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Stress, predators, and survival: Exploring permit (*Trachinotus falcatus*) catch-and-release fishing mortality in the Florida Keys

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ABSTRACT

The biological consequences of catch-and-release (C&R) angling revolve around interactions between the physiological and behavioural responses of the captured fish and ecological conditions such as the presence of opportunistic predators. Here, we explored the consequences of C&R on adult permit (*Trachinotus falcatus*), including assessments of depredation rates, their physiological and reflex responses prior to release, post-release behaviour, and post-release predation in diverse habitats in the Florida Keys, USA. We found pre-capture depredation rates were highly variable amongst habitat types, ranging from zero on shallow water flats, to 35.3% and 90.1% on specific reef and shipwreck locations, respectively. Observed predators were all large sharks. Importantly, one of the high predation sites is an important permit spawning location, thus C&R fishing in that locale may be a conservation concern. Physiological stress responses (blood lactate, glucose, pH) and reflex tests indicated that permit were relatively robust to routine angling (fight durations of 1 to 12 min) and handling (air exposure up to 2 min). Short duration post-release tracking using tri-axial acceleration biologgers identified no differences in swimming activity for fish that were kept in water versus those held in the air for 2 min to simulate an admiration period. While this study indicates that permit are relatively robust to C&R angling in terms of stress responses and behavioural impairment, high densities of opportunistic predators at certain fishing locations can result in high rates of pre-capture depredation independent of the state of the animal. Permit angling in locations with high predator densities is a potential conservation issue, especially if specific locations represent important pre-spawning aggregation sites for fish populations that may be more vulnerable to predation and thus depredation. Current C&R best practices (i.e., limiting fight times and air exposure) may not be adequate to ensure permit survival at high predator density sites. Angling-related depredation is often cryptic, yet is a growing conservation concern in many fisheries – we developed and applied a novel framework for identifying cryptic depredation that may be applicable across fisheries.

1. Introduction

Recreational angling is a globally popular activity with participants exhibiting an increasing interest in ensuring its sustainability (Coleman et al., 2004; Cooke and Cowx, 2004). Although angled fish are sometimes harvested for food (Cooke et al., 2018), catch-and-release (C&R) angling has become common conservation practice in many established recreational fisheries (Arlinghaus et al., 2007). Reasons for anglers to engage in C&R are varied, but most commonly it is to comply with harvest regulations (e.g., releasing undersized fish, releasing fish that out of season) or a result of angler conservation ethic (Arlinghaus et al., 2007). The premise behind C&R is that there is high survival and negligible fitness consequences for released fish (Cooke and Schramm, 2007). Yet, it is widely known that the condition of fish upon release is highly dependent on species-specific physiology, angler behaviour, interactions with environmental factors (e.g., water temperature, salinity) and ecological conditions (e.g., predators, refuge) (Cooke and Suski, 2005; Arlinghaus et al., 2007). Studies also show that fish physiological stress arising from a fisheries interaction can lead to a number of behavioural alterations including impairment of reflexes (Davis, 2010;

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Brownscombe et al., 2015), decreased swimming performance (Brownscombe et al., 2015), and a compromised ability to seek refuge (Brownscombe et al., 2014; Cooke et al., 2013a), all of which can lead to post-release predation (Danylchuk et al., 2007; Donaldson et al., 2008; Guindon, 2011). Therefore, identifying best practices that ensure fish are in optimal condition before release is critical, particularly for fish that live in regions with high densities of predators (Raby et al., 2014).

Permit (*Trachinotus falcatus*) are a widely distributed marine fish that inhabit coastal areas including expansive seagrass flats, nearshore reefs and offshore structure in the Western Atlantic, Gulf of Mexico, and Caribbean Sea (Adams et al., 2006). The permit fishery in the southern United States represents a significant economic contribution to recreational fisheries, particularly in Florida where the annual economic impact of the flats fishery alone exceeds $465 million dollars (Fedler, 2013). This is especially important in the Florida Keys ecosystem which supports a wide range of habitats where permit can be angled using a variety of recreational fishing techniques. In the Florida Keys, permit are often targeted on shallow, nearshore flats: a unique blend of seagrass, mangroves, sand, mud, benthic algae and coral rubble (Adams and Cooke, 2015). These flats serve as important habitats for resident and transient communities of fauna (Barbier et al., 2011), and are particularly significant to the growth and development of popular gamefish including permit, bonefish (*Albula vulpes*) and tarpon (*Megalops atlanticus*). As such, C&R is a practice that is commonly applied to the flats fishery of the Florida Keys.

In contrast to the flats, offshore habitats, like reefs, shipwrecks and towers (navigation and/or decommissioned radio towers), represent target areas for anglers practicing both voluntary C&R and harvest-based angling. Recreational fishing for permit is common on nearshore and offshore habitats in the Florida Keys, where large schools of permit aggregate at spawning or pre-spawning sites. Telemetry data show spawning permit aggregating at known locations in the Florida Keys as early in the year as March (Brownscombe et al., 2019). In addition to attracting large schools of permit, some of these locations also attract fish communities that vary in taxonomic levels (Koenig et al., 2000), including apex and mesopredators such as sharks and groupers. Anecdotal evidence suggests that boat activity at these locations can attract sharks before angling begins at sites where permit spawning aggregations occur (J. Brownscombe, personal observation), and sharks are known to change their behaviour given the presence or absence of vessels (Fitzpatrick et al., 2011). Consequently, there are reports of sharks depredating hooked permit from the end of fishing lines at specific offshore locations in the Florida Keys (J. Brownscombe, personal observation). Though the flats and offshore fisheries may differ with respect to the applied angling techniques and the equipment used, the connectivity shared between populations of fish from both locations must be considered. If fish that are captured on the flats come from offshore spawning aggregations, it is important to understand how fish lost from one location can impact their population, especially if specific spawning locations are more susceptible to depredation events.

