FL for each species and system using linear models. We applied an analysis of covariance (ANCOVA) to evaluate for differences in slopes and intercepts between reef systems.

For all statistical analyses, values of morphological variables were log-transformed [$\log(\text{value} + 1)$] to conform to model assumptions and computed using SAS (SAS Institute) statistical software. Given multiple comparison, we applied a conservative alpha level of $\alpha = 0.01$ for assessing statistical significance.

RESULTS

A total of 611 fishes of 7 species were collected from the 3 reefs at the Rowley Shoals and the 2 reefs within the Scott Reefs and used in this morphological analysis. Species ranged in size (FL), with *Lutjanus kasmira* the smallest and *L. bohar* the largest (Table 1). For all 7 fish species, both absolute mean values (CFL, CFH, CFA, ED and EA) were higher on the Rowley Shoals compared to the Scott Reefs (Table S3 in the Supplement).

Mean values of sCFA for each species were 14 to 40% larger on the Rowley Shoals relative to the Scott Reefs (Figs. 2 & 3). Similarly, mean values of sEA for each species were 12 to 46% larger at the Rowley Shoals relative to the Scott Reefs (Figs. 2 & 3). MANOVAs revealed that the discriminate morphological variable was significantly higher at the Row-

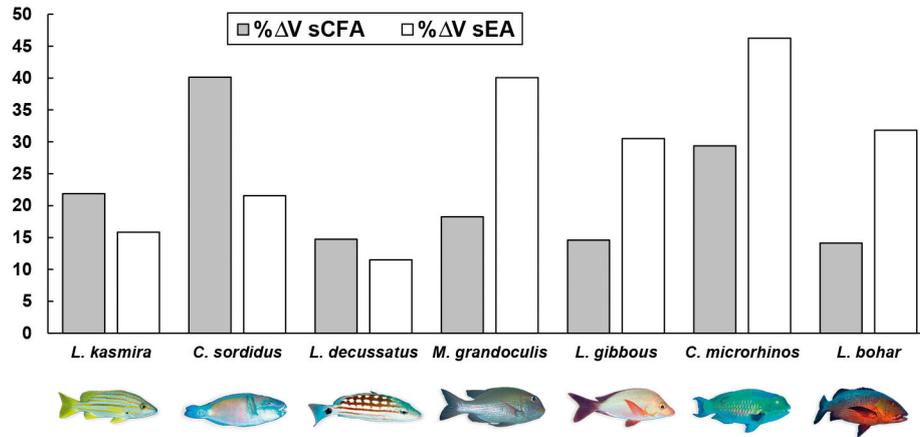


Fig. 2. Percent relative difference ($\% \Delta V$) in size-adjusted mean caudal fin area (sCFA) and eye area (sEA) at the Rowley Shoals compared to the Scott Reefs calculated using Eq. (1). Species are ordered left to right from smallest to largest mean length; full species names are given in Fig. 1

