

Using functional responses to quantify interaction effects among predators

Ryan J. Wasserman^{*,1,2}, Mhairi E. Alexander^{2,3,4}, Tatenda Dalu⁵, Bruce R. Ellender^{1,2}, Horst Kaiser⁶ and Olaf L. F. Weyl^{1,2,6}

¹South African Institute for Aquatic Biodiversity (SAIAB), P. Bag 1015, Grahamstown 6140, South Africa; ²Centre for Invasion Biology, South African Institute for Aquatic Biodiversity (SAIAB), P. Bag 1015, Grahamstown 6140, South Africa; ³Institute for Biomedical and Environmental Health Research (IBEHR), School of Science and Sport, University of the West of Scotland, Paisley PA1 2BE, Scotland, UK; ⁴Department of Botany and Zoology, Centre for Invasion Biology, Stellenbosch University, Matieland 7602, South Africa; ⁵Department of Zoology and Entomology, Rhodes University, P.O. Box 94, Grahamstown 6140, South Africa; and ⁶Department of Ichthyology and Fisheries Science, Rhodes University, P.O. Box 94, Grahamstown 6140, South Africa

Summary

1. Predator diversity alterations have been observed in most ecosystems as a result of the loss and/ or addition of species. This has implications for predator–prey dynamics as non-trophic interactions among predators, so-called multiple predator effects (MPE), are known to influence predation success. In addition, there is often a density-dependent relationship between prey availability and prey consumption (functional response). While MPE investigations are common in the literature, functional responses have rarely been incorporated into this field of predation ecology.

2. Here, we outline an experimental procedure that incorporates functional responses into multiple predator effect studies. Using three fish species with different functional traits as model predators (bluegill *Lepomis macrochirus*, southern mouthbrooder *Pseudocrenilabrus philander* and banded tilapia *Tilapia sparrmanii*), we assess intra- and interspecific predator interaction outcomes on predator–prey dynamics. This was done by contrasting observed functional responses of heterospecific and conspecific combinations of predators with expected responses based on those of individual predators.

3. Multipredator combinations produced variable results. Bluegill were the only species in which observed conspecific multipredator functional responses matched those of expected based on individual performance (prey risk neutral effects). In contrast, prey risk reduction was observed for both mouthbrooder and tilapia conspecific multipredator trials. Heterospecific combinations revealed strong prey risk reduction effects for mouthbrooder–tilapia and bluegill–tilapia trials, while mouthbrooder–bluegill multipredator functional responses combined additively. These results are discussed within the context of behavioural traits of the species and the development of a trait-based predictive framework.

4. Using a functional response approach allowed for the assessment of multiple predator effects across a range of prey densities. We propose that the incorporation of within-guild predator combinations into classic functional response investigations will enhance predictive capacity development in competition and predation ecology.

Key-words: conspecific aggression, fish predators, heterospecific aggression, multiple predator effect, predictive theory, trait-based framework

Introduction

Across most ecosystems, biodiversity levels are changing as a result of human-mediated extinctions and

introductions of species outside of their native ranges (Sala *et al.* 2000; Naeem, Duffy & Zavaleta 2012). These human-driven impacts often result in predator biodiversity alterations, which can involve guild homogenization or increases in species richness (Simberloff & Von Holle 1999; Griffin, Byrnes & Cardinale 2013).

*Correspondence author. E-mail: ryanwas21@gmail.com

Understanding predator diversity and its implications for trophic interactions is therefore important for predictive theory (Schmitz 2007). Assessments of predator diversity effects represent an important avenue of ecological research as predators provide valuable ecosystem services, and these services are likely to fluctuate when predator diversity levels are altered (Ives, Cardinale & Snyder 2005; Duffy, Richardson & France; Schmitz 2007). At the predator level, considerations on the functional role of within-guild interactions on prey consumption are mixed, with implications for process level understanding of ecosystem functioning (Schmitz 2007; Griffin, Byrnes & Cardinale 2013).

Since virtually all ecological communities are comprised of multiple predators that utilize mutual prey resources, the potential for competition is always present (Sih, Englund & Wooster 1998; Barrios-O'Neill *et al.* 2014). In this way, predators not only interact with their prey, but they are often forced to interact with one another (Polis, Myers & Holt 1989; Johnson *et al.* 2009). Considering the nature and strength of interactions among predators has been identified as an important component of predation studies as predator–predator exchanges can have implications for predator–prey interaction outcomes (Sih, Englund & Wooster 1998; Bolker *et al.* 2003; Vonesh & Osenberg 2003). As such, it is now well recognized that the effects of predators on prey do not necessarily combine additively as multiple predators can modify predator–prey interactions (Soluk 1993; Sih, Englund & Wooster 1998; Sokol-Hessner & Schmitz 2002; Barrios-O'Neill *et al.* 2014). Multiple predator effects (MPE) resulting from non-additive competition among predators are generally assessed across species, that is the outcome of interactions between two species (predator and prey) as altered by an additional predatory species (Weigelt *et al.* 2007; Barrios-O'Neill *et al.* 2014; Anderson & Whiteman 2015). However, studies contrasting heterospecific and conspecific non-additive effects are less common (Young 2004; Forrester *et al.* 2006; Anderson & Whiteman 2015). Conspecific non-additive effects are also relevant within the context of biodiversity as introduced non-native predators can reduce within-guild species richness while still functionally maintaining the trophic level in which they are positioned, representing a type of predator homogenization (Lohrer & Whitlatch 2002; Griffin, Byrnes & Cardinale 2013). Additionally, while a wealth of information is available regarding MPE on prey consumption, most of these studies assess these effects at single prey densities (Harvey, White & Nakamoto 2004; Griffen 2006; Porter-Whitaker *et al.* 2012). However, density-dependent prey acquisition is an important element of predator–prey interactions and predation efficiency is known to vary with prey density (Abrams 1982, 2000; Alexander *et al.* 2012). To this end, the present study proposes an application of a classic ecological approach for the assessment of intra- and inter-specific predator interaction outcomes on predator–prey dynamics.

The relationship between prey density and consumption rate is known as the predator functional response (Holling 1959). Functional responses are extensively used by ecologists and have helped gain insight into optimal foraging theory (Abrams 1982), invasion biology (Dick *et al.* 2014) and the field of predation in general (Soluk 1993; Jeschke, Kopp & Tollrian 2002). There is, however, a relatively small body of literature that has empirically assessed the effects of interactions among predators on density-dependent prey acquisition (Soluk 1993; Losey & Denno 1998; Barrios-O'Neill *et al.* 2014). The little work that has been done in this regard has produced varied results. Firstly, it has been shown that when combined, the overall functional response of heterospecific predator combinations may be lower than one would expect based on single predator functional responses (Soluk 1993). However, this is in contrast to heterospecific predator combinations that result in an overall functional response that doubles the predicted functional response based on single predator performance (Losey & Denno 1998). In addition, multiple predator effects at the conspecific level have also shown that consumption can be additive and group consumption is therefore predictable from individual-level consumption (Barrios-O'Neill *et al.* 2014). These contrasting results highlight that multipredator interactions are complex and that predator combinations can result in prey risk reduction as a result of antagonism among predators, risk enhancement as a result of synergism among predators or a risk neutral scenario whereby predator effects simply combine additively. Here, we account on an experiment assessing the effects of multiple fish predators on shared prey. In this experiment within the context of risk reduction, risk enhancement and risk neutral multiple predator effects, we contrast the overall functional responses of heterospecific and conspecific combinations of predators with predicted responses based on individual predator performances.