Depredation occurs when hooked fish are removed or killed from fishing lines and/or gear by predators prior to landing (Raby et al., 2014). Though depredation has been studied in other taxa (Knowlton et al., 1999; Naughton-Treves et al., 2003), with several examples coming from the marine environment (Garrison, 2007; Powell and Wells, 2011; Hamer et al., 2012; Mitchell et al., 2018a), little is known about its impact with respect to recreational C&R angling. Depredation is not unique to the Florida Keys (O'Toole et al., 2010; Mitchell et al., 2018a, 2018b), however, the incidence of permit depredation on offshore structures represents a potential threat to their conservation. Management and regulation of recreational permit angling is difficult when spawning aggregations form on offshore structures, and understanding the potential impact of depredation on permit populations begins with identifying its incidence. Research supporting management of permit fisheries is limited (Crabtree et al., 2002; Adams et al., 2006) as the study of their physiology, behaviour, and spatial ecology is in its infancy. Moreover, no C&R science studies on this species currently exist. This lack of knowledge specific to the permit fishery in the Florida Keys, combined with potential impact of depredation on the population, presents a complex challenge to fisheries managers tasked with identifying appropriate C&R practices and instituting other conservation action. To address this, we examined the effects of C&R angling on permit depredation rates, physiological stress, reflex impairment responses, post-release behaviour and survival of permit in diverse habitats in proximity to the Florida Keys. We aimed to identify potential conservation issues associated with rate of depredation in addition to identifying the best angling practices for permit using observational and experimental approaches.

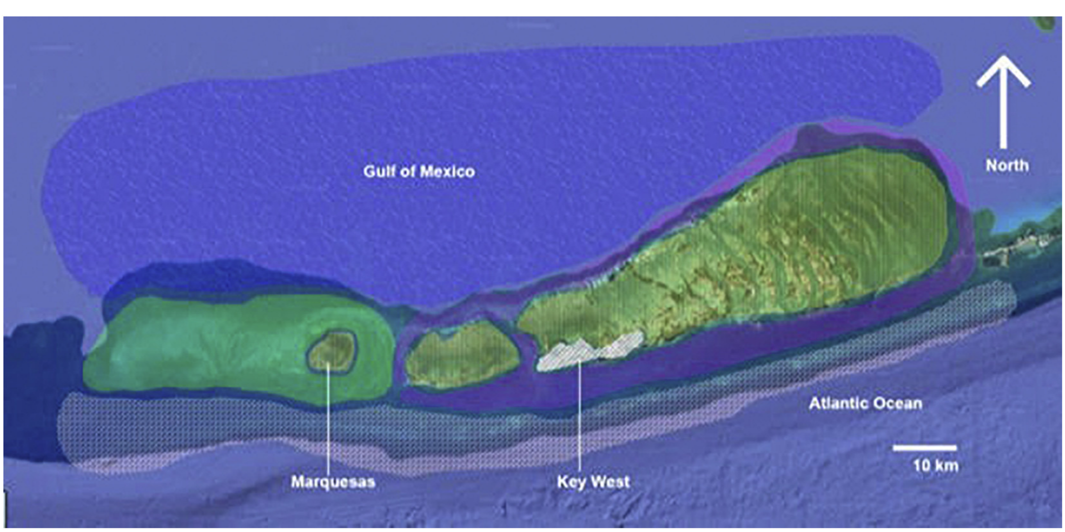

Fig. 1. Habitat distribution for the Western Florida Keys. The white (lined) area is the population centre of Key West. Yellow areas indicate flats habitats. Purple habitats indicate nearshore structures. White (hashed) indicates Atlantic structures. Green indicates western structures. Blue indicates Gulf (of Mexico) structures. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

2. Methods

2.1. Depredation

Permit were angled from 33 general locations that were classified into 5 distinct habitats sharing similar characteristics that included: flats, nearshore structures, Atlantic structures, Gulf structures and western structures (Fig. 1). Flats were defined as expansive seagrass flats, found near shore that ranged from 0.5 to 2 m in depth. Nearshore structures were defined as either shipwrecks, towers or reefs found within 8 miles of shore (Fig. 1). Atlantic structures included shipwrecks and coral reefs located on the Atlantic (southern) side of the Florida Keys, Gulf structures included offshore shipwrecks and a decommissioned Navy radio tower in the Gulf of Mexico (northern) side of the Florida Keys, and western structures included shipwrecks directly west of the Marquesas at the interface of the Atlantic Ocean and the Gulf of Mexico (Fig. 1).

Permit angling events were documented from March of 2016 to June 2018, and included catches by researchers, fishing guides, and recreational anglers on guided trips. Angling was conducted with a range of fishing gear, from medium strength fishing rods with 6.8 kg break strength line to heavy strength rods with 22.7 kg break strength line. Medium strength setups were utilized on the flats (enabling farther casting distances for sight fishing and stealthier line), and heavy strength setups on deeper-water structures. All fishing setups included braided fishing line attached to ~1.5 m of fluorocarbon leader which ranged from 6.8 kg to 22.7 break strength. Fish were often angled using small crabs (carapace width of approximately 4–5 cm) as live bait at the end of small octopus hooks (2/0) or weighted jigs (1/2 oz to 1 oz).

Incidences of confirmed permit depredation (n = 12, Table 2) were documented via visual observations made by anglers on the boat when they occurred near the water surface. These incidences were used to develop a specific set of criteria (Table 1) to assess the probability that permit lost prior to landing resulted from depredation or to other causes (i.e., line breaking on structures or the fish coming loose from the hook) (Fig. 2). This was based on the series of events that occurred in observed depredation events. In all cases when predation was visually observed, permit exhibited a marked transition from a low energy resistance response (i.e., slow swimming to resist capture) to rapid burst swimming (resulting in pulling line off the fishing reel through the drag) as the predator(s) approached to mount an attack. This was followed by a period of erratic evasive swimming behaviour prior to the predator capturing the permit (Fig. 3B), which, in every observed instance, resulted in a broken fishing line at the terminal end of the leader. In instances where permit were hooked but not landed and depredation was not visually observed, depredation was considered probable when all events described above occurred (Fig. 2). In these cases, it was also common for anglers to visually observe previous depredation events, or to visually observe predators in the vicinity of the fishing vessel. Depredation was considered improbable when no major changes in fish behaviour were observed in concert with fish loss, either through the line breaking (likely due to underwater structures or application of too much drag resistance on the fishing reel) or the fish becoming unhooked. A visualization of the intensity and duration of the fight of a hooked fish, combined with the potential factors contributing to depredation, was created to aid in describing potential depredation events (Fig. 3). This figure is meant strictly for descriptive purposes, and is based on the collective descriptions of catches where fish were landed successfully (Fig. 3A), depredation was confirmed (Fig. 3B), depredation was probable (Fig. 3B) or depredation was improbable (Fig. 3C).