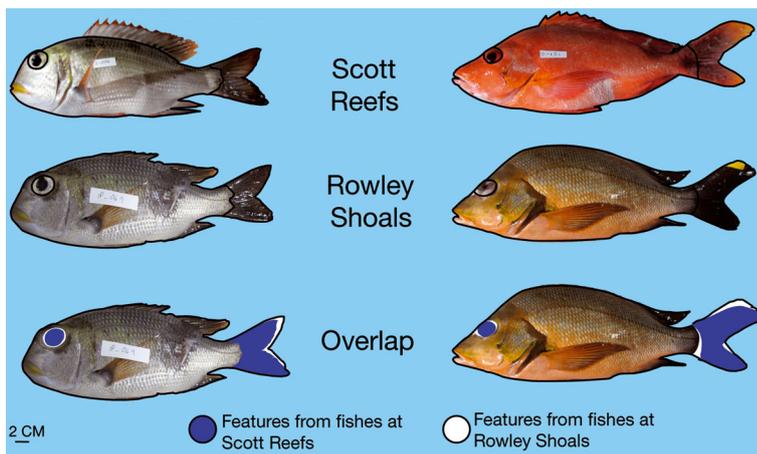


Fig. 3. Representative examples of morphological comparisons in eye area and caudal fin areas of *Monotaxis grandoculis* (left) and *Lutjanus gibbus* (right) of the same length collected from the Scott Reefs (SR, top row) and the Rowley Shoals (RS, middle row). The bottom row illustration shows the eye and caudal fin areas of the SR individual (dark blue) overlaid on the eye and caudal fin areas from the RS fish (white)

ley Shoals compared to the Scott Reefs for all 7 species (Table 2). The factor reef system contributed significantly to the variation in both sCFA and sEA for 4 of 7 species, sEA for 2 species and sCFA for 1 species (Table 2, Fig. 4). In these cases, 10–20% of the variation in sCFA and 7–31% of variation in sEA was accounted for by reef system (Table 2, Fig. 4). There were no differences in morphological variables among individual reefs nested within each system across species, with the exception of sEA for *L. kasmira* and *Monotaxis grandoculis* (Table 2).

Regression analysis revealed significant linear increases in both CFA and EA with increasing fish length within both reef systems, indicating allometric scaling (Table 3a, Fig. 5). ANCOVA of CFA and EA

versus FL revealed that the rates of change due to allometric scaling were comparable between the Rowley Shoals and the Scott Reefs, with the exception of CFA for *Chlorurus sordidus* (higher at the Scott Reefs) and *M. grandoculis* (higher at Rowley Shoals) and EA for *M. grandoculis*, *L. gibbus*, *C. microrrhinos* and *L. bohar*, where slopes were significantly higher at the Rowley Shoals than at the Scott Reefs (Table 3a). There were significant differences in the intercepts between reef systems for all 7 species, except EA versus FL for *L. gibbus* (Table 3b).

DISCUSSION

We took advantage of a unique study system to examine the potential indirect impacts of targeted shark removals on the morphology of 7 fishes across multiple coral reefs. While both the Rowley Shoals and Scott Reef systems have similar areas, biological communities, physiochemical conditions, productivity levels and disturbance histories, they differ significantly in the abundance and trophic composition of sharks. Ecological theory predicted that among coral reefs that have experienced nearly exclusive fishing of sharks (the Scott Reefs), prey fishes would have relatively smaller caudal fins and eyes compared to conspecifics on similar coral reefs with healthy shark populations (the Rowley Shoals). We found empirical support for these predictions. For example, mean sCFA of *Chlorurus sordidus* was 40% larger and mean sEA of *C. microrrhinos* was 46% larger on the Rowley Shoals relative

Table 2. Results of multivariate analysis of variance for evaluating differences in size-adjusted morphological traits between (a) reef systems and (b) individual reefs nested within each system, with test statistics for the discriminate response variable and size-adjusted caudal fin area (sCFA) and eye area (sEA). R² indicates the proportion of variation in morphological traits explained by the differences in reef system. Full species names are given in Table 1

(a) Reef system (Rowley Shoals vs. Scott Reefs)									
Species	Discriminate variable		sCFA			sEA			
	F	p	F	p	R ²	F	p	R ²	
<i>L. kasmira</i>	9.51	0.0002	19.2	<0.0001	0.19	6.39	0.01	0.07	
<i>C. sordidus</i>	13.22	<0.0001	24.31	<0.0001	0.20	7.61	<0.007	0.07	
<i>L. decussatus</i>	6.15	0.003	12.45	0.0007	0.13	3.42	0.07	0.04	
<i>M. grandoculis</i>	22.10	<0.0001	3.51	0.06	0.04	42.1	<0.0001	0.31	
<i>L. gibbus</i>	12.76	<0.0001	8.4	0.005	0.10	24.43	<0.0001	0.24	
<i>C. microrhinos</i>	17.7	<0.0001	13.88	0.0004	0.15	34.08	<0.0001	0.31	
<i>L. bohar</i>	8.63	<0.0004	3.52	0.065	0.05	17.49	<0.0001	0.20	

