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Microplastics do not affect the feeding rates of a marine predator

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Author contributions

EMC, NEC, LK and JTAD designed the study. EMC and VC collected and maintained the animals in the laboratory and carried out the functional response experiments. EMC and RNC conducted the analysis. All authors contributed to the writing, which was led by EMC.

Abstract

Microplastics may affect the physiology, behaviour and populations of aquatic and terrestrial fauna through many mechanisms, such as direct consumption and sensory disruption. However, the majority of experimental studies have employed questionably high dosages of microplastics that have little environmental relevance. Predation, in particular, is a key trophic interaction that structures populations and communities and influences ecosystem functioning, but rarely features in microplastic research. Here, we quantify the effects of low (~65-114 MP/L) and high (~650-1140 MP/L) microplastic concentrations on the feeding behaviour of a ubiquitous and globally representative key marine predator, the shore crab, *Carcinus maenas*. We used a functional response approach (predator consumption across prey densities) to determine crab consumption rates towards a key marine community prey species, the blue mussel *Mytilus edulis,* under low and high microplastic concentrations with acute (8h) and chronic (120 h) microplastic exposure times. For both the acute and chronic microplastic exposure experiments, proportional prey consumption by crabs did not differ with respect to microplastic concentration, but significantly decreased over increasing prey densities. The crabs thus displayed classical, hyperbolic Type II functional responses in all experimental groups, characterised by high consumption rates at low prey densities. Crab attack rates, handling times and maximum feeding rates (ie functional response curves) were not significantly altered under lower or higher microplastics concentrations, or by acute or chronic microplastic exposures. Here, we show that functional response analyses could be widely employed to ascertain microplastic impacts on consumer-resource interactions. Furthermore, we suggest that future studies should adopt both acute and chronic microplastic exposure regimes, using environmentally-relevant microplastic dosages and types as well as elevated future scenarios of microplastic concentrations.

Keywords:

Animal behaviour; blue mussel; functional response; pollutants; shore crab; synthetic polymers

1. Introduction

The prevalence and persistence of plastic pollution in all environments globally has become one of the most topical environmental issues since the beginning of the 21st century. Global production levels of plastic are now exceeding values of 350 million tons per year (Plastics Europe, 2020), and the input of plastic waste into the global oceans is estimated to be between 4.8 and 12.7 million tons on an annual basis (Jambeck et al. 2015). More recently, the ubiquity of microplastic pollution (<5 mm; Thompson et al. 2004) in marine systems has caused global concern among scientists and the public alike (Cunningham et al. 2020). Termed as either industry-made primary microplastics, or secondary fragmented particles from larger macro-sized items (Thompson et al. 2004), microplastics have been found in nearly every imaginable marine habitat, from transitional estuarine environments to deep-sea trenches (Welden & Lusher, 2017). Microplastics have also been shown to be ingested by a range of marine-dwelling fauna at different trophic levels, from zooplankton (Zheng et al. 2020) and decapods (*Nephrops norvegicus*; Hara et al. 2020), to fish (*Chelon aurata*; Zakeri et al. 2020), birds (*Branta leucopsis*; Coughlan et al. 2020), turtles (*Caretta caretta*; Duncan et al. 2019) and marine mammals (*Delphinus capensis*; Nelms et al. 2019).

While in recent years the ubiquity of microplastics in the environment, and ingestion by marine animals, has been described extensively, the potential physiological and behavioural impacts these micro-sized particles can have are much more complex than just quantifying the amount of particles in a given environment. Microplastic pollution has been shown to negatively affect the physiology of many marine species in laboratory studies (Franzellitti et al. 2019), with impacts ranging from organ damage to mortality. In contrast, the behavioural impacts have only been assessed for a limited number of fauna, including habitat selection in the purple shore crab (Prestholdt & Kemp, 2020), disrupted cognition and reduced shell selection in the European hermit crab (Crump et al. 2020), and reduced swimming velocity in European seabass (Barboza et al. 2018). In both the range of behavioural and physiological impacts found during microplastic exposure trials, the severity of impacts have been driven by factors such as, polymer type, shape, size, and dosage (Bucci et al. 2020).