2.2. Physiological and behavioural responses to C&R

Permit were angled from offshore shipwrecks around the Florida Keys between April 7 of 2018 and June 8 of 2018 and were also included in the data set listed in the above section (2.1). Fish were captured from offshore shipwreck habitats because researchers had access to greater numbers of fish compared to flats habitats. These fish were captured using conventional spinning tackle (medium-heavy rods and 22 kg breaking-strength braided line, 18 kg breaking-strength fluorocarbon leader material, 2/0 octopus hooks) and small live crabs (carapace width of approximately 4–5 cm). Following capture, permit were handled either completely in the water (0 min of air exposure; 0MIN) or exposed to the air for 2 min (2 min of air exposure; 2MIN), simulating a fisheries interaction that may include hook removal out of water and time for photographs to be taken (Brownscombe et al., 2015; Prystay et al., 2017; Suski et al., 2007).

Immediately following air exposure (0MIN or 2MIN), fish were assessed using reflex action mortality predictors (RAMP) that include equilibrium, tail grab, eye roll (vestibular ocular response, VOR), body flex and head complex assessments (Davis, 2010; Brownscombe et al., 2017). Reflex impairment is a common predictor of fish behavioural impairment and post-release survival (Davis, 2010; Raby et al., 2012), and is therefore a useful tool to assess the condition of fish in the context of recreational angling (Brownscombe et al., 2017). Equilibrium was assessed by turning the fish upside-down, with the ability for the fish to right itself within 3 s indicating a positive response. Tail grab was assessed by grabbing the fish by the tail, with fish attempting to escape handling indicating a positive response. VOR was assessed by rolling the fish on its side and observing the eyeball movement of the fish, with eyeballs tracking level indicating a positive response. Body

Table 1
Criteria for determining the possible occurrence of a depredation event.

Outcome	Observations (Criteria)
Confirmed predation	• Fish transitioned from low energy resistance response to high energy escape response with erratic swimming behavior • Predator visually observed depredating hooked permit • Fishing line cut, hook lost • Predators observed in the area
Probable predation	• Fish suddenly transitioned from low energy resistance response to high energy escape response with erratic swimming behavior • Hooked permit were not lost in proximity to any underwater structure • Fishing line cut, hook lost • Predators observed in the area • Other permit depredated during fishing session
Improbable predation	• Fish exhibits regular transition from high to low energy escape response • Fishing line breaks or fish becomes unhooked • No predators observed in area • No depredation events observed on fishing day • Close proximity to submerged structure (eg. Shipwrecks or reefs)

Criteria used to evaluate likelihood of depredation events. Combinations of multiple factors is positively proportional to likelihood of depredation, where depredation is more likely to happen if more factors are present during a fisheries interaction.

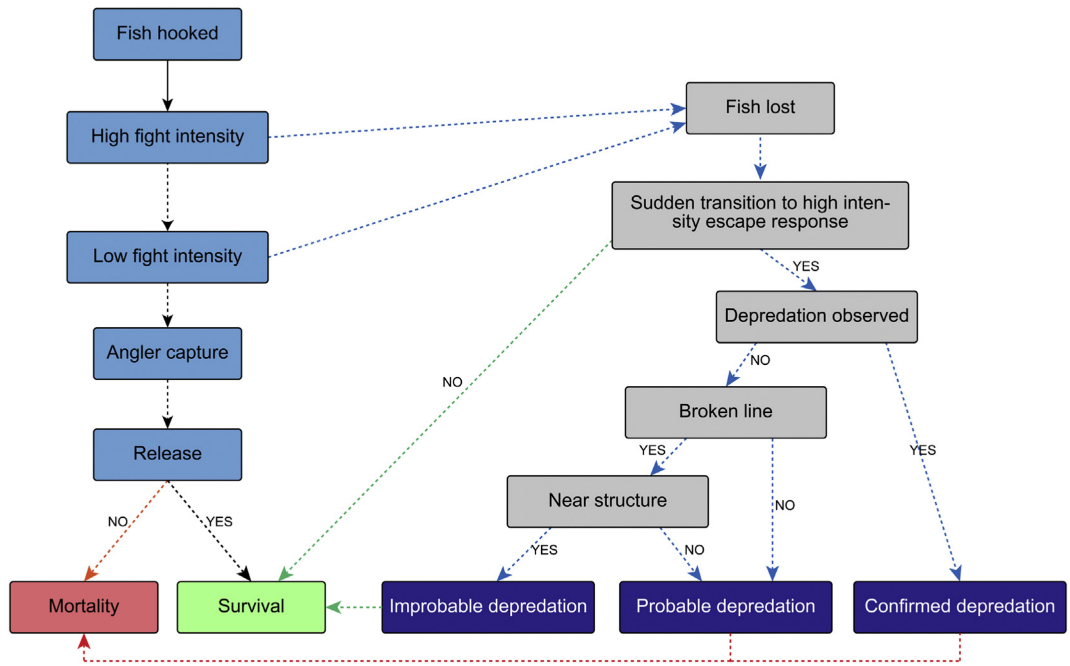

Fig. 2. Flow chart describing observable criteria that contribute to depredation events in permit.

flex was assessed by lifting the fish from the centre of its body, with body flexion as an attempt to escape indicating a positive response. Head complex was assessed by observing opercular movement, with regular ventilation indicating a positive response. Positive responses were scored as 1, negative responses (absent reflexes) were scored as 0. Previous literature indicates these tests are viable measures of fish vitality, and often predictive of post-release behavioural impairment and/or mortality (Raby et al., 2012; Brownscombe et al., 2013, 2015).