(b) Individual reef nested within system						
Species	Discriminate variable		sCFA		sEA	
	F	p	F	p	F	p
<i>L. kasmira</i>	4.57	0.002	2.76	0.07	9.21	0.0002
<i>C. sordidus</i>	2.95	0.6	0.85	0.36	3.67	0.06
<i>L. decussatus</i>	0.92	0.48	0.84	0.48	1.24	0.3
<i>M. grandoculis</i>	3.78	0.001	0.75	0.5	6.25	0.0007
<i>L. gibbus</i>	1.1	0.4	0.89	0.45	1.39	0.25
<i>C. microrhinos</i>	0.25	0.78	0.03	0.87	0.49	0.49
<i>L. bohar</i>	1.81	0.1	1.32	0.28	1.23	0.31

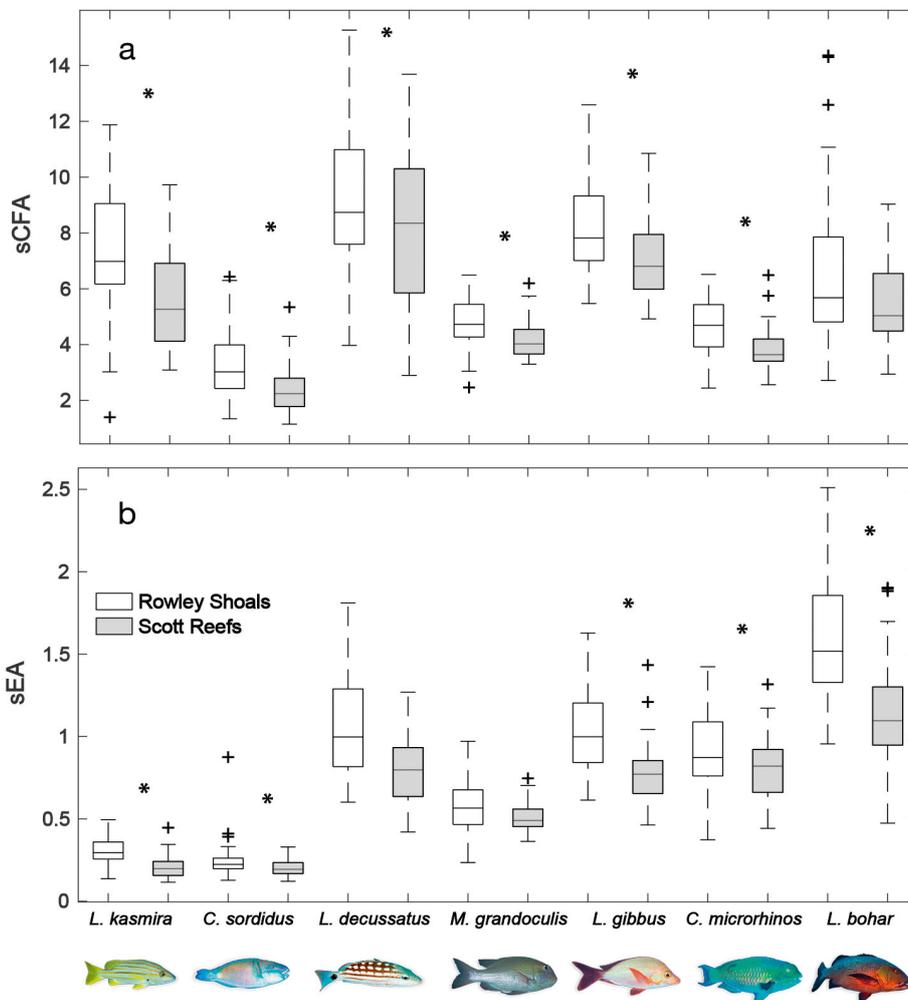


Fig. 4. Size-adjusted (a) caudal fin area (sCFA) and (b) eye area (sEA) for focal species at the Rowley Shoals (RS) compared to the Scott Reefs (SR). Species are ordered left to right from smallest to largest mean length; full species names are given in Fig. 1. Asterisks indicate significant differences between RS and SR based on multivariate analysis of variance (test statistics in Table 2). Box upper and lower edges are the interquartile range, the line within each box is the median, the whiskers represent the minimum and maximum values, and the + indicate outliers

Table 3. (a) Results of linear regressions of length against caudal fin area (CFA) and eye area (EA) for each system for the focal species and (b) results of ANCOVA tests for differences in slopes and intercepts between systems. In (a), asterisks