Although the range of surface water values of microplastic pollution are estimated between 0-100 mps/L (Burns & Boxall, 2018), a number of experimental studies have utilised values 1000-fold higher. Recently, Cunningham & Sigwart (2019) highlighted that 82% (105/128) of microplastic studies utilise higher amounts of microplastics on marine animals than what is found in the environment. Few studies have adopted more realistic dosages for microplastics in exposure experiments, which would provide a more accurate representation of the impacts of microplastics. For example, environmentally-relevant levels of microplastic fibres (5 fibres/1.5 g feed) have been shown to become lodged in the digestive system of *Nephrops norvegicus* which has led to a reduction in body mass (Welden & Cowie, 2016). Further to this, microplastic fibres at low dosages (3 fibres/L) have also been shown to reduce the growth of Pacific mole crabs (*Emerita analoga*; Horn et al. 2020). Environmental levels of microplastics found in Burns & Boxall (2018) have also been used in behavioural studies. A previous study demonstrated that the emergence behaviour of the intertidal gastropod *Littorina littorea* was not affected by microplastic levels within its habitat (~0.68 mps/individual; Doyle et al. 2020). Additionally, the duration of microplastic exposure has shown to drive the level of impacts on marine animals (Anbumani & Kakkar, 2018). For example, increasing the exposure time has been shown to affect the cognition of hermit crabs during a chronic 5-day exposure to low dosages of polyethylene particles (25 mps/L) and, therefore, impact upon one of their basic survival behaviours (i.e. shell selection, Crump et al. 2020).

Predation is a fundamental biotic process that influences the structuring and functioning of ecosystems globally (Sih et al. 1985). Functional responses have been used to determine relationships between predation rates and prey densities and are a critical tool in quantifying interaction strengths throughout ecology (Holling, 1959; Dick et al. 2014). More recently, functional responses have been used in microplastic studies to determine the uptake rates of particles at a range of microplastic densities by fishes (Mbedzi et al. 2020), as well as the effects of microplastic exposure on prey, and how this impacts predator feeding rates in freshwater systems (Cuthbert et al. 2019). While previous studies have only considered a single prey density (e.g. Cole et al. 2015), the functional response approach can provide greater insights into how microplastics affect predator-prey interaction strengths across a range of prey densities (Cuthbert et al. 2019). Hence, functional responses could be used to discern how the feeding efficiency of predators is affected by microplastic exposure, and in turn, predict population stability implications (Dick et al. 2014).

Predatory crabs are ubiquitous in marine environments, and given the rate of consumption in comparison to other animal groups (i.e. echinoderms), crabs have been described as keystone predators in certain circumstances due to their influence on ecosystem functionality (Hull & Bourdeau, 2017). Mussel reefs, which are commonly consumed by predatory crabs (Joyce et al. 2019), are a critically important habitat that support a wide range of biodiversity and provide refuge for a variety of juvenile species (Mascaró & Seed, 2001). While high levels of predation can homogenise reefs and reduce overall biodiversity (Hollebone & Hay, 2007), predators may also be a stabilising force in communities by regulating population levels.

Hence, the effects of microplastic pollution on this key trophic interaction need to be examined. In this study, we aimed to determine whether low and high levels of microplastic pollution would affect the feeding behaviour of a common shoreline crustacean, *Carcinus maenas.* Given the deleterious effects of microplastics reported in other study systems, we expected that exposure to microplastics would reduce the intensity of predator-prey interactions between crabs and mussels. We fed blue mussel (*Mytilus edulis*) in increasing densities to *Carcinus maenas* and assessed the functional responses of the predator in two separate experiments. Firstly, we adopted an 8 hour-exposure of microplastics as an acute treatment, as this duration was previously used to assess the feeding rates of shore crabs and mussel prey (Joyce et al. 2019). Secondly, a five-day chronic exposure to microplastics was used as behavioural impacts were found in hermit crabs for this duration (Crump et al. 2020). We employed the use of polyethylene particles in an effort to make our study relevant to the environment with standardised particle types, but without using microplastic fibres, which are the most prevalent in the environment (de Haan et al. 2019).