Once reflexes were assessed, permit were held in a 106l livewell with a constant flow of fresh seawater while blood samples were taken from caudal vasculature punctures immediately (0 min) and 30 min post-capture. Although peak physiological stress varies between species, samples taken at 30 min were used to examine near-peak physiological stress responses in permit based on practices in other studies (Flodmark et al., 2002; Bracewell et al., 2004; Suski et al., 2007; Cooke et al., 2013b). Approximately 1–2 ml of blood was taken with each sample using 38 mm 21-gauge needles (Catalog no. 305917, Becton and Dickinson and Company, Franklin Lakes, NJ, USA) with 4 ml heparinized vaccutainers (Catalog no. 367884, Becton and Dickinson and Company, Franklin Lakes, NJ, USA). Blood was immediately analyzed for glucose (mmol/l, Accu Check Compact Plus, Roche Diagnostics, Basel, Switzerland), lactate (mmol/l, Lactate Plus, Nova Biomedical Corporation, Waltham, MA, USA) and pH (HI-99161 with automated temperature compensation, Hanna Instruments, Woonsocket, Rhode Island, USA) using previously validated point-of-care devices (Stoot et al., 2014).

2.3. Post-release behaviour and survival

Tri-axial accelerometers (Gulf Coast Data Concepts X16-mini, Waveland, Mississippi, USA) are useful for evaluating the swimming behaviour and survival of fish (Cooke et al., 2016) including in the context of C&R studies (Brownscombe et al., 2018; Lennox et al., 2018). For this study, an additional group of permit (n = 8; separate from the group used for physiological and behavioural responses to C&R; section 2.2) were captured using hook and line from offshore wreck habitats.

Immediately after capture, fish were either held in the water for the entire duration of the experiment or subjected to 2 min of air exposure in order to simulate a fisheries interaction. Following the exposure to either 0 min (n = 3) or 2 min (n = 5) of air, tri-axial accelerometers were attached to the caudal peduncle of the fish with a custom designed acrylic plate which had bungee cords and attachment points for elastic bands (Lennox et al., 2018). A large snap swivel was attached to the acrylic plate, which was used as an anchor point for 22 kg breaking strength braided fishing line (0.36 mm diameter) attached to a spin fishing rod. The spool of fishing line was left open during the recording period, allowing fish free swimming movement without tension. Once the recording period was complete (20 min), the bail was closed, and the line was pulled taught until the elastic bands broke to retrieve the accelerometer package. The use of this technique also allowed for quantifying potential post-release predation if fish exhibited erratic, burst swimming behaviour, if swimming ceased or if accelerometer packages were broken off prematurely when fish were not in proximity to any underwater structures. Absolute displacement from the original site of capture to the end of the 20 min recording period was not measured as ocean currents moved the free-spooled line between the accelerometer package fixed to the fish and the spool from which it was fixed, making it difficult to determine the absolute position of the fish.

2.4. Data analysis

All data analysis was conducted using R (R Team, 2016) via RStudio (R Team, 2016). Depredation data presented analytical challenges because some habitat types had no predation values and had hence zero variance. Therefore, to explore the influence of habitat type on permit depredation rates, conditional inference trees (CIT) were used, which identify significant binary recursive partitions in the data to predict the response (Hothorn et al., 2006). CITs were fitted to 1) known predation events (observed depredation = 1, other outcomes = 0), and 2) probable predation events (observed and probable depredation = 1, improbable depredation and known survival = 0). Each model included habitat type (flats, nearshore structures, western structures, Gulf structures, Atlantic structures) as the predictor. In this case the algorithm identified significant partitions in the data with t-tests. To examine the impacts of various factors on permit stress responses, blood lactate, glucose and pH were compared using linear mixed effects models ('nlme' package, RStudio) containing fight time, temperature, air exposure, collection time (ie. The effect of extracting blood samples

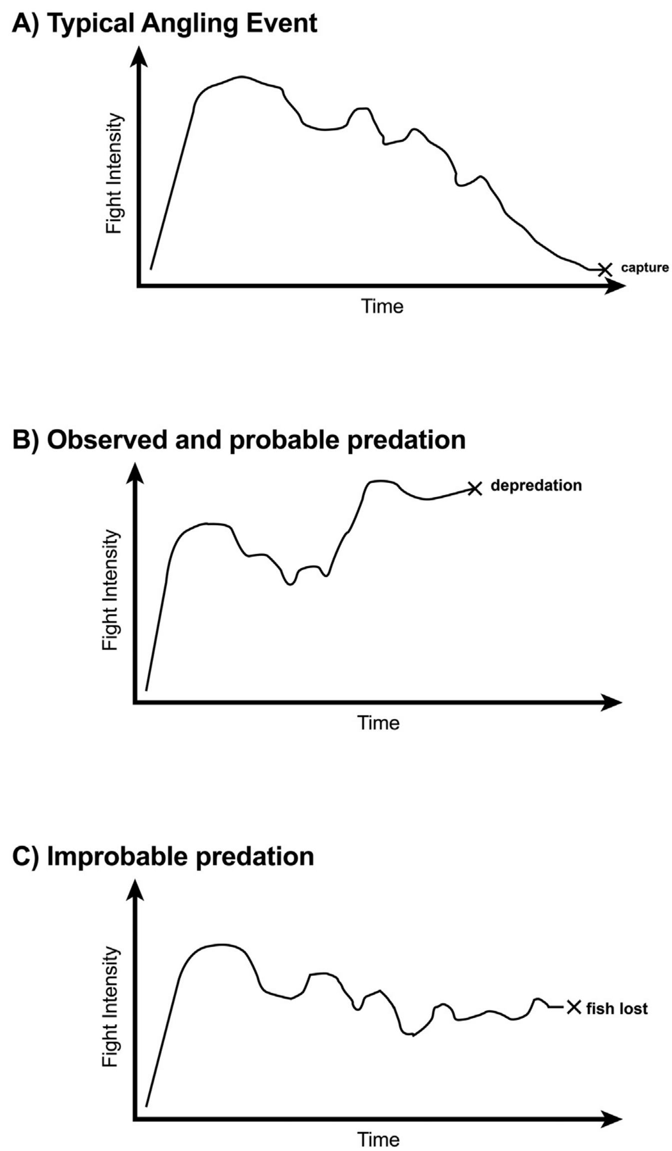

Fig. 3. Fight trajectories for simulated fisheries interactions comparing changes in fight intensity over time that fish are hooked. These tracings describe A) typical angling events, B) observed predation events and C) improbable predation.