next to slopes indicate p values associated with linear regression against length. In (b), asterisks associated with slope and intercept tests are based on ANCOVA. Analysis based on log (value+1)-transformed data. Significance level indicated as follows: – = non-significant, *p ≤ 0.01, **p ≤ 0.001, ***p ≤ 0.0001. Full species names are given in Table 1

Variable	Species	(a) Linear regression vs. length results				(b) ANCOVA results	
		Rowley Shoals (sharks present)		Scott Reefs (sharks depleted)		Slope test	Intercept test
		R ²	Slope	R ²	Slope		
CFA	<i>L. kasmira</i>	0.55	2.7***	0.53	2.0***	–	***
CFA	<i>C. sordidus</i>	0.32	1.72***	0.28	1.92***	**	**
CFA	<i>L. decussatus</i>	0.59	2.0***	0.64	1.32***	–	***
CFA	<i>M. grandoculis</i>	0.91	2.1***	0.89	1.69***	*	***
CFA	<i>L. gibbus</i>	0.68	1.99***	0.78	2.3***	–	***
CFA	<i>C. microrhinos</i>	0.46	1.95***	0.74	2.28***	–	***
CFA	<i>L. bohar</i>	0.90	2.0***	0.94	1.96***	–	***
EA	<i>L. kasmira</i>	0.48	2.23***	0.46	1.8***	–	***
EA	<i>C. sordidus</i>	0.16	0.52*	0.40	0.82***	–	**
EA	<i>L. decussatus</i>	0.33	1.36***	0.56	0.91***	–	***
EA	<i>M. grandoculis</i>	0.72	1.15***	0.48	0.87***	***	***
EA	<i>L. gibbus</i>	0.27	1.1**	0.15	0.72*	***	–
EA	<i>C. microrhinos</i>	0.38	0.96**	0.26	0.53***	***	**
EA	<i>L. bohar</i>	0.71	1.28***	0.70	0.9***	**	*