Three predatory fish species were used in the experiment, bluegill *Lepomis macrochirus* Rafinesque, 1819, southern mouthbrooder *Pseudocrenilabrus philander* (Weber, 1897) and banded tilapia *Tilapia sparrmanii* A. Smith, 1840. Bluegill (Centrarchidae) are native to North America, while southern mouthbrooder and banded tilapia (Cichlidae) are native to parts of Africa. All three fishes have been distributed extensively outside of their native ranges and at some localities in South Africa occur sympatrically as non-native predators (Ellender & Weyl 2014; Muller, Weyl & Strydom 2015). Aside from the fact that the presence of these fish species now represent a 'non-native species cocktail' in select regions, these fishes have certain trait differences making for ideal combinations to highlight a suite of possible MPE outcomes. Bluegill are a shoaling species (Colgan *et al.* 1979; Gross & MacMillan 1981; Wahl & Stein 1988; Savino & Stein 1989; Dugatkin & Wilson 1992) and are primarily predators incorporating mostly animal prey into their diets (Marshall 2011; Taguchi *et al.* 2014) and while southern

mouthbrooders (hereafter referred to as mouthbrooders) are also primarily predatory, they are a non-shoaling species not found in close association with conspecifics (Ribbink 1971; Polling, Schoonbee & Saayman 1995). Banded tilapia (hereafter referred to as tilapia), however, occasionally occur in small shoals or pairs, but are also often solitary (Skelton 2002; Marshall 2011). While this species readily consumes animal prey, it is more omnivorous than the bluegill and mouthbrooder, incorporating plant material into its diet (Zengeya & Marshall 2007; Marshall 2011). Bluegill are considered a flexible species with proportions of populations often found in both open waters and in close association with structure and littoral regions (Werner & Hall 1988; Yonekura, Kohmatsu & Yuma 2007). While published information on the habitat preference of the mouthbrooder and tilapia is limited, they are most commonly encountered in shallow littoral habitats with the former often associated with sandy or rocky substrates and vegetation structure and the latter more closely associated with vegetation structure (Bruton 1978; Khoza, Potgieter & Vlok 2012). The three species therefore overlap in habitat use in environments in which they co-occur, as has been observed in impoundments and pools of the Kariega River system, South Africa, where experimental animals were collected for the present study (Table 1).

In this study, we firstly determined single species functional responses at the individual level for bluegill, mouthbrooder and tilapia, towards small fish prey. We then use this information to explore how individual performance translates to multiple predator overall performance within the context of prey risk reduction, neutral or enhancement effects. This was achieved by contrasting individual functional response information with that of both conspecific and fully crossed mixed predatory fish species pairs (Fig. 1). We predicted that for conspecific multipredator pairs, prey risk reduction (rather than risk neutral or risk enhancement) would be more prevalent for species that do not typically utilize habitats in close conspecific association. For heterospecific combinations, however, we predicted that different multispecies combinations would not result in similar prey risk scenarios and explored mechanisms potentially driving any observed differences. In this way, the study highlights the complexities of multiple predator effects and how prey consumption at different prey densities is a potentially important addition warranting consideration in MPE studies.

Table 1. Summary of conspecific association level, predominant feeding mode and habitat association of the three species employed in the multipredator functional response experiment

	<i>Lepomis macrochirus</i>	<i>Pseudocrenilabrus philander</i>	<i>Tilapia sparrmanii</i>
Common name	Bluegill	Mouthbrooder	Tilapia
Conspecific association	Shoaling	Solitary	Small shoals/solitary
Feeding	Predator	Predator	Omnivore
Habitat association	Open water/littoral	Rocky substrate/vegetated littoral	Vegetated littoral

Materials and methods

EXPERIMENTAL DESIGN

All predatory fish were collected in March 2015 by seine netting in Craig Doone Dam (33°21'35.65"S; 26°28'41.29"E), near Grahamstown, South Africa. Prey used were Mozambique tilapia *Oreochromis mossambicus* captive bred offspring, supplied by AquaCulture Innovations, Grahamstown. All fish were transported to the Department of Ichthyology and Fisheries Science, Rhodes University, Grahamstown and were housed in separate 600-L tanks in a closed recirculating system (water flow to each tank 1 L min⁻¹; 18 ± 1 °C). All fish were allowed to acclimate to the system for at least 72 h prior to use in feeding trials and were maintained on a diet of earthworms to standardize prior experience.

Experiments were conducted in individual 26-L cages constructed from 1.5 mm mesh and floated using buoyancy aids in 15 separate 300-L fibreglass tanks that were part of the same flow-through system as the holding tanks so that each fibreglass tank held one mesh cage. Predators were size-matched with respect to total length (TL): bluegill (mean ± SD) = 75.1 ± 1.9 mm TL; mouthbrooder = 76.4 ± 2.6 mm TL; and tilapia = 76.7 ± 2.2 mm TL. Prey were selected from a common size class (10.0 ± 0.5 mm TL). Preliminary trials on prey in the holding tanks indicated that in the absence of predators the prey used the entire tank with individuals observed actively swimming throughout the water column. Fish predators were randomly selected 2 h prior to use and placed in the mesh cage to reacclimate, either individually, with a conspecific individual, or in a mixed-species pair depending on the treatment. Single- and mixed-predator treatments were fully crossed so that all predator combinations were trialled with fish only being used once for a single feeding trial. After the reacclimation period, fish were presented with tilapia prey at six densities (2, 4, 8, 16, 32 and 64) with at least three replicates per density per treatment combination. Feeding trials were run for 1 h, after which prey consumption was examined. Controls were three replicates of each prey density in the absence of predators. Since all predators are active diurnally, feeding trials were only conducted during daylight hours.

As feeding behaviour was not continuously observed, it was not possible to know which predator species was responsible for prey consumption in the heterospecific combinations. For this reason, additional trials were run for all multispecies combinations at a single prey density of 32 prey and filmed ($n = 3$ for each combination), which allowed the number of fish consumed by the respective species during each combination to be determined. For this component of the study, however, novel fish were not available and as such we used fish that had been employed in the functional response component of the study. The same experimental procedures as outlined above were employed for this component, and GoPro cameras (Hero 3) were placed directly overhead the cages allowing for an aerial view of the inside of the cages. Based on this footage, the number of fish consumed by the respective species in each combination of predators was determined. In addition, interactions between the predators could be quantified and related to interference. Behavioural traits were scored for each species of fish