from the same fish at both 0 and 30 min), and their interactions as predictors, and individual fish as a random intercept to account for repeated measures in individuals. For both linear models, backwards model selection via AIC was used to determine the final model. With the permit post-release accelerometer data, the raw acceleration values were used to calculate overall dynamic body acceleration (ODBA), which is a reliable measure of overall animal activity levels (Gleiss et al., 2011; Brownscombe et al., 2018). The effect of air exposure and time elapsed on ODBA was evaluated using linear mixed effects modelling ('nlme' package, RStudio).

3. Results

3.1. Depredation

In total, 205 permit (n = 205, 66.2 ± 0.81 cm fork length, mean ± SEM in landed fish) were hooked by recreational angling. Depredation rates (which include confirmed, probable and possible depredation events) varied greatly amongst sites and ranged from 0% to 90% (Fig. 4). However, only 6 of 33 sites (18.18%) had incidences of depredation (including confirmed, probable and possible depredation events). With observed depredation events, CIT identified a significant partition between two groups: 1) Atlantic structures and Gulf structures (n = 88, misclassification rate = 13.6%, depredation rate = 14%), and 2) flats, nearshore structures, and western structures (n = 117, misclassification rate = 0%, depredation rate = 0%) (t = 17.0, p = *.002*), indicating the two groups of habitat types had significantly different depredation rates. With observed and probable predation rates combined, the same habitat partitions were observed (t = 32.0, p < *.001*). Geographically, the locations with the highest incidence of depredation were found in the Gulf structure and Atlantic structure habitats, with a lower frequency in western structures, and no incidences of depredation in any other habitat type. Coordinates of the sites cannot be publicly shared to protect the interests of the collaborative members of the fishing community.

3.2. Physiological and behavioural responses to C&R

Fish held exposed to 0 min of air (n = 17) had a mean blood lactate concentration of 7.35 ± 0.62 mmol/l (Table 3) and fish exposed to 2 min of air (n = 14) had a mean blood lactate concentration of 7.46 ± 0.97 mmol/l (Table 3) immediately after capture. When compared to their blood lactate concentration following a 30-min holding period, both groups exposed to either 0 or 2 min of air exposure experienced significant increases in blood lactate (0MIN, 13.82 ± 0.28 mmol/l; 2MIN, 14.32 ± 0.28 mmol/l; Table 3), though increases were not significantly different between groups. Blood glucose exhibited the same pattern following a 30-min holding period, with 0MIN fish increasing from 6.19 ± 0.64 mmol/l (Table 3) to 15.85 ± 0.85 mmol/l (Table 3) and 2MIN fish increasing from 6.23 ± 0.65 mmol/l (Table 3) to 14.93 ± 11.1 mmol/l (Table 3) without any significant difference between groups. Blood pH was not significantly affected by any measurable parameter and was not significantly different between time periods or 0MIN and 2MIN treatment groups (Table 3). Using linear mixed effects modelling, fight time, temperature and air exposure were not significant predictors of changes in blood lactate, blood glucose or blood pH. The only significant predictor for both lactate and pH was collection time (0 min vs. 30 min post-capture; Table 4).

RAMP reflex testing results indicated that only one fish showed any impairment, which included both equilibrium and tail grab reflexes immediately following air exposure. However, this fish regained equilibrium within 60 s and, like all fish from both groups (n = 17 for 0 min air exposure, n = 14 for 2 min air exposure), had no signs of reflex impairment before release following a 30-min holding period in a livewell containing fresh, circulating seawater.

3.3. Post-release behaviour and survival

A comparison of permit swimming activity (Overall Dynamic Body Acceleration; ODBA) measured with accelerometers showed that fish in the 0MIN group had higher mean swimming activity rates, but there was a high level of variance within groups due to small sample sizes (Fig. 5). There was therefore no significant difference between fish held in water (n = 3) and fish exposed to 2 min of air exposure (n = 5) (t = −0.884, p = *.410*). All released fish were successfully tracked with accelerometers for the entire 20-min period, with the exception of one individual, from which the accelerometer was released 8-min post-release near an offshore shipwreck. The fish suddenly began swimming rapidly, followed by the loss of all terminal tackle, including the accelerometer, all of which indicated a potential depredation event.