2. Methods

2.1. Animal collection and husbandry

Shore crabs (*Carcinus maenas*) were collected between July and August 2020 using herring-baited crab pots at the Ballyhenry shore, Strangford Lough, Northern Ireland, UK (54° 23' 36.6576" N, 5° 34' 27.7356" W), and maintained in outdoor ~500 L tanks at the Queen's University Marine Laboratory (QML), Portaferry. A constant flow of seawater was pumped from the adjacent Strangford Lough into the tanks using a flow-through, sand-filtered seawater system (13 ± 1 °C). The tanks were covered with a thin mesh to ensure animals could not escape. All crabs were fed herring (*Clupea* sp.) bait every three days and maintained for a minimum of one week prior to the start of experiments which ran from July to August 2020. The prey, blue mussel, *Mytilus edulis*, were collected from Horse Island, Strangford Lough (54° 27' 29.0736" N, 5° 33' 2.5956" W), and were also maintained under the same flow-through seawater system in 5 L buckets at QML. For consistency and comparability across treatments, crabs with a carapace width of 60-70 mm, and mussels with shell lengths of 15-21 mm, were used for the experiments. These size ranges had been used previously in a study that calculated the feeding rates of shore crabs towards blue mussel (Joyce et al. 2019). Additionally, only male crabs were used to avoid any effects of sex and reproductive status on predator-prey interactions, and only crabs that were free of parasites and had both chelipeds intact were used in the experiments.

2.2 Microplastic preparation

To test the effects of microplastic exposure on our predator prey interactions, a 5 L solution of seawater with microplastic particles was prepared, i.e. 0.1 g of ultra-high molecular weight, surface-modified multi coloured polyethylene powder, 40-48 μm particle size (Sigma-Aldrich, UK). The densities for each microplastic treatment were as follows: no added microplastics (i.e. control); 65.98 – 114.02 microplastics per litre (i.e. MPL); 659.84 – 1140.25 microplastics per litre (i.e. MPH). The MPL densities were used to represent low numbers of microplastics identified from marine sediment samples (Cunningham & Sigwart, 2019) and the highest known surface water values (100 microplastics/L; Burns & Boxall 2018). We used the MPH treatment to assess the effects of an elevated (e.g. future) microplastic concentration, in comparison to the low MPL treatment. Aeration was used to aid the distribution of microplastic particles and reduce the settlement on the bottom of the tanks. Although microplastic fibres are the most prominent type found in the environment, polyethylene powder has been used as a proxy for environmental microplastics in the past (Mbedzi et al. 2020). Background levels of microplastic pollution from our animal sampling site are low (1 MP/L; Green et al. 2018) and were therefore not considered during the exposure trials. Nevertheless, the crab and mussel species examined in this study have a widespread, pancontinental distribution, and therefore are likely to be exposed to a broad range of microplastic concentrations outside the immediate sampling location.

2.3 Experimental design

We tested the effects of our three microplastics exposure concentration groups (control/MPL/MPH) in two separate experiments. The first experiment consisted of both the predator and prey exposed to three microplastic treatments (see below for densities of microplastic) for an acute exposure of eight hours during the functional response feeding trial. A second, separate experiment consisted of a chronic five day exposure to three microplastic treatments, where only the crabs were exposed to the microplastic treatments prior to the beginning of the feeding trial (e.g Crump et al. 2020). This chronic microplastic exposure period was then followed with a functional response feeding trial over an eight hour period with no further microplastic exposure. Each experiment (i.e., acute or chronic exposures) thus considered two effects in a cross-factorial design: microplastics exposure concentration (3 levels) and prey density (6 levels; see below).