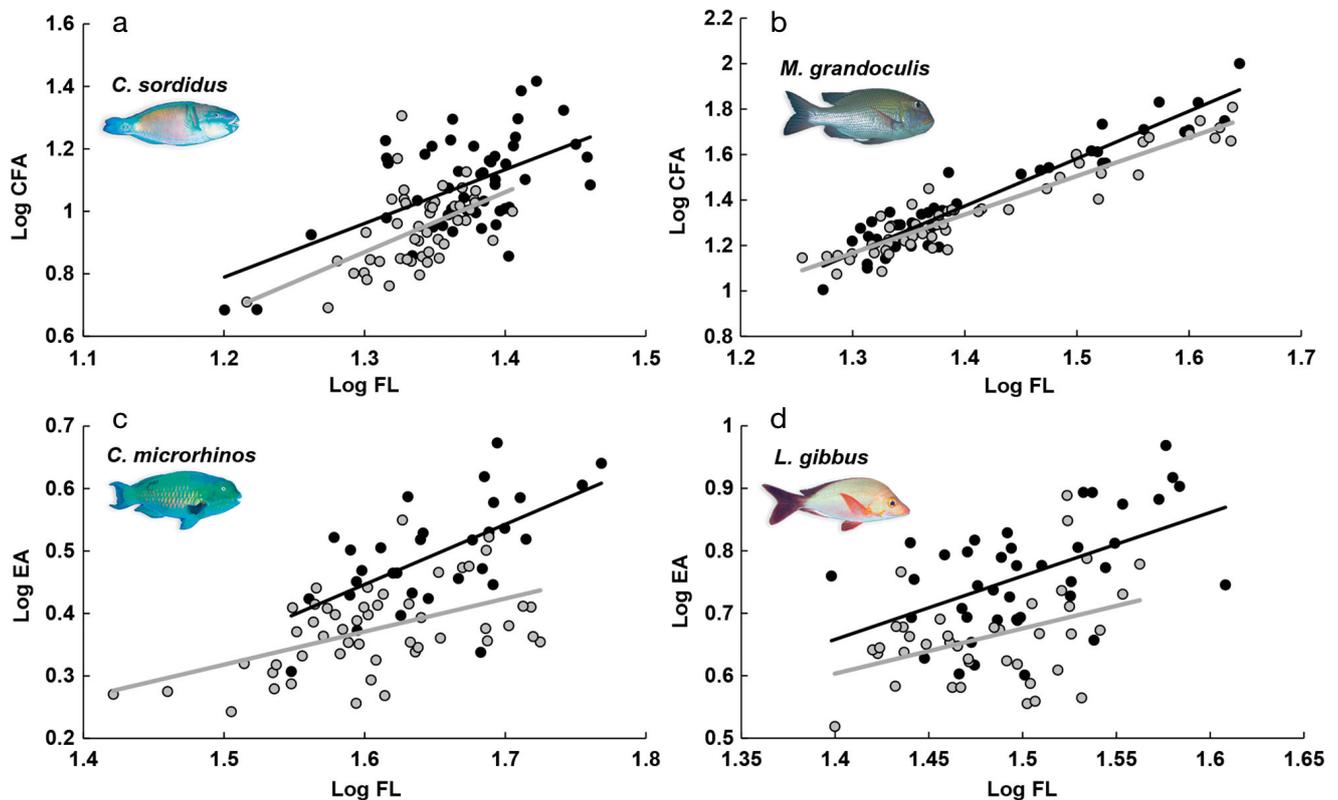


Fig. 5. Representative examples of relationship between fish morphologies (y-axis) and fork length (FL; x-axis) by reef system. Caudal fin area (CFA) in (a) *Chlorurus sordidus* and (b) *Monotaxis grandoculis*; eye area (EA) in (c) *C. microrhinos* and (d) *Lutjanus gibbus*. Lines are fitted linear regression models for the Rowley Shoals (black symbols) versus the Scott Reefs (gray symbols). Values (circles) are log(value+1)-transformed

to the Scott Reefs. Moreover, differences in fin and eye areas were consistent across the length range of fishes we sampled. However, it is important to note that in a correlative study such as ours, alternative explanations of these patterns cannot be fully discounted and may have contributed, at least in part, to the patterns we measured. However, such factors are unlikely to be the primary driver of the morphological patterns observed (see 'Alternative explanations' below).

Our study design cannot distinguish between the possibility of plasticity or genetic evolution via natural selection due to predator removals as the drivers of the morphological differences measured between systems. However, either option would be significant since it would represent, to our knowledge, the first quantitative evidence of a shift in prey morphology potentially resulting from the removal of predators by humans. Distinguishing between these mechanisms would require genetic data, and experimental studies under controlled laboratory conditions, for example, where fishes were raised with and without predators (Carroll et al. 2007). Such approaches are difficult to envision when the predators in questions are large reef-roaming sharks; however, studies of the genes that regulate body and eye shape and size could also offer insights into which of these possibilities are more likely (Irschick & Reznick 2009).