4. Discussion

The goal of C&R fishing is for the fish to survive and experience

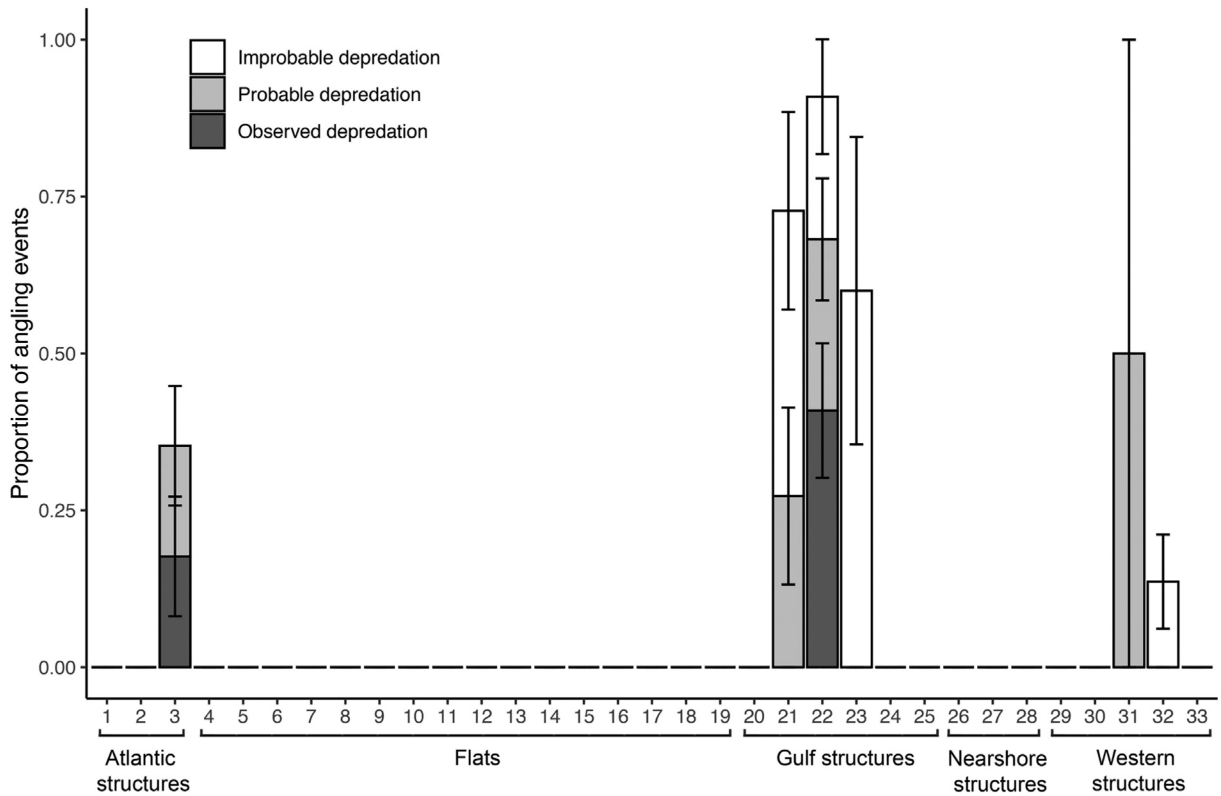

Fig. 4. The likelihood of depredation on angled permit organized by site (#) and habitat type in the Florida Keys.

limited biological fitness consequences. This can be compromised due to a number of factors, including depredation prior to capture, C&R stressors leading to immediate mortality, delayed mortality, or reduced fitness, or post release predation related to behavioural impairment (Brownscombe et al., 2016). In this study, we found that permit had moderate physiological stress responses to angling stressors and

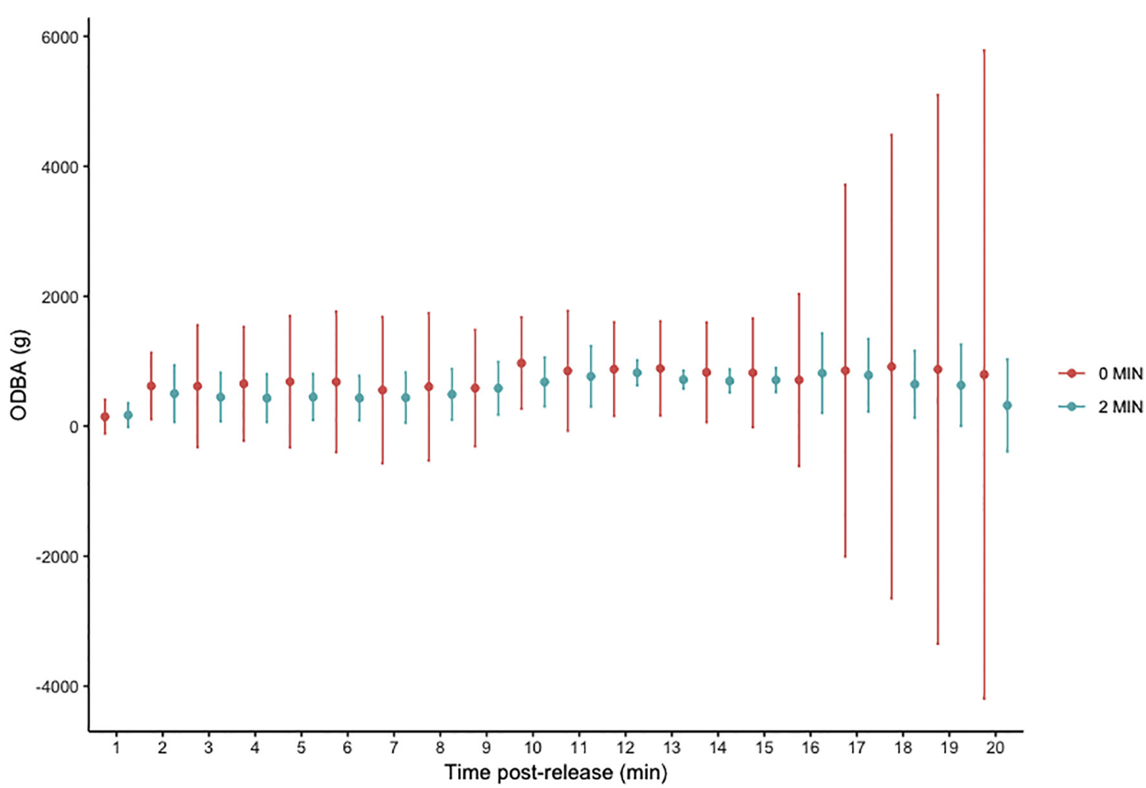

Fig. 5. Overall dynamic body action (ODBA) vs. time of permit exposed to 0 min of air exposure (0 MIN; n = 3) or 2 min of air exposure (2 MIN; n = 5) over the course of a 20-min trial period.

exhibited very little reflex impairment; however, high depredation rates prior to capture in some locations is concerning. The former is encouraging since a species' response to C&R is important in evaluating the suitability of C&R angling as an effective management strategy (Cooke and Suski, 2005; Arlinghaus et al., 2007). The latter is worrisome since high depredation rates, especially at sites used by permit for spawning, could have population-level implications (Sadovy and Domeier, 2005).