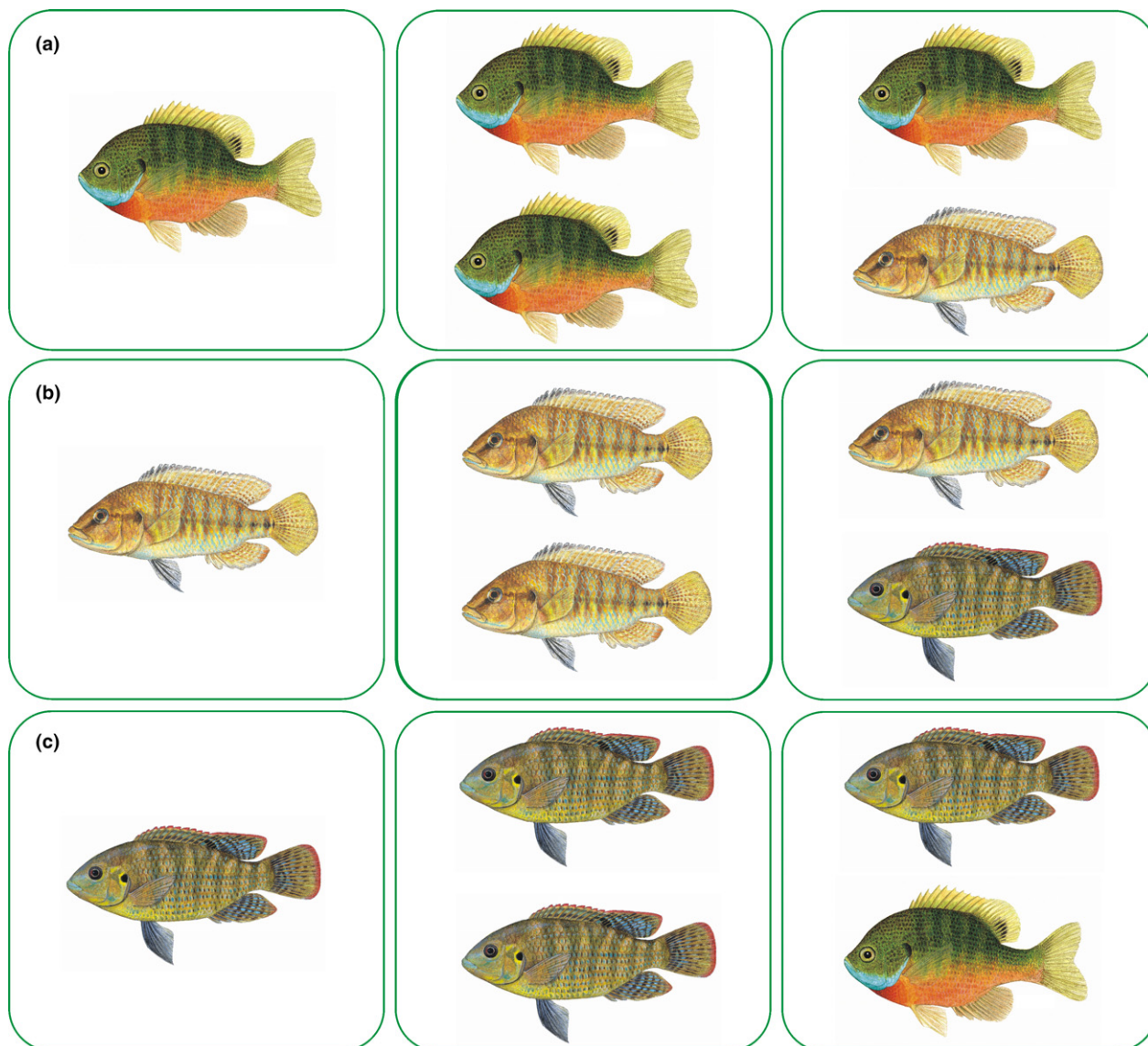


Fig. 1. Experimental predator treatments comprising individual predator, conspecific and heterospecific predator combinations of (a) bluegill (*Lepomis macrochirus*), (b) mouthbrooder (*Pseudocrenilabrus philander*) and (c) tilapia (*Tilapia sparrmanii*).

from analysis of video footage that was gathered during filmed trials. Four traits were selected for analysis that provided unambiguous assessment of behaviour: (i) lateral displays, defined as the spreading of operculum and erecting of fins by an individual when it was within one body length of the heterospecific; (ii) lunging, defined as the quick movement towards the heterospecific and included chasing (where one fish was chased in short, rapid bursts by the other); (iii) follow, defined as the slow following of a heterospecific within a distance of one body length; and (iv) retreat, defined as the rapid swimming away from a heterospecific in response to displays or any type of approach by the partner fish. These behaviours were counted for both species in each filmed multipredator trial.

DATA ANALYSIS

To test whether functional responses of two fish predators (conspecifics and mixed-species pairs) could be predicted by summing individual responses, the predicted combined consumption

was calculated using the following multiplicative model (Soluk 1993);

$$C_{ab} = N_p(P_a + P_b - P_aP_b)$$

where C_{ab} is the predicted combined consumption for a particular initial prey density (N_p) and P_a and P_b are the probabilities of being consumed by each predator present (fish a and fish b), respectively, over a 1-h period of exposure. This multiplicative model calculates predicted combined consumption that cannot exceed the total number of prey introduced. Data for predator a and predator b were generated from single fish functional response experiments. The predicted combined consumption was calculated as the expected data, while consumption from the observed multipredator trials was modelled as the observed data below.

Functional response type was first determined using logistic regression that tests for a negative linear coefficient (fitted using the maximum likelihood procedure) in the relationship between the proportion of prey eaten and prey density. This would indicate a type II functional response (Juliano 2001). We then modelled by

maximum likelihood estimation (Bolker 2008) type II functional responses (see Results) using the 'random predator equation' (Rogers 1972), which is appropriate where prey are not replaced as they are consumed, as was the case here (Juliano 2001);

$$N_e = N_0 \{1 - \exp[a(N_e h - T)]\}$$

where N_e is the number of prey eaten, N_0 is the initial density of prey, a is the attack constant, h is the handling time and T is the experimental period. To compare the obtained functional responses, datasets were nonparametrically bootstrapped ($n = 2000$) in order to construct 95% confidence intervals around functional response curves and their associated parameters. The random predator equation was fitted to each bootstrapped dataset using starting values of a and h that were obtained from the original maximum likelihood estimates. As bootstrapping allows data to be considered in terms of populations, if the confidence intervals do not overlap we were able to deduce that the functional responses and/or their associated parameters were different. Generalized linear models (GLMs) assuming quasi-poisson distributions were used to compare overall prey consumption between appropriate predator treatments.

Behavioural traits were scored and data arranged so that behaviour of the focal predator species towards each of the other predator species could be compared. A mixed-effects ANOVA (between-factor = partner fish; within-factor = behavioural trait), which accounted for non-independence of behavioural data, was used for each focal species. Data were log-transformed prior to analyses to meet normality assumptions. Significant interactions were investigated via post hoc t -tests that compared frequency of each of the behaviours performed towards partner fish. All analyses were carried out in R v. 2.15.1 (R Development Core Team 2013), and functional response modelling was undertaken using the 'frair' package (Pritchard 2014).

Results

In control trials, no prey deaths were recorded; therefore, prey mortality was attributed to predation, which was also visually observed. First-order terms derived from logistic regressions were all significantly negative, indicating that the functional responses obtained were all type II (Table 2; Figs 2–4), and the attack constant and handling time parameters of the functional response models were also all significant at $P < 0.001$ (Table 2).

Overall consumption of prey by individual predators was dependent on predator identity ($F_{(2,54)} = 6.66$, $P = 0.003$; Fig. 2), with tilapia consuming significantly fewer prey in comparison with both bluegill ($z = 2.41$, $P < 0.05$) and mouthbrooder ($z = 3.48$, $P < 0.05$). This was further evidenced in functional response curves where 95% confidence intervals did not overlap across the full prey density range between mouthbrooders and tilapia, and only at prey densities above 34 between bluegill and tilapia (Fig. 2).

Consumption by conspecific pairs was also dependent on the species identity ($F_{(2,54)} = 9.34$, $P < 0.001$; Fig. 3), and bluegill pairs consumed significantly more prey than pairs of both mouthbrooder ($z = 3.18$, $P = 0.004$) and tilapia ($z = 3.82$, $P < 0.001$). Differences in the magnitude of the functional responses were also evident among the conspecific pairs, with bluegill pairs producing heightened responses in comparison with the other two species (solid lines in Fig. 3). Differences in these functional responses were also demonstrated in the increased attack ($a = 4.931$) and decreased handling ($h = 0.047$) parameters in bluegill pairs in comparison with pairs of mouthbrooders ($a = 1.530$; $h = 0.162$) and tilapia ($a = 0.922$; $h = 0.093$) (Table 2). Of the three species, the observed functional response of the bluegill pairs overlapped with the predicted response across the full prey density range (Fig. 3a); however for both mouthbrooder and tilapia conspecific pairs, the observed functional responses were significantly reduced in comparison with the predicted curves (Fig. 3b,c). This reduction was greatest, however, in mouthbrooder comparisons, and this was further evident in the reduction in the observed attack and handling time values in comparison with the predicted parameters (Table 2).

A significant difference in prey consumed by mixed-pair treatments ($F_{(2,54)} = 3.71$, $P = 0.03$; Fig. 4) was driven by greater predation by bluegill and mouthbrooder in combination compared to the mix of bluegill and tilapia

Table 2. Parameter estimates and significance levels from first-order logistic regression analyses of the proportion of prey killed against initial prey density, with functional response parameters (a and h) and significance levels from the Rogers random predator equation

Predators	Data	First-order term, P	a	P	h	P
1 Bluegill	O	-0.037, <0.001	4.517	0.008	0.087	<0.001
1 Mouthbrooder	O	-0.040, <0.001	3.099	<0.001	0.051	<0.001
1 Tilapia	O	-0.027, <0.001	0.765	<0.001	0.098	<0.001
2 Bluegill	O	-0.048, <0.001	4.931	<0.001	0.047	<0.001
2 Mouthbrooder	O	-0.025, <0.001	1.530	0.003	0.162	<0.001
2 Tilapia	O	-0.027, <0.001	0.922	<0.001	0.093	<0.001
2 Bluegill	E	-0.045, <0.001	3.758	<0.001	0.043	<0.001
2 Mouthbrooder	E	-0.038, <0.001	3.844	<0.001	0.028	<0.001
2 Tilapia	E	-0.028, <0.001	1.299	<0.001	0.051	<0.001
1 Bluegill, 1 Mouthbrooder	O	-0.047, <0.001	5.523	<0.001	0.035	<0.001
1 Bluegill, 1 Tilapia	O	-0.036, <0.001	2.183	<0.001	0.065	<0.001
1 Mouthbrooder, 1 Tilapia	O	-0.053, <0.001	6.472	<0.001	0.063	<0.001
1 Bluegill, 1 Mouthbrooder	E	-0.043, <0.001	4.654	<0.001	0.034	<0.001
1 Bluegill, 1 Tilapia	E	-0.037, <0.001	3.209	<0.001	0.048	<0.001
1 Mouthbrooder, 1 Tilapia	E	-0.039, <0.001	3.389	<0.001	0.037	<0.001

O, Observed; E, Expected.