To assess crab functional responses, all feeding trials were carried out in circular 5 L arenas (diameter = 22 cm, depth = 17 cm) containing 4 L of aerated and sand-filtered seawater in a constant temperature room (12 °C). The blue mussels were acclimated to the constant temperature room 16 hours prior to both experiments, which also ensured attachment to the base of the arena with their byssus threads (Joyce et al. 2019). The crabs were acclimated for 16 hours and five days, for the acute and chronic exposures, respectively. We note that although different acclimation times were used for these two exposures, they were considered as separate experiments and analysed in different models (see below). Each arena was covered with mesh to prevent the crabs from escaping. The prey, blue mussels, were added to individual arenas at six densities (2, 4, 8, 16, 32, and 64) and each of the three experimental groups was replicated three times [i.e., 3 (microplastics exposure concentrations) × 6 (densities) × 3 (replicates) = 54 arenas for each of the two experiments). All crabs were starved for 48 hours prior to the feeding trials to standardise hunger levels. Each trial started at 09:00, when crabs were added to the arenas, with crabs allowed to subsequently feed for eight hours. The crabs were then removed and the remaining live mussels were counted in each arena. One additional replicate was performed for the acute experiment without a predator to assess any prey background mortality under each microplastics exposure concentration and prey density.

2.4 Statistical analyses

All statistical analyses were computed in R v3.4.4 (R Core Development Team 2018). Two separate generalised linear models (GLM) were used to examine the proportion of available prey consumed from the acute and chronic exposure experiments. In both models, ‘microplastics concentration’ and ‘prey density’ were included as single and interacting terms. Both GLMs assumed a quasi-binomial distribution, to account for overdispersion in the proportional data (proportions of available prey consumed), given that residual deviance exceeded degrees of freedom. In each model, we specified the proportions of prey eaten versus alive as the response variable (the sum of which corresponds to the initial prey density). We determined the coefficient of determination (R^2) for both GLMs using the ‘rsq’ package in R (Zhang, 2020). F-tests were used to infer main effect sizes from the resulting GLM outputs with Type III sums of squares using the ‘car’ package in R (Fox and Weisberg, 2019).

Functional responses were modelled using the ‘frair’ package in R (Pritchard et al. 2017). This procedure followed two steps. Firstly, for each exposure concentration (C/MPL/MPH) within both experiments (i.e. acute/chronic), logistic regression was used to analyse the proportional consumption of prey in relation to ‘prey density’ to categorise the functional response type (i.e. Types I, II or III). Type II and III functional responses are common in invertebrate predators, whilst Type I functional responses are mechanistically reserved to filter feeding organisms (Jeschke et al. 2004). Here, a Type II functional response was statistically indicated by a significantly negative first order term (i.e. linear coefficient), whereas a Type III functional response is inferred from a significantly positive first order term followed by a significantly negative second order term (i.e. quadratic coefficient). Ecologically, a Type II functional response is characterised by a high rate of resource consumption at low resource densities, which then reaches a plateau. Whereas, a Type III functional response exhibits a low rate of consumption at low densities, which then increases initially before again reaching a plateau. As such, the Type II functional response may be more likely to extirpate populations due to a lack of low density refuge, compared to the Type III functional response where a low density refuge is theoretically imparted to populations (Dick et al. 2014).

Secondly, as prey were not replaced as they were consumed, and owing to the functional response type we found, we fitted Rogers’ random predator equation (Rogers, 1972), which is appropriate for instances of prey depletion (Cuthbert et al. 2020):

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

where N_e is the number of prey killed, N_0 is the initial density of prey, a is the attack rate, h is the handling time and T is the total experimental period. Both a and h are parameters of the functional response curve, whereby *a* corresponds to the search efficiency of predators at low densities (i.e. initial curve slope) and *h* inversely to the maximum feeding rate (i.e. curve asymptote). The random predator equation was fit for each microplastics concentration treatment group using maximum likelihood estimation for both experiments, with the Lambert W function implemented to make the equation solvable (Bolker, 2008). Additionally, to generate 95% confidence intervals around the functional response curve, a non-parametric bootstrapping procedure (n = 2000 iterations) was used (Pritchard et al. 2017). As this procedure allows functional responses to be considered at the population-level as opposed to the sample-level, differences in functional responses were made on the basis of confidence interval divergence across prey densities.