Living in a high predator density environment in the Florida Keys, permit have the potential to be susceptible to the negative impacts of depredation. In this study, depredation rates were variable amongst locations and habitat types. Flats and nearshore structure habitats had no incidences of depredation: a stark contrast when compared to offshore habitats, including Gulf and Atlantic structures, where depredation rates were variable but highly prevalent at specific sites. Most alarmingly, permit depredation rates were high in certain locations (up to 90%, one specific site), including a well-known spawning location during the spawning season (35%). Higher depredation rates at specific offshore structure habitats may be related to predator (sharks) densities, although sharks do inhabit all of these habitat types (J. Brownscombe, unpublished data). A key factor may be that permit are more transient on the shallow-water flats and nearshore structures, moving to locations with the tides, while they remain resident at offshore structures for longer periods in large schools (J. Brownscombe, unpublished data). Likelihood of depredation may change if fish spend less time in areas with lower predator densities (ie. The flats) and spend more time at these offshore structures where predator densities may be higher. These behavioural patterns may cause permit to be more vulnerable to opportunistic predation when they reside at defined structures and can represent a stationary food source for predators. When combined with angling pressure, these factors may enable sharks to capitalize on opportunities to depredate hooked fish. Indeed, the two locations with the highest incidences of confirmed depredation rates (one shipwreck in the Gulf of Mexico and one in the Atlantic Ocean on the Florida Reef Tract) are widely known and publicly shared fishing locations. The potential population impacts of fishing-related depredation depends on a number of factors, including fishing pressure, the number of permit spawning locations, and natural permit predation rates. Although these factors are largely unknown, an ongoing tracking study suggests the majority of permit in the region utilize a single spawning site where depredation rates were 35%. Regardless of natural predation rates, there is certainly potential that high fishing pressure at this site could have a negative impact on the population.

Based on habitat, fish caught on western Structures account for the bulk of the total catches (n = 73, 35.6%, Table 2), with flats (n = 39, 19.1%, Table 2) accounting for the second largest group. Although not quantified here, our fishing efforts were likely similar or even higher on flats habitats compared to nearshore or western structure, meaning that more time and effort was spent targeting permit in flats habitats. When considered alongside the catch data, this may reflect a general pattern in the permit fishery where hooking and capture rates in shipwreck habitats are higher compared to other habitat types. This is logical given that permit aggregations occur in predictable, stationary locations on these offshore structures that are easier to identify (visually: near the surface, or electronically: using sonar equipment at greater depths) and capture compared to fish in low predation risk habitats. It has also become common for recreational anglers to use sonar technology to locate and target large schools of fish in water that is too deep to see in. Fishing for permit at greater depths may increase the likelihood of depredation given that there is a greater distance between the hooked fish and the boat, though this study did not account for depth of capture. In some locations, depredation rates that encompass confirmed, probable and possible events approach > 90% of angled permit, as observed in this study. Thus, depredation may represent a significant conservation concern to the Florida Keys permit fishery and reinforces the need to understand the behaviour of predators habituated to the presence of anglers on offshore structure. It must also be considered that depredation is a dynamic threat given its potential for variability over both space and time, making management of its impact difficult. Moreover, understanding the regional connectivity between different habitat types that permit use, in addition to understanding how predators use the same habitat(s), is essential to identifying how and why these fish are more susceptible to depredation.

Table 2
Permit angling events examining catches and depredation, sorted by habitat.

	Likelihood of depredation				All events	
Habitat	Yes (100%)	Probable (> 50%)	Improbable (≤ 50%)	No (0%)	Total	Total (%)
Flats	0	0	0	39	39	19.1%
Nearshore Structure	0	0	0	5	5	2.4%
Atlantic Structure	3	3	0	19	25	12.2%
Western Structure	0	1	3	69	73	35.6%
Gulf Structure	9	9	13	32	63	30.7%
TOTAL	**12**	**13**	**16**	**164**	**205**	**100%**
TOTAL (%)	5.9%	6.3%	7.8%	80.%	100%	

Likelihood of depredation indicates if a fish (permit) was attacked or removed from the end of a line for reported catch data. Total (in all cases) refers to the total number of catches for each habitat type (horizontal rows) or based on likelihood of depredation (vertical columns). Total (%) refers to the number of catches in each column or row represented by percentage. Yes (100%) indicates a conclusive event where predators were physically observed removing angled fish. Probable (> 50%) indicates a higher likelihood of depredation occurring, based on angler/guide experience (see Methods, 2.1). Improbable (≤50%) indicates a lower likelihood of depredation occurring, based on angler/guide experience (see Methods, 2.1). No (0%) indicates no depredation occurring (i.e. angled fish were successfully captured). Habitats are defined in Methods (2.1) and are oriented according to Fig. 1.

For fish that were captured, our physiological, reflex impairment and swimming performance findings suggest that permit were robust to the effects of C&R under our experimental conditions. No significant differences in blood lactate, blood glucose or blood pH were found between fish exposed to 0 or 2 min of air exposure. Fish can compartmentalize lactate produced by exhaustive exercise in muscle tissue, and its detection in the blood is likely a result of passive lactate diffusion into the bloodstream as it continues to accumulate following bouts of exercise (Wood, 1991). The observed increase in circulating glucose is likely a result of increased energy mobilization as fish attempted to recover physiologically from increased swimming activity. Muscle glycogen stores may be depleted with evasion tactics, burst swimming and regular muscle contraction; thus, glucose is mobilized to replenish these stores. However, the increases observed in lactate and glucose could have resulted from a combination of repeated blood sampling and/or confinement stress associated with fish being held in a livewell for 30 min before a second sample was obtained. Notably, pH did not change significantly between 0MIN and 2MIN fish, or 30 min after fish were caught. It is possible that permit were able to excrete excess H^+ ions during the 30 min time period they were being held, similar to skipjack tuna (Perry et al., 1985), thus accounting for the lack of change in blood pH. When compared to other targeted flats fishing species (bonefish; sp. *Albula vulpes;* (Suski et al., 2007), permit exhibit a similar secondary stress response with increases in blood glucose and lactate (Table 3) measured 30 min post capture. However, unlike bonefish (Brownscombe et al., 2013), permit reflex impairment (RAMP testing) or swimming impairment was not significantly different between 0 and 2 min of air exposure. Additionally, no significant difference in ODBA was found between fish exposed to 0 or 2 min of air exposure (Fig. 5), although this is potentially attributed to the low sample size between groups (n = 5 and n = 3 respectively). The lack of significance in ODBA may have also been caused by a change in fish behaviour in response to the fixation of accelerometer packages, or by the drag they create when fixed to the caudal peduncle of the fish. Further evaluation

Table 3
Summary table of blood physiology parameters and size data from permit exposed to minutes 0 or 2 min of air exposure held for 30 min.