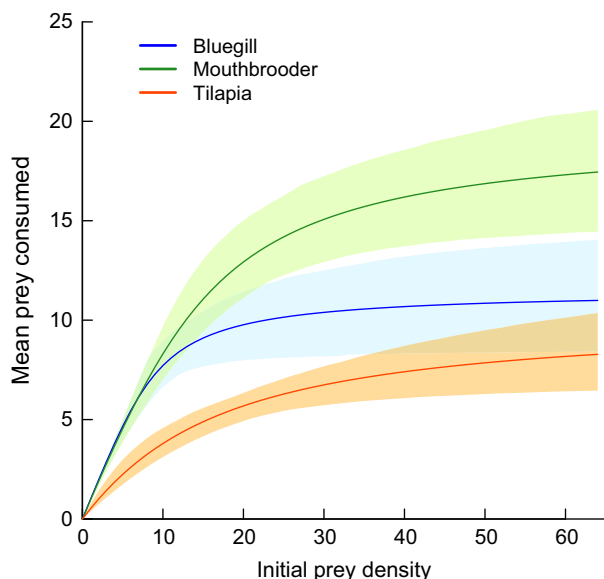


Fig. 2. Functional responses of individual bluegill (*Lepomis macrochirus*), mouthbrooder (*Pseudocrenilabrus philander*) and tilapia (*Tilapia sparrmanii*) towards common fish prey. Shaded areas are 95% confidence intervals.

($z = 2.54$, $P = 0.02$; solid lines in Fig. 4a vs. 4b). The observed functional response of bluegill combined with mouthbrooder overlapped with predicted values across the full prey density range, and the response curves were overlaid on each other (Fig. 4a). Observed responses of bluegill and tilapia in combination overlapped with predicted responses for the most part, although there was separation in the 95% confidence limits between prey densities of around 15–25 (Fig. 4b). Difference in functional response between the observed and predicted responses of mouthbrooder and tilapia were detected, however, and although the initial response was similar, responses diverged at around prey densities just above 26 (Fig. 4c). This divergence at greater prey densities is also evidenced in the two-fold increase in the handling parameter that was observed in mouthbrooder and tilapia pairs ($h = 6.472$) compared to those that were expected ($h = 3.389$).

Video analyses revealed that when combined, bluegill and mouthbrooder consumed (mean \pm SD) 13.33 ± 5.51 and 18.67 ± 5.51 prey items, respectively. However, the trials in which tilapia was used in combination with another fish species, both bluegill and mouthbrooders were responsible for 100% of prey consumption, eating all 32 prey. In behavioural observations, mouthbrooders failed to display retreating behaviour from either bluegill or tilapia; thus, this was removed from the analysis of this species. There were no significant main effects or interaction of partner fish and behavioural type on the frequency of behaviours observed in mouthbrooders. The frequency of behaviours of bluegill varied depending on behavioural type and the species of partnered fish ($F_{(3,12)} = 6.798$, $P < 0.01$). This was driven by a significantly greater number of retreats from mouthbrooders compared to tilapia

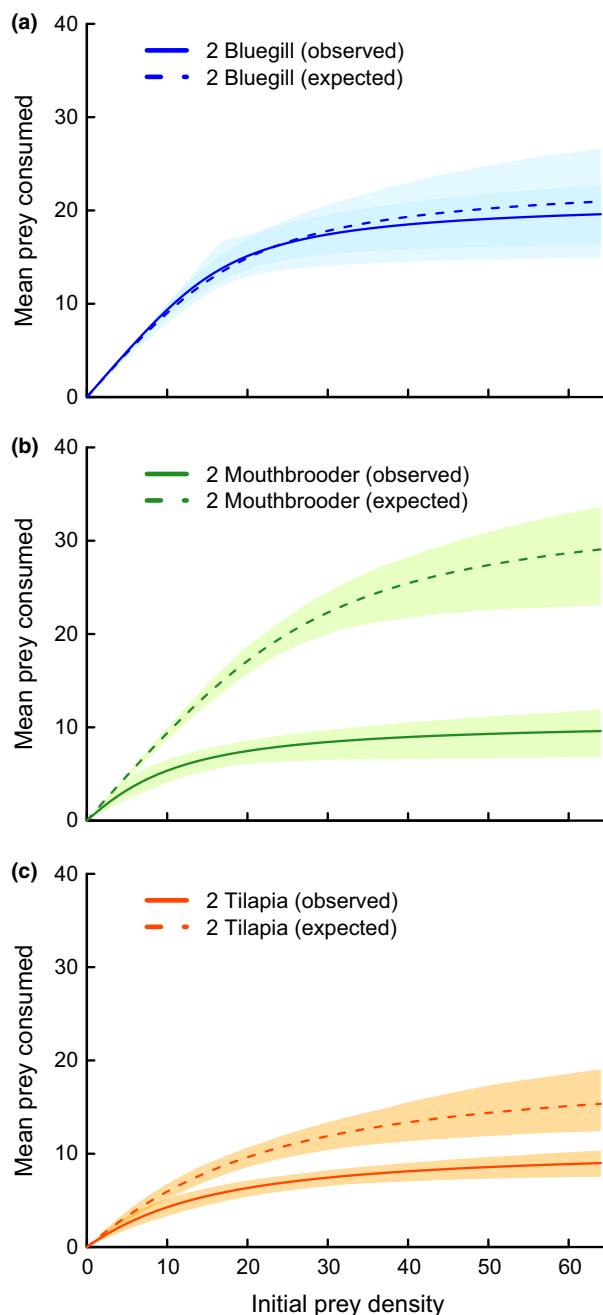


Fig. 3. Observed (solid) and expected (dashed) functional responses of conspecific pairs of (a) bluegill (*Lepomis macrochirus*), (b) mouthbrooder (*Pseudocrenilabrus philander*) and (c) tilapia (*Tilapia sparrmanii*). Solid/dashed lines represent model curve, while shaded areas are 95% confidence intervals. Expected responses calculated from the performance of individuals of each species (Fig. 2). Note the different y-axis scales when comparing with Fig. 2.

($t_4 = 3.069$, $P < 0.05$). In tilapia, frequencies of observed behaviours varied depending on behavioural type and the species of partnered fish ($F_{(3,12)} = 22.448$, $P < 0.01$). This was driven by a significantly greater number of chases of bluegills compared to mouthbrooders ($t_4 = 10.128$, $P < 0.05$) and greater retreats from mouthbrooders compared to bluegill ($t_4 = 4.247$, $P < 0.05$).