3. Results

In the absence of predators, 100% of blue mussel prey survived throughout the duration of the acute experiment for all treatments, indicating that mortality found during the experiments was a result of predation, rather than microplastics exposure or any other causes. We therefore did not need to adjust feeding rates in experimental groups to account for any background prey mortality.

Microplastics exposure did not significantly affect the proportion of mussels consumed following either acute ($F_{2,48} = 0.292, p = 0.748$) or chronic exposures ($F_{2,48} = 0.219, p = 0.804$; Fig.1). In both experiments, proportional prey consumption decreased significantly with increasing prey densities (acute: $F_{1,48} = 92.944, p < 0.001$; chronic: $F_{1,48} = 43.776, p < 0.001$; Fig.1). The lack of effect of microplastics exposure on the proportion of prey consumed in both experiments was consistent across prey densities, owing to a lack of significant interaction (acute: $F_{2,48} = 0.091, p = 0.913$; chronic: $F_{1,48} = 0.082, p = 0.922$;

Fig.1). The R^2 values for acute and chronic exposure GLMs were 0.719 and 0.572, respectively.

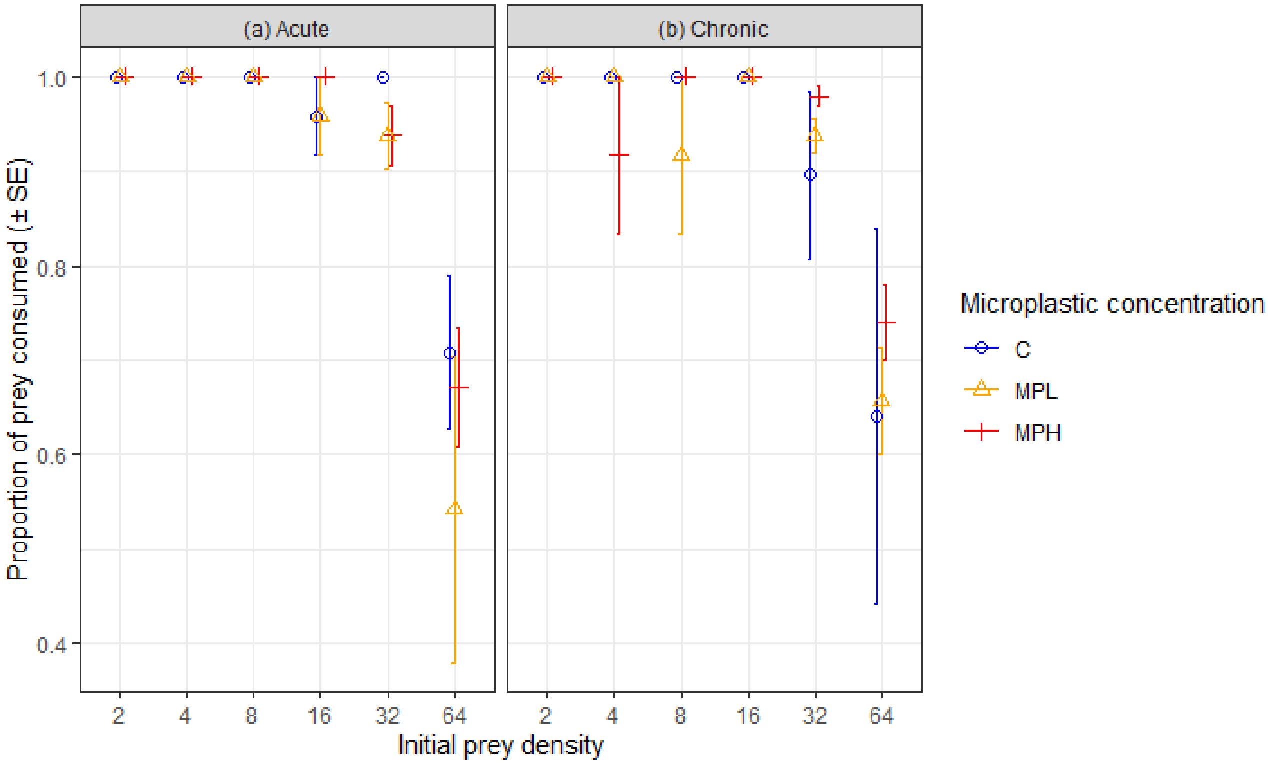

Fig.1: Proportions of *M. edulis* prey consumed across increasing microplastic concentrations (Control/ MPL/ MPH) and prey densities by *C. maenas* for (a) the acute eight-hour microplastic exposure experiment and (b) the chronic five-day microplastic exposure experiment. Means are shown with standard error (SE) (n = 3).