	0 Minutes air exposure		2 Minutes air exposure	
	0 min	30 min	0 min	30 min
Lactate (mmol/l)	7.35 ± 0.62	13.82 ± 0.28*	7.46 ± 0.97	14.32 ± 0.28*
Glucose (mmol/l)	6.19 ± 0.64	15.85 ± 0.85*	6.23 ± 0.65	14.93 ± 1.11*
pH	6.85 ± 0.11	6.84 ± 0.09	6.82 ± 0.11	6.92 ± 0.17
Fork length (cm)	55.27 ± 3.97		51.25 ± 6.30	

Blood physiology data for permit exposed to 0 min of air exposure ($n = 17$) and 2 min of air exposure ($n = 14$). All fish were captured from offshore shipwrecks. Groups are divided into 2 categories: 0 min (blood collected immediately after capture) and 30 min (blood collected from the same fish following a 30-min holding period). * $p < .05$ for 0 min vs. 30 min blood collection within group. Data are presented as mean ± SEM.

Table 4
Linear mixed effects model outputs for permit blood lactate concentrations, blood glucose concentrations and blood pH in comparison to fight time, temperature, air exposure and collection time.

Model variable	Coefficient	*t-value*	*p-value*
Lactate			
Intercept	5.148	0.320	0.754
Fight time	0.133	0.771	0.456
Temperature	0.025	0.127	0.901
Air exposure	−0.240	−0.482	0.638
Collection time	0.211	8.289	< 0.001
Glucose			
Intercept	571.653	2.203	0.048
Fight time	5.383	1.507	0.158
Temperature	−6.019	−1.866	0.087
Air exposure	−1.359	−0.169	0.868
Collection time	6.491	7.803	< 0.001
Fight time*air exposure	−0.311	−2.078	0.059
pH			
Intercept	3.681	0.917	0.394
Fight time	−0.003	−0.142	0.894
Temperature	0.042	0.803	0.467
Air exposure	0.124	1.181	0.303
Collection time	0.000	1.169	0.287

Fight time was measured in minutes and includes the duration of time spanning from initial hook up to landing the hooked fish. Temperature is water temperature measured in Fahrenheit. Air exposure describes fish that were exposed to either 0 or 2 min of air prior to blood collection, simulating an admiration period. Collection time refers to the effect of the timing of blood collection and compares blood taken immediately after capture (0 min) and blood taken following a 30-min holding period.

of post-release swimming performance with larger sample sizes would be warranted. Despite the potential added stress of repeated blood sampling, permit stress responses were moderate relative to teleost fish, and very little reflex impairment was observed, supporting the notion that permit are resilient to common recreational angling stressors. The possibility that permit may have been pushed to their recovery threshold upon capture must also be considered, and that the additional stress of air exposure may not have been evident in the changes observed in their blood physiology parameters.

Though depredation is an emerging challenge to conservation and management, small changes in angler behaviour could mitigate the potentially negative impact of depredation on permit populations, especially during the spawning season. Through regulations or voluntarily, there is opportunity for anglers to alter their behaviour and move between multiple habitats to responsibly target permit. Avoiding sites where depredation is a known problem (or ceasing fishing on days when depredation rates are observed to be high) could be effective in reducing the risk that predators depredate angled fish, as there are many fishing locations where depredation rates are low. However, importantly, anywhere that permit are aggregated for long periods (Brownscombe et al., n.d.), and angling pressure is high, could result in rapidly increasing depredation rates. Avoidance of fishing permit in known spawning aggregations with high predator densities would be a particularly valuable conservation action. Site-based, seasonal closures could also provide protection for fish aggregating at these sites, making them less vulnerable to mortality caused by depredation. Gear changes, like using stronger rod, reel and line combinations, would allow anglers to bring fish in more quickly, although the efficacy of this tactic in reducing depredation has yet to be tested. Anglers can also use word-of-mouth and social media to communicate locations that are particularly bad for depredation, encouraging other members of the community to avoid these areas if possible. These are easy and important steps that observant anglers can take to minimize their potential impact on fish populations until appropriate regulatory action can be taken.

The study of depredation in C&R recreational fisheries is still in its infancy; in the case of permit in the Florida Keys, it presents itself as a potential threat to conservation at certain locations with high predator densities and fishing pressure. Our findings suggest that the C&R practices we tested (varied fight times and air exposures) had a minimal impact on permit physiological stress responses, reflex impairment, and post-release swimming behaviour, however, enforcing C&R regulations on the permit fishery may not be sufficient enough to protect spawning populations of fish. Studies have shown that harvesting fish from spawning aggregations is detrimental to fish populations (Coleman et al., 2011), especially if species exhibit a strong fidelity for yearly spawning sites (Domeier and Colin, 1997; Sadovy and Domeier, 2005); a particular concern for permit (Brownscombe et al., 2019). Angling for permit at specific sites where fish are aggregating could render C&R fishing unsustainable if rates of depredation are too high. Future research could address such concerns by identifying the site-specific relationship shared between the frequency of recreational fishing, the residency of predators at these locations and the rate at which depredation occurs. Additionally, finding tools that are able to reliably and accurately quantify absolute depredation events, where individual sharks and fish can be identified, would provide valuable insight into the potential impact of depredation on specific fish populations.

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Declaration of Competing Interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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