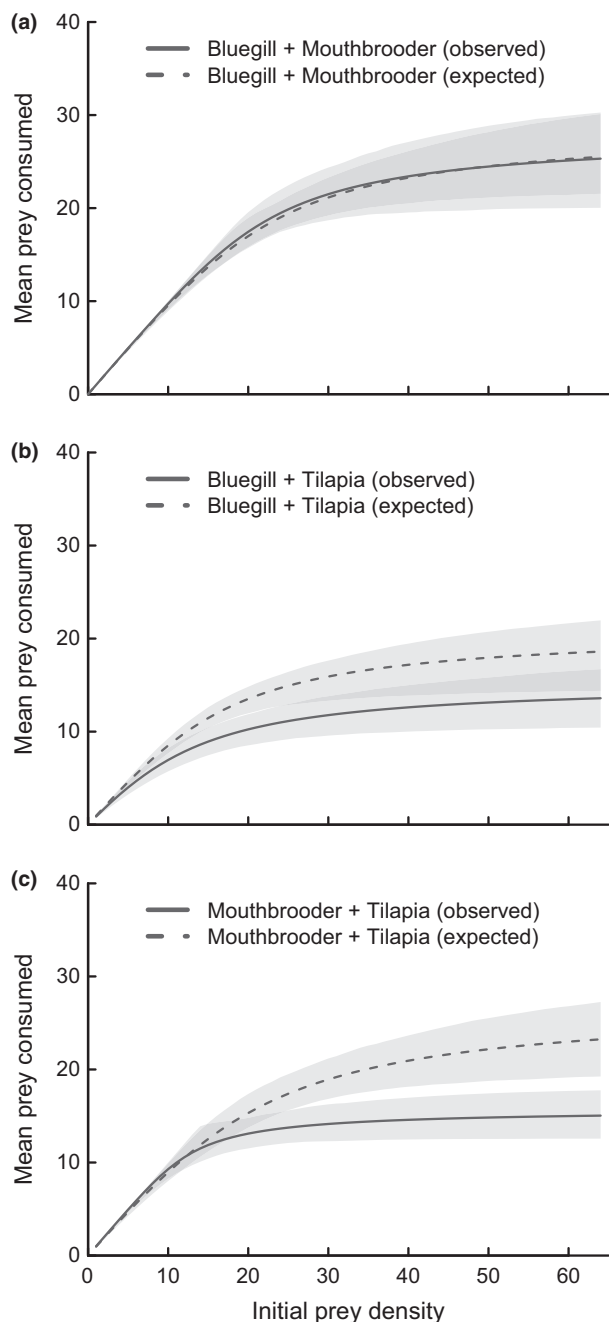


Fig. 4. Observed (solid line) and expected (dashed line) functional responses of heterospecific pairs of (a) bluegill (*Lepomis macrochirus*) and mouthbrooder (*Pseudocrenilabrus philander*) (b) bluegill and tilapia (*Tilapia sarrmanii*) and (c) mouthbrooder and tilapia. Expected responses calculated from the performance of individuals of each species (Fig. 2). Note the different y-axis scales when comparing with Fig. 2.

Discussion

In ecological communities, prey are seldom exposed to single predators and, as predators occur in a variety of combinations, varying degrees of impact on prey populations have been observed (Schmitz 2007; Barrios-O'Neill *et al.* 2014). In this study, we demonstrate that predator–predator interactions can be important in determining prey

consumption by investigating functional responses in individual as well as conspecific and heterospecific pairs of predatory fish. Specifically, we show that at both the conspecific and heterospecific level, prey risk varies as a result of predator–predator effects. Differences in the functional responses of individual species were found with mouthbrooders exhibiting heightened responses compared to both bluegill and tilapia towards the prey Mozambique tilapia. In conspecific pairs, however, this pattern did not hold as prey risk enhancement was observed for bluegill pairs which exhibited heightened functional responses in comparison with pairs of both mouthbrooder and tilapia where prey reduction was observed. Prey risk also differed among mixed-species pairs with combinations of bluegill and mouthbrooder producing a risk neutral scenario whereby observed functional responses were congruent with expected, based on individual predator performance. Often these results did not align with expected outcomes and we show that differences in predator identity and diversity can have important consequences for altering the outcome of what might be expected in density-dependent relationships between predators and their prey.

In predator–prey interactions with single predators, the expectation that mouthbrooders, as a solitary and predatory species (Polling, Schoonbee & Saayman 1995), would exhibit increased resource use in comparison with the other species held true. Indeed, overall consumption by this species was significantly greater than that of tilapia and a divergence in functional responses with bluegill was evident from intermediate to high prey densities. Bluegill are also predatory; however, this reduced consumption compared to the solitary mouthbrooder predator may be reflective of a less efficient individual predator that most commonly occurs in shoals with conspecifics (Smith & Warburton 1992). Tilapia exhibited reduced functional responses in comparison with bluegill and mouthbrooders, and this was similar across single fish and conspecific pair treatments. As an omnivorous species (Zengeya & Marshall 2007; Marshall 2011), it is likely that tilapia is less motivated to feed on this prey when it is rare, resulting in a reduction in the initial response, with increased consumption occurring only at higher prey densities.

Observed and expected responses of conspecific pairs diverged among the species as was evident via overlapping and separate confidence levels of the functional responses curves. Of the three fish species investigated here, only the functional response of bluegill aligned with what was expected. This suggests that although bluegill pairs do not facilitate feeding that results in prey risk enhancement, or synergistic consumption, they may not act to aggravate conspecifics. Conversely, a reduction in the responses of observed mouthbrooder pairs in comparison with the expected response suggests that this non-shoaling fish responds to the presence of conspecifics. Mouthbrooders are recognized as being an aggressive species (Ribbink 1971; Polling, Schoonbee & Saayman 1995; Marshall 2011), and the results reported here reflect this. In tilapia,

observed responses for two individuals were comparable to that for one individual. As a species that spends its time in loose association with conspecifics, these results suggest that interference levels lie somewhere between the shoaling bluegill and the solitary mouthbrooder. Additionally, tilapia are omnivorous and readily consume a broad range of prey and so competition for live prey, even at the conspecific level, may not be as fierce as in primarily predatory fish species.

All species exhibited or responded to aggression when in a heterospecific combination. These combinations revealed that there was strong interference competition between mouthbrooder and tilapia, with mouthbrooder out-competing the tilapia for prey resources at high prey densities. The same was observed for bluegill–tilapia combinations, with tilapia not consuming prey in the presence of bluegill. Video observations of heterospecific combinations highlighted strong interference between the fish species, with mouthbrooder and bluegill both reducing tilapia prey resource consumption. The mechanisms by which mouthbrooder and bluegill reduced prey consumption by tilapia differed, however, as indicated by the behavioural analyses. In the mouthbrooder–tilapia combinations, mouthbrooder were highly aggressive towards tilapia and this resulted in tilapia spending much time retreating and ultimately not feeding. In the bluegill–tilapia combinations, tilapia were observed slowly following the bluegill rather than actively feeding. The combination of bluegill and mouthbrooder functional responses combined additively, and video analyses revealed that these species in combination each consumed prey at a similar rate as each did in the single functional response trials. While this suggests that interference interactions between solitary and shoaling species are less pronounced than between two solitary species, aggressive behaviour was observed between mouthbrooder and bluegill. Indeed, mouthbrooder were equally as aggressive towards bluegill as they were towards tilapia. Bluegill also showed a high frequency of retreat behaviour towards mouthbrooder when in combination. But unlike tilapia, bluegill returned non-contact aggression towards mouthbrooder and spent time following the heterospecific, while repeatedly making successful attempts at the prey between contact bouts.

These findings highlight subtle behavioural difference among the three predator species which ultimately have implications for foraging. In addition, it is evident that similar outcomes, such as the prey risk reduction observed in both the mouthbrooder–tilapia and bluegill–tilapia combinations, can arise through alternate behaviourally mediated mechanisms. The fish used for the behavioural component of the study had, however, already been used once in functional response trials. It is therefore possible that there was an element of learning associated with the behavioural component of the study, as suggested in the overall higher consumption of prey in the behavioural experiment when compared to the functional response experiment at the same density. Despite this consideration,

the behavioural results highlight key interactions between predator species in heterospecific trials and provide mechanistic information into how multispecies observed differ from expected functional responses based on individual performance.