Crabs consistently displayed Type II functional responses irrespective of microplastic concentrations for both acute and chronic exposures, owing to significantly negative first order terms (Table 1). Accordingly, the proportions of available prey eaten were highest at the lower prey densities (Fig.1). Attack rates and handling times were always significantly different to zero and were thus well predicted by the random predator equation (Table 1). Both attack rates and handling times were similar among microplastic concentrations following acute exposures. Likewise, for chronic exposures, attack rates and handling times remained similar among microplastics exposures (Table 1). Maximum feeding rates were slightly highest in the control and high microplastics concentration groups for acute and chronic exposures, respectively (Fig.2). However, functional response confidence intervals always overlapped across prey densities, indicating similarities among feeding rates irrespective of microplastics exposure treatment (Fig.2).

Table 1: First-order terms from logistic regressions of proportional prey consumption for shore crabs *C. maenas* consuming blue mussel *M. edulis* at different microplastics concentrations (Control, MPL; 65.98 – 114.02 MP/L, MPH; 659.84 – 1140.25 MP/L) under acute and chronic microplastic exposure, as well as functional response parameter estimates from the random predator equation.

Treatment	First-order term	*p*-value	FR Type	Attack rate (*a*)	*p*-value	Handling time (*h*)	*p*-value
Acute Control	-0.086	<0.001	II	7.232	<0.001	0.017	<0.001
Acute MPL	-0.075	<0.001	II	7.043	<0.001	0.023	<0.001
Acute MPH	-0.072	<0.001	II	7.514	<0.001	0.020	<0.001
Chronic Control	-0.062	<0.001	II	6.665	<0.001	0.021	<0.001
Chronic MPL	-0.061	<0.001	II	5.308	<0.001	0.018	<0.001
Chronic MPH	-0.075	<0.001	II	6.561	<0.001	0.016	<0.001