We hypothesize that the large differences in eye and fin morphology we measured between the study reefs were likely driven by the costs and benefits associated with expressing these traits under conditions of high and low shark predation. While larger eyes may help with predator detection, or assessing predator intent, especially under low-light conditions when sharks may be hunting in the reef, there are also costs associated with increasing eye size (Land & Nilsson 2012). These include energetic costs of physically moving with larger eyes, the costs of manufacturing and maintaining millions of nerve cells and the increased risk of eye damage and greater neural processing (Thomas et al. 2006). Larger eyes also require more brain space for image processing than smaller eyes (Møller & Erritzøe 2014). Changes in eye size are thus likely a reflection of the costs and benefits of increased visual acuity (Thomas et al. 2006). With respect to the caudal fin shape, larger fins will enhance movement performance, and thus may aid in predator escape, but as with eyes, there are potential costs associated with larger fins. These costs include the increased energy required for growth and maintenance of a large caudal fin and associated structures (e.g. caudal peduncle) and the energetics of physically carrying larger fins (e.g. additional drag).

The morphological patterns that we found were relatively consistent across all 7 focal species, despite these fishes differing in size and trophic guild. We hypothesize that this may be a result of sharks feeding across trophic levels (Barley et al. 2017a), so that a reduction in shark numbers has possibly caused morphological changes in prey across trophic levels. Some recent studies have revealed that smaller reef sharks occupy trophic levels comparable to large teleosts (e.g. Frisch et al. 2016). Accordingly, it is likely that predation pressure on the larger teleosts at the Rowley Shoals is from the large-bodied apex predatory sharks found there, which are almost absent from the Scott Reefs. However, it is also possible that reef sharks may be consuming the juvenile stages of these larger teleosts, whereby larger eyes and tails would improve survivorship. If larger eyes and tails are not a plastic trait throughout a lifetime, the differences in morphologies we recorded could also be a carry-over effect displayed in adults of larger species that survived a juvenile stage vulnerable to reef shark predation. It is worth noting that our study cannot determine whether the system differences in fish morphology attributed to shark presence/absence arose through differences in consumptive or non-consumptive predator effects (i.e. predator-induced mortality versus risk effects), but the significant differences in shark abundance between the reef systems are likely to influence both (Creel & Christianson 2008).

Alternative explanations

Large-scale, unreplicated natural experiments have a unique power to test hypotheses at ecologically realistic scales and have delivered insights of great power into cosmology, evolution and geology (Barley & Meeuwig 2017). However, as with any study of this type, we cannot exclude the possibility that some unmeasured factor(s) other than predation could be driving the patterns we observed. Such variables include temperature, habitat complexity, light levels, current speed, food availability/quality, prey capture requirements and competition for resources. However, we found little evidence that this was likely the case, discussed below. In terms of environmental conditions, average temperatures recorded over the past 5 yr at the Rowley Shoals and Scott Reefs did not differ significantly between systems (Barley et al. 2017b). While habitat differences could also possibly impact morphology, habitat complexity does not differ between the Rowley Shoals and the Scott Reefs (Barley et al. 2017a). Experimen-

tal work has shown that juvenile brook trout *Salvelinus fontinalis* reared in high-velocity streams had maximum caudal fin heights that were 7.8% larger on average than those of fish reared in streams where flows were an order of magnitude lower (Imre et al. 2002). Similarly, cichlids (*Astatotilapia burtoni*) reared in total darkness had eye diameters that were only 2.5% larger on average than individuals reared under conditions of full white light (Kröger & Fernald 1994). We did not measure current speeds or light levels between reef systems, but both the Rowley and Scott reef systems occupy similar oceanographic settings and positions at the edge of the continental shelf, have the same habitat complexity (Ruppert et al. 2013) and are also subject to similar light and tidal regimes. Thus, it seems unlikely that these factors could account for the morphological differences we observed.