Incorporating a simple approach contrasting functional responses of expected and observed multiple predator combinations provides much information. For example, in addition to the information acquired in classic MPE experiments, this approach can provide valuable information on the importance of prey density dependence on predator–predator interactions. This is highlighted by the conceptual model (Fig. 5) that exemplifies the ways in which predator–prey dynamics can be altered. While both functional response and MPE investigations are well-established and independently highlight shortcomings of simple pairwise predator–prey interaction experiments, the convergence of these two fields of study is slowly beginning to be realized. In the present study, the incorporation of multiple prey densities over a single prey density approach resulted in a more robust determination of potential multiple predator effects. This was particularly evident in the multispecies combinations whereby in contrast to the overall functional response data, at certain prey densities no differences between observed and expected multiple predator effects would have been detected, despite the lack of 100% prey consumption at those densities. Similarly, under certain single prey densities, differences were evident between observed and expected overall consumption in multipredator combinations, contrary to the overall functional response derived outputs. The use of a functional response approach therefore provides an added degree of confidence in results over the single prey density MPE approach, given that the design requires the use of many predators thus minimizing random effects. Furthermore, a major advantage to using a functional response approach to assessing predator–prey interactions is seen in the attack rate parameter, which provides information into prey resource utilization at lower prey densities and is important for the determination of functional response type. While in the present study a type II functional response was observed in all trials, other types of responses are possible. Functional response type is thought to have implications for prey population persistence or the point at which prey-switching by a predator occurs (Hassell 1978; Abrams 1982). The incorporation of this aspect of predation into MPE studies is lacking and using a functional response approach in combination with behavioural observations, as in the present study, could add insight into our understanding of predator induced prey extinctions or Allee effects.

The next step in integrating functional responses into the MPE literature would be to develop a predictive framework whereby multiple species with various trait suites are trialled. To do this, future studies will need to use conceptual models (Fig. 5) to identify the factors that may alter predator–prey dynamics at each step. These factors can

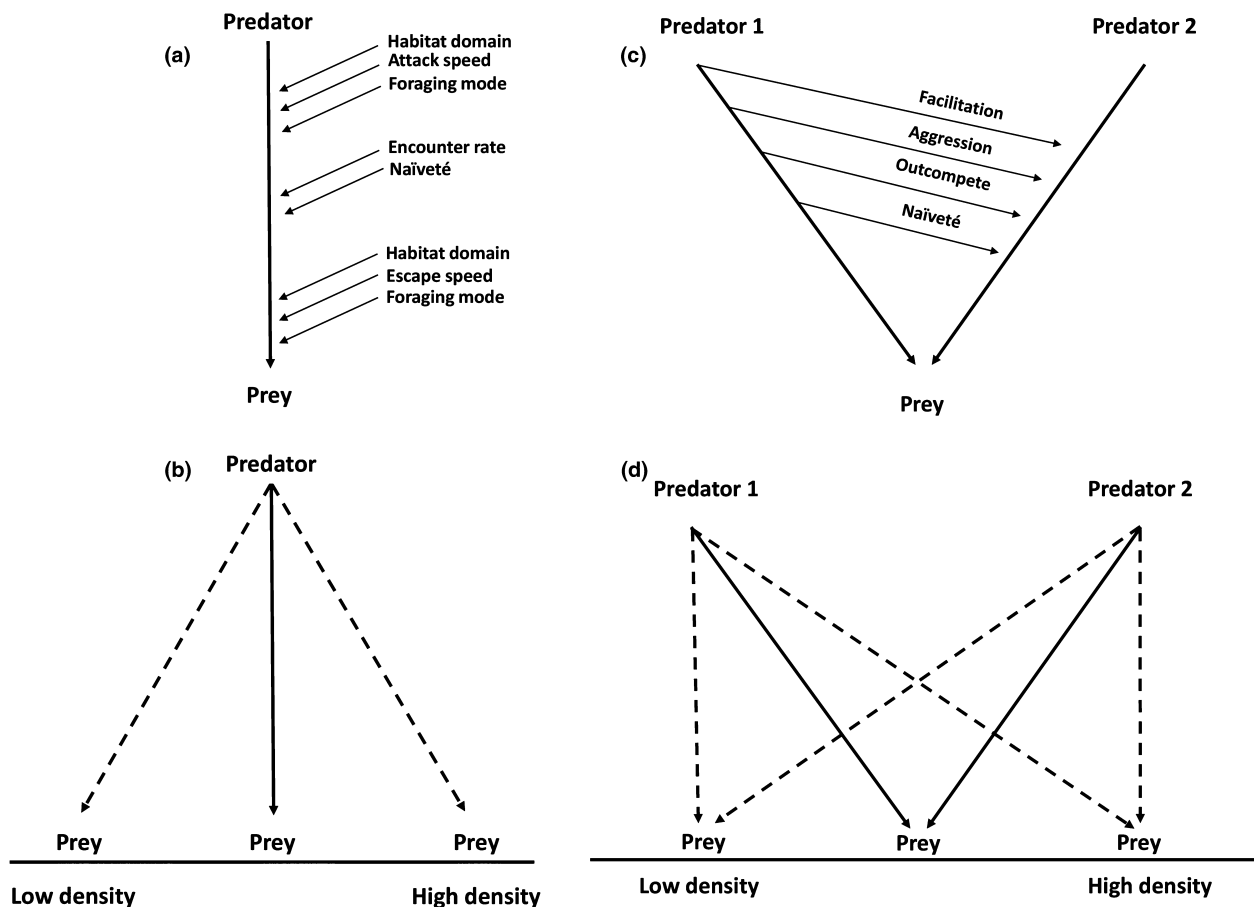


Fig. 5. Conceptual models outlining example factors affecting predator–prey interactions when considering (a) simple pairwise interaction between a single predator and prey (at fixed density), (b) pairwise interactions between a single predator and prey at multiple prey densities (functional response) (c) interaction between predator and prey as mediated by the presence of an additional predator (multiple predator effect) and (d) interaction between predator and prey at multiple prey densities as mediated by the presence of an additional predator. Bold solid lines represent interactions between predators and prey. Thin solid lines represent factors that influence predator–prey interaction outcomes. Hashed lines represent shifting predator–prey interaction outcomes in response to changes in prey densities. Note that factor examples from model (a) (habitat domain, attack speed, naïveté) are still present in models (b), (c) and (d) as are model (c) factors (facilitation, aggression, interference, distract/ court, naïveté) present in model (d). Models (c) and (d) are relevant at both the conspecific and heterospecific multipredator level and factors highlighted in (c) are contingent on species traits.

then be associated with species traits. As such, species selection should be based on available autecological information in order for traits such as habitat domain, territoriality, reliance on visual or olfactory cues and foraging mode to be incorporated. In the present study, we only used three species, and much of this information is not available for tilapia and mouthbrooder. This makes it difficult to develop such a framework using this data. The study does, however, provide a working example of how such studies could be conducted to provide insight into how key traits can be highlighted for predictive capacity regarding MPE outcomes. For example in the present study, while maintenance of foraging area size was an unavoidable component of the design, this experimental requirement may provide insight into how the importance of space depends on functional traits of the test species. On the one hand, mouthbrooder are solitary and potentially territorial and for this species, space may be important.

Indeed mouthbrooder pairs did not consume prey proportional to its availability, exhibiting overall attack rates and handling times that were lower than even individual functional responses for the species. On the other hand, extreme are bluegill, a shoaling species. Shoaling species are less likely to be threatened by conspecifics in close proximity as this is the mode in which they typically forage (Wahl & Stein 1988; Savino & Stein 1989). Thus, bluegill pairs exhibited functional responses as expected based on individual performance. Space therefore seems to be less of a factor affecting prey resource utilization in bluegill. In the context of this study, such statements are largely speculative given the limited number of species tested and the lack of sufficient species information such as mouthbrooder and tilapia habitat domains (Schmitz 2007). However, future studies on larger datasets incorporating more species could result in the development of a trait-based framework with potential predictive capacity.