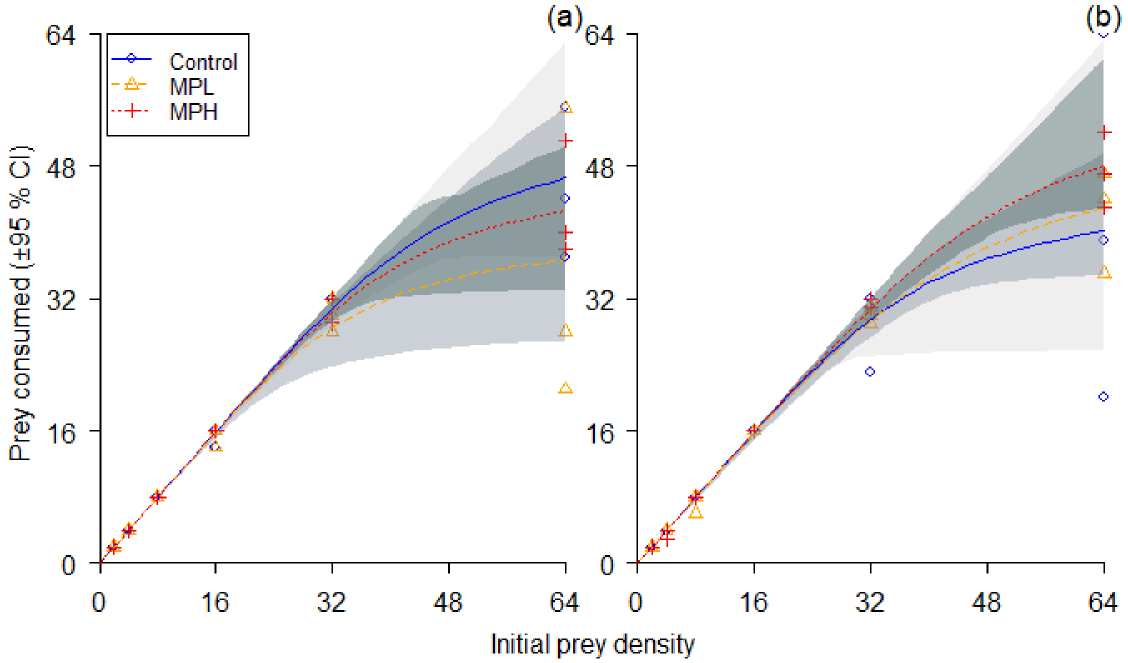

Fig.2: Type II functional responses of shore crabs *C. maenas* consuming blue mussel *M. edulis* during (a) the acute eight-hour exposure and (b) the chronic five-day exposure to microplastics (Control, MPL; 65.98 – 114.02 MP/L, MPH; 659.84 – 1140.25 MP/L). The functional response curves include bootstrapped 95% confidence intervals (*n* = 2000). Points are raw data.

4. Discussion

To our knowledge, this is the first study to quantify how the functional response of marine fauna are affected by the presence of varying dosages of microplastic pollution. Our data show that both low and high levels of microplastics, which have been utilised in previous studies, do not supress the feeding efficiency of shore crabs upon blue mussel prey. We found that prey consumption was significantly driven by initial prey density only, aligning with Type II functional responses, and no other significant effects were detected.

Although the behaviour of the predators in this study was not affected by microplastic exposure, other key animal behaviours aside from predation and feeding have previously been impacted by exposure to microplastic pollution; for example, beachhoppers

(*Platorchestia smithi*) displayed a reduction in locomotive behaviours (Tosetto et al. 2016), reduced cognition was highlighted in hermit crabs (*Pagurus bernhardus*; Crump et al. 2020), and a decrease in social and shoaling behaviours was also found in fish (*Carassius carassius*; Mattsson et al. 2015). Furthermore, while prey consumption was not affected in our study, microplastics may also have negative effects on the reproductive success of individuals (Horn et al. 2020) and may cause organ damage (Barboza et al. 2018), or gut blockage (Watts et al. 2015), which could lead to reduced environmental fitness of individuals and therefore impact upon community structure and diversity (Brown et al. 2016).

It is imperative that future studies adopt more environmentally-realistic microplastic levels, firstly, because a large number of studies use elevated and unrealistic microplastic dosages (Cunningham & Sigwart, 2019); and secondly, because low dosages of microplastics have shown to have an effect on animal behaviour (Crump et al. 2020) and physiology (Horn et al. 2020). Although the number of particles within the low dosage used in this study (MPL; 65.98 – 114.02 MP/L) are in line with values highlighted by Burns & Boxall (2018) as being environmentally-relevant, the use of microplastic fibres as opposed to fragments would have increased the relevance of this study (Horn et al. 2020; Watts et al. 2015; Woods et al. 2018). Previous studies have also utilised secondary microplastics such as fragments or aged particles (Jaikumar et al. 2019) to make the study more relevant to the environment (Lehtiniemi et al. 2018). Also, the site in which we sampled our animals was previously shown to have very low levels of microplastic pollution (~1 MP/L; Green et al. 2018); therefore, it is important that site specific microplastic pollution is considered when trying to adopt an environmentally-relevant approach. It has also been shown that the effects of microplastics at environmental levels may be species-specific (Suckling et al. 2021), and therefore, further studies need to be carried out to highlight which species are most vulnerable. This will help to further our understanding of the impacts microplastic pollution has on marine systems. Likewise, examining how microplastics pollution effects interact with other aspects of global change, such as climatic warming, could help to further improve understandings of multiple stressors associated with human activity.