Currens et al. (1989) found that food quantity can affect the morphology of fishes, with poorly-fed fishes having longer upper jaws and smaller body depths. However, they also demonstrated that caudal fin areas were unaffected by changes in feeding regime, which suggests that our results were not influenced by differences in food availability between the 2 study systems. Indeed, even if caudal fin area and other body dimensions were influenced by food availability, fishes at the Scott Reefs should theoretically have had larger eye and caudal fin dimensions than at the Rowley Shoals, as Barley et al. (2017a) found that prey of lower trophic levels that would be consumed by the focal fishes were significantly more abundant at the former location than the latter. However, we found the opposite pattern, further suggesting that food availability is likely not the primary driver of the morphological patterns found. Alternatively, the observed patterns might have been due to differences in the food targets between reef systems. Fishes have been shown to develop larger caudal fin areas in response to an increased need to pursue mobile prey (Webb 1982). Again, based on the earlier studies at the study reefs, this is unlikely to explain our results, since mesopredatory teleosts at the Scott Reefs, where eye and caudal fin areas are smaller, consume more mobile fishes and fewer benthic invertebrates than conspecifics at the Rowley Shoals (Barley et al. 2017b). Moreover, our finding that the direction of morphological differences between reef systems was consistent across different fish trophic levels, regardless of diet type (herbivore, piscivore, omnivore) is also difficult to reconcile with explanations solely based on diet.

Given the higher measured abundances of mesopredatory fishes at the Scott Reefs attributed to predation release (Ruppert et al. 2013), it is also possible that competition for space and food in some way impacts fin and eye morphology. Resource competition often drives divergence, favoring increased ecological and phenotypic variation (Smith 1962, Maret & Collins 1997, Bolnick 2004). Accordingly, an increase in competition due to higher fish abundances should result in more individuals with extreme morphologies, enabling them to access alternative resources (Maret & Collins 1997, Swanson et al. 2003). However, our results show the opposite pattern. Across all 7 focal species, variance in all the morphological traits examined were higher at Rowley Shoals, where focal fish abundances were lower (Table S3, Fig. 4). On the other hand, predation is known to cause shifts in both the density and frequency of prey phenotypes, potentially leading to increased phenotypic variation (Langerhans et al. 2004, Eklöv & Svanbäck 2006, Ingley et al. 2014). Data from the current study were consistent with this hypothesis, since there was greater morphological variability at the Rowley Shoals than at the Scott Reefs.

While it is possible that some spatial gradient in unmeasured variables could influence morphological patterns, we found no evidence of this in our analysis of patterns in morphology across individual reefs within each system. Thus, while other variables could have contributed, at least in part, to the differences in morphological patterns found between reef systems, they are unlikely to be the primary source of variation. Rather, our results are consistent with predictions based on predator-prey ecological theory.

CONCLUSION

In summary, we provide evidence from a large-scale field study that depletions of predators by humans may cause changes to morphological traits of prey. This study contributes empirical evidence of the potential ecological and evolutionary consequences of shark population depletions, further suggesting the need for timely conservation of these top predators. If the measured differences in morphological traits between reef systems were driven by the associated energetic trade-offs of expressing them under varying levels of shark predation, then fishes released from predation pressure may shift energetic resources from somatic growth to reproduction, which could improve their fecundity and reproductive suc-

cess, ultimately leading to shifts in demography, population dynamics and trophic relationships. Indeed, mesopredatory fishes at the shark-depleted Scott Reefs are relatively more abundant (Ruppert et al. 2013, Barley et al. 2017a) and in better condition than conspecifics at the Rowley Shoals (i.e. wider at a given length), likely as a result of changes in foraging behavior (Barley et al. 2017b). Thus, future research on trophic cascades, including work in our study system, should consider how human-driven predator declines and subsequent alterations in prey behavior, physiology and/or morphology could be affecting energy flow throughout the ecosystem, ultimately impacting trophic relationships and population dynamics.

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