Within the context of developing a trait-based framework, prey species traits would also need to be incorporated as prey are often not passive players in predator–prey interactions (Schmitz 2007; Dodd *et al.* 2014; Wasserman *et al.* 2016a). In the present study, early life-history fish prey were employed. These prey were active swimmers and would likely have been capable of a degree of escape, more so than slower swimming prey such as certain species of tadpole, for example. Prey selection is therefore an important consideration in any predator–prey experimental study as prey species may respond differently to the same predator (Schmitz 2007). The development of a predictive framework using a functional response approach would advance the field of trophic ecology within the context of predator diversity effects given that functional responses, in theory, account for predator performance with and without prey as a limited resource. Such information would have various ecological applications. One example would be for the prioritization of invasive species management. Functional response studies have gained much momentum in the field of invasion biology, as it has been highlighted that invasive species often have elevated and different functional responses to similar native species (Dick *et al.* 2013a, 2014; Alexander *et al.* 2014; Barrios-O'Neill *et al.* 2015). As such, it has been proposed that functional responses could be used as a tool to identify problematic invasive species or used in combination with other information to potentially quantify impact (Alexander *et al.* 2014; Dick *et al.* 2014). There is, however, limited information on the role of competition and predator–predator interactions within the context of invasion biology for functional response studies. This is relevant as the invasion meltdown theory predicts that one invasive species could facilitate a second invader (Simberloff & Von Holle 1999; Jackson 2015), while biotic resistance theory specifies that certain species facilitate immunity to further invasions (DeRivera *et al.* 2005; Dick *et al.* 2013b). The incorporation of functional responses into MPE studies therefore has much potential for the field of invasion biology.

Simplification is key to the development of ecological theory and functional response procedures offer relatively straightforward means of exploring aspects of optimal foraging (Abrams 1982). While laboratory studies are often criticized for having limited field relevance, they are still important for the development of ecological theory as they provide insight into specific aspects of ecology (Lawton 1995; Chapman 2000; Barrios-O'Neill *et al.* 2015). Controlled experiments are often one of the only ways with which we can gain a mechanistic understanding of how certain processes unfold (Benton *et al.* 2007; Alexander *et al.* 2012). We therefore propose that the incorporation of predator combinations into classic functional response investigations would be useful for the development of competition and predation ecology. Notably, this approach can be utilized in a comparative fashion in regional, taxonomic and phenotypic, invasion or size structuring predation assessments with the ultimate goal of enhancing

predictive capacity development for multipredator aspects of predator–prey dynamics.

Acknowledgements

This work was financially supported by the National Research Foundation of South Africa (NRF, UID: 77444, 88746) and the Department of Science and Technology/NRF Centre of Excellence in Invasion Biology (CIB). Gratitude is extended to Phumza Ndalen, Emiel Roels and Rachel Ndhlovu for field assistance. The Department of Economic Development, Environmental Affairs and Tourism (Cacadu Region) is thanked for issuing research permits (11/15CR & 12/15CR).

Data accessibility

The data are deposited in the Dryad Digital Repository: <http://dx.doi.org/10.5061/dryad.4v2s0> (Wasserman *et al.* 2016b).

References

- Abrams, P.A. (1982) Functional responses of optimal foragers. *American Naturalist*, **120**, 382–390.
- Abrams, P.A. (2000) The evolution of predator–prey interactions: theory and evidence. *Annual Review of Ecology and Systematics*, **31**, 79–105.
- Alexander, M.E., Dick, J.T., O'Connor, N.E., Haddaway, N.R. & Farnsworth, K.D. (2012) Functional responses of the intertidal amphipod *Echinogammarus marinus*: effects of prey supply, model selection and habitat complexity. *Marine Ecology Progress Series*, **468**, 191–202.
- Alexander, M.E., Dick, J.T., Weyl, O.L., Robinson, T.B. & Richardson, D.M. (2014) Existing and emerging high impact invasive species are characterized by higher functional responses than natives. *Biology Letters*, **10**, 20130946.
- Anderson, T.L. & Whiteman, H.H. (2015) Non-additive effects of intra- and interspecific competition between two larval salamanders. *Journal of Animal Ecology*, **84**, 765–772.
- Barrios-O'Neill, D., Dick, J.T.A., Emmerson, M.C., Ricciardi, A., MacIsaac, H.J., Alexander, M.E. *et al.* (2014) Fortune favours the bold: a higher predator reduces the impact of a native but not an invasive intermediate predator. *The Journal of Animal Ecology*, **83**, 693–701.
- Barrios-O'Neill, D., Dick, J.T.A., Emmerson, M.C., Ricciardi, A. & MacIsaac, H.J. (2015) Predator-free space, functional responses and biological invasions. *Functional Ecology*, **29**, 377–384.
- Benton, T.G., Solan, M., Travis, J.M.J. & Sait, S.M. (2007) Microcosm experiments can inform global ecological problems. *Trends in Ecology & Evolution*, **22**, 516–521.
- Bolker, B.M. (2008) *emdbook: Ecological Models and Data in R*. Princeton University Press, Princeton, NJ, USA.
- Bolker, B., Holyoak, M., Krivan, V., Rowe, L. & Schmitz, O. (2003) Connecting theoretical and empirical studies of trait-mediated interactions. *Ecology*, **84**, 1101–1114.
- Bruton, M.N. (1978) The habitats and habitat preferences of *Clarias gariepinus* (Pisces: Clariidae) in a clear coastal lake (Lake Sibaya, South Africa). *Journal of the Limnological Society of Southern Africa*, **4**, 81–88.
- Chapman, M.G. (2000) Poor design of behavioural experiments gets poor results: examples from intertidal habitats. *Journal of Experimental Marine Biology and Ecology*, **250**, 77–95.
- Colgan, P.W., Nowell, W.A., Gross, M.R. & Grant, J.W. (1979) Aggressive habituation and rim circling in the social organization of bluegill sunfish (*Lepomis macrochirus*). *Environmental Biology of Fishes*, **4**, 29–36.
- DeRivera, C.E., Ruiz, G.M., Hines, A.H. & Jivoff, P. (2005) Biotic resistance to invasion: native predator limits abundance and distribution of an introduced crab. *Ecology*, **86**, 3364–3376.
- Dick, J.T., Gallagher, K., Avlijas, S., Clarke, H.C., Lewis, S.E., Leung, S. *et al.* (2013a) Ecological impacts of an invasive predator explained and predicted by comparative functional responses. *Biological Invasions*, **15**, 837–846.
- Dick, J., MacNeil, C., Alexander, M., Dodd, J. & Ricciardi, A. (2013b) Predators vs. alien: differential biotic resistance to an invasive species by two resident predators. *NeoBiota*, **19**, 1–19.
- Dick, J.T.A., Alexander, M.E., Jeschke, J.M., Ricciardi, A., MacIsaac, H.J., Robinson, T.B. *et al.* (2014) Advancing impact prediction and