While the present study assessed a five-day chronic exposure period, which has previously been observed to cause negative behavioural changes to hermit crabs at low microplastic dosages (25 microplastics per litre; Crump et al. 2020), other studies have considered longer durations for chronic exposure. For example, exposure times lasting six weeks have been used to assess behavioural changes in fish (Critchell & Hoogenboom, 2018); although, only slight changes in animal activity were observed during behavioural analysis. Further to this, an exposure period of 95 days highlighted how environmental levels of microplastics affected growth and survivability of fish species due to long-term particle exposure and ingestion (Naidoo & Glassom, 2019). Hence, longer exposure times may be needed to tease out impacts at low or environmentally-relevant dosages of microplastic dosages and types.

Effects of microplastics on trophic interactions may be most profound should they affect keystone predators that govern the abundance and distribution of other species. The shore crab *C. maenas* is a common predator with a broad, pancontinental distribution considering both native and invaded ranges (Howard et al., 2018). Crab predation on mussel reefs maybe seen as a key trophic regulator, as it influences community structuring and biodiversity, because mortality of individual mussels can leave space for other species to colonise or seek refuge (Enderlein & Wahl, 2004; Barrios-O’Neill et al. 2018). Increased predation pressure can have negative effects for community structure as biogenic reefs can become homogenised (Hollebone & Hay, 2007). However, predation on mussel reefs by crabs can be depressed via the introduction of other predators such as whelks, and is also limited by size of the predator, and reef complexity (Quinn et al. 2012). With the introduction of an anthropogenic stressor, we show that low levels of microplastics do not decrease predation rates. In essence, this indicates that mussel population vulnerability to predation may be relatively unaffected by similar types of microplastics pollution to this study under varying concentrations. This corroborates with a freshwater study, whereby predation rates were not impeded by the presence of microplastics-exposed dipteran prey (Cuthbert et al. 2019).

Functional response experiments can be used to assess how interaction strengths are affected by anthropogenic influence; for example, increasing temperature and other climatic variables (Munari et al. 2011; Uiterwaal & DeLong, 2020). Whilst a classical ecological concept that has been pervasive in the ecological literature for decades (Holling, 1959), functional responses have only been applied very recently to quantify effects of microplastics on trophic interactions (Cuthbert et al. 2019). That is despite their predictive power in determining interaction strengths in other ecological fields (e.g. invasion biology, Dick et al. 2014). In contrast, the use of functional responses to determine the interaction strengths of aquatic fauna when exposed to organic and other pollutants dates further back in the literature (e.g. Havens, 1994; Matozzo et al. 2001). Functional response experiments can also be useful tool to assess the uptake of microplastic particles at increasing densities directly, as opposed to prey items (Wood et al., 2018; Mbedzi et al. 2019). A previous study adopting this method showed that fish (*Tilapia sparrmanii*) displayed a Type II functional response when ingesting microplastic particles mixed with fish bait, meaning that fish consumed high numbers of microplastics even at low densities (Mbedzi et al. 2020). Therefore, functional responses can be used to determine how microplastics enter the food chain at a range of densities via numerous trophic levels (Woods et al. 2018). In the present study, as microplastics did not significantly affect feeding rates across resource densities, no evidence for significant effects on attack rates, handling times or maximum feeding rates towards prey were discerned. We also found Type II hyperbolic functional responses across all microplastics treatments, and therefore microplastics exposure did not impede predation efficiencies at low prey densities, with Type II functional response thought to be more destabilising to prey populations than sigmoidal Type III feeding relationships that can impart low density prey refuge (Dick et al. 2014). However, we note that our experiment did not incorporate any habitat structure, which can impart refuge to prey and cause shifts in functional response types (e.g. Alexander et al. 2012).

In conclusion, no significant effects of feeding efficiency of predators were discerned as a result of microplastic exposure at concentrations or durations. This research highlights the need for future studies to assess environmentally-relevant dosages of microplastics in laboratory exposure trials. Overall, the present study advances our understanding of the effects of microplastics concentration and exposure on trophic interactions between key species, highlighting that microplastics may not impact upon this ecosystem function. Nevertheless, other impacts of microplastic exposure, such as bioaccumulation and biomagnification of chemical contaminants within food webs (Scopetani et al. 2018; Le Bihanic et al. 2020), and on other species, requires further investigation.

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