- hypothesis testing in invasion ecology using a comparative functional response approach. *Biological Invasions*, **16**, 735–753.
- Dodd, J.A., Dick, J.T., Alexander, M.E., MacNeil, C., Dunn, A.M. & Aldridge, D.C. (2014) Predicting the ecological impacts of a new freshwater invader: functional responses and prey selectivity of the 'killer shrimp', *Dikerogammarus villosus*, compared to the native *Gammarus pulex*. *Freshwater Biology*, **59**, 337–352.
- Duffy, J.E., Richardson, J.P. & France, K.E. (2005) Ecosystem consequences of diversity depend on food chain length in estuarine vegetation. *Ecology Letters*, **8**, 301–309.
- Dugatkin, L.A. & Wilson, D.S. (1992) The prerequisites for strategic behaviour in bluegill sunfish, *Lepomis macrochirus*. *Animal Behaviour*, **44**, 223–230.
- Ellender, B.R. & Weyl, O.L.F. (2014) A review of current knowledge, risk and ecological impacts associated with non-native freshwater fish introductions in South Africa. *Aquatic Invasions*, **9**, 117–132.
- Forrester, G.E., Evans, B., Steele, M.A. & Vance, R.R. (2006) Assessing the magnitude of intra- and interspecific competition in two coral reef fishes. *Oecologia*, **148**, 632–640.
- Griffen, B.D. (2006) Detecting emergent effects of multiple predator species. *Oecologia*, **148**, 702–709.
- Griffin, J.N., Byrnes, J.E. & Cardinale, B.J. (2013) Effects of predator richness on prey suppression: a meta-analysis. *Ecology*, **94**, 2180–2187.
- Gross, M.R. & MacMillan, A.M. (1981) Predation and the evolution of colonial nesting in bluegill sunfish (*Lepomis macrochirus*). *Behavioral Ecology and Sociobiology*, **8**, 163–174.
- Harvey, B.C., White, J.L. & Nakamoto, R.J. (2004) An emergent multiple predator effect may enhance biotic resistance in a stream fish assemblage. *Ecology*, **85**, 127–133.
- Hassell, M.P. (1978) A basic model. *The Dynamics of Arthropod Predator-Prey Systems* (ed. M. Hassell), pp. 12–27. Princeton University Press, Princeton, NJ, USA.
- Holling, C. (1959) The components of predation as revealed by a study of small-mammal predation of the European Pine Sawfly. *The Canadian Entomologist*, **91**, 293–320.
- Ives, A.R., Cardinale, B.J. & Snyder, W.E. (2005) A synthesis of subdisciplines: predator–prey interactions, and biodiversity and ecosystem functioning. *Ecology Letters*, **8**, 102–116.
- Jackson, M.C. (2015) Interactions among multiple invasive animals. *Ecology*, **96**, 2015–2041.
- Jeschke, J., Kopp, M. & Tollrian, R. (2002) Predator functional responses: discriminating between handling and digesting prey. *Ecological Monographs*, **72**, 95–112.
- Johnson, P.T.J., Olden, J.D., Solomon, C.T. & Vander Zanden, M.J. (2009) Interactions among invaders: community and ecosystem effects of multiple invasive species in an experimental aquatic system. *Oecologia*, **159**, 161–170.
- Juliano, S.A. (2001) Nonlinear curve fitting: predation and functional response curves. *Design and Analysis of Ecological Experiments*, 2nd edn (eds S.M. Scheiner & J. Gurevitch), pp. 178–196. Oxford University Press, Oxford, UK.
- Khoza, Z.C., Potgieter, M.J. & Vlok, W. (2012) A preliminary survey of biotic composition of the Olifantspruit catchment, South Africa. *African Journal of Aquatic Science*, **37**, 201–208.
- Lawton, J.H. (1995) Ecological experiments with model systems. *Science*, **269**, 328–331.
- Lohrer, A.M. & Whitlatch, R.B. (2002) Interactions among aliens: apparent replacement of one exotic species by another. *Ecology*, **83**, 719–732.
- Losey, J.E. & Denno, R.F. (1998) Positive predator–predator interactions: enhanced predation rates and synergistic suppression of aphid populations. *Ecology*, **79**, 2143–2152.
- Marshall, B. (2011) *The fishes of Zimbabwe and their biology*. *Smithiana Monograph No. 3*. South African Institute for Aquatic Biodiversity, Grahamstown, South Africa.
- Muller, C., Weyl, O.L.F. & Strydom, N.A. (2015) Introduction, establishment and spread of the Southern mouthbrooder *Pseudocrenilabrus philander* in the Baakens River, Eastern Cape, South Africa. *African Zoology*, **50**, 259–262.
- Naem, S., Duffy, J.E. & Zavaleta, E. (2012) The functions of biological diversity in an age of extinction. *Science*, **336**, 1401–1406.
- Polis, G.A., Myers, C.A. & Holt, R.D. (1989) The ecology and evolution of intraguild predation: potential competitors that eat each other. *Annual Review of Ecology, Evolution, and Systematics*, **20**, 297–330.
- Polling, L., Schoonbee, H.J. & Saayman, J.E. (1995) Observations on aspects of the biology of *Pseudocrenilabrus philander* (Weber, M., 1897) from a subtropical South-African impoundment. *Water SA*, **21**, 371–378.
- Porter-Whitaker, A.E., Rehage, J.S., Liston, S.E. & Loftus, W.F. (2012) Multiple predator effects and native prey responses to two non-native Everglades cichlids. *Ecology of Freshwater Fish*, **21**, 375–385.
- Pritchard, D.W. (2014) frair: a package for functional response analysis in R.
- R Development Core Team. (2013) *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria. <http://www.R-project.org>.
- Ribbink, A.J. (1971) The behaviour of *Hemihaplochromis philander*, a South African cichlid fish. *Zoologica Africana*, **6**, 263–288.
- Rogers, D. (1972) Random search and insect population models. *The Journal of Animal Ecology*, **41**, 369–383.
- Sala, O.E., Chapin, F.S., Armesto, J.J., Berlow, E., Bloomfield, J., Dirzo, R. et al. (2000) Global biodiversity scenarios for the year 2100. *Science*, **287**, 1770–1774.
- Savino, J.F. & Stein, R.A. (1989) Behavioural interactions between fish predators and their prey: effects of plant density. *Animal Behaviour*, **37**, 311–321.
- Schmitz, O.J. (2007) Predator diversity and trophic interactions. *Ecology*, **88**, 2415–2426.
- Sih, A., Englund, G. & Wooster, D. (1998) Emergent impacts of multiple predators on prey. *Trends in Ecology & Evolution*, **13**, 350–355.
- Simberloff, D. & Von Holle, B. (1999) Positive interactions of nonindigenous species: invasional meltdown? *Biological Invasions*, **1**, 21–32.
- Skelton, P.H. (2002) *A Complete Guide to the Freshwater Fishes of Southern Africa*. Struik, Cape Town, South Africa.
- Smith, M.F. & Warburton, K. (1992) Predator shoaling moderates the confusion effect in blue-green chromis, *Chromis viridis*. *Behavioral Ecology and Sociobiology*, **30**, 103–107.
- Sokol-Hessner, L. & Schmitz, O.J. (2002) Aggregate effects of multiple predator species on a shared prey. *Ecology*, **83**, 2367–2372.
- Soluk, D.A. (1993) Multiple predator effects: predicting combined functional response of stream fish and invertebrate predators. *Ecology*, **74**, 219–255.
- Taguchi, T., Miura, Y., Krueger, D. & Sugiura, S. (2014) Utilizing stomach content and faecal DNA analysis techniques to assess the feeding behaviour of largemouth bass *Micropterus salmoides* and bluegill *Lepomis macrochirus*. *Journal of Fish Biology*, **84**, 1271–1288.
- Vonesh, J.R. & Osenberg, C.W. (2003) Multi-predator effects across life-history stages: non-additivity of egg- and larval-stage predation in an African treefrog. *Ecology Letters*, **6**, 503–508.
- Wahl, D.H. & Stein, R.A. (1988) Selective predation by three esocids: the role of prey behavior and morphology. *Transactions of the American Fisheries Society*, **117**, 142–151.
- Wasserman, R.J., Alexander, M.E., Weyl, O.L.F., Barrios-O'Neill, D., Froneman, P.W. & Dalu, T. (2016a) Emergent effects of structural complexity and temperature on predator–prey interactions. *Ecosphere*, **7**, e01239.
- Wasserman, R., Alexander, M., Dalu, T., Ellender, B., Kaiser, H. & Weyl, O. (2016b) Data from: Using functional responses to quantify interaction effects among predators. *Dryad Digital Repository*, doi:10.5061/dryad.4v2s0.
- Weigelt, A., Schumacher, J., Walther, T., Bartelheimer, M., Steinlein, T. & Beyschlag, W. (2007) Identifying mechanisms of competition in multi-species communities. *Journal of Ecology*, **95**, 53–64.
- Werner, E.E. & Hall, D.J. (1988) Ontogenetic habitat shifts in bluegill: the foraging rate-predation risk trade-off. *Ecology*, **69**, 1352–1366.
- Yonekura, R., Kohmatsu, Y. & Yuma, M. (2007) Difference in the predation impact enhanced by morphological divergence between introduced fish populations. *Biological Journal of the Linnean Society*, **91**, 601–610.
- Young, K.A. (2004) Asymmetric competition, habitat selection, and niche overlap in juvenile salmonids. *Ecology*, **85**, 134–149.
- Zengeya, T.A. & Marshall, B.E. (2007) Trophic interrelationships amongst cichlid fishes in a tropical African reservoir (Lake Chivero, Zimbabwe). *Hydrobiologia*, **592**, 175–182.

Received 31 October 2015; accepted 22 April 2016

Handling Editor: James Harwood