Aquatic Invasions (2019) Volume 14, Issue 3: 502–517

Special Issue: Behaviour in Aquatic Invasions
Guest editors: Kit Magellan, Amy Deacon, Marian Wong and Mhairi Alexander

Research Article

Temperature effects on exploratory behaviour and learning ability of invasive mosquitofish

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Co-Editors' Note: This paper is a contribution to the **Behaviour in Aquatic Invasions** Special Issue of *Aquatic Invasions*. Papers in this Special Issue explore how behaviour contributes to invasion success; native species' behavioural strategies that reduce the impacts of invasions; how knowledge of behaviour can enhance management of invasive species; and potential effects of climate change on the behavioural impacts of aquatic invasive species.

Citation: Magellan K, Bonebrake TC, Dudgeon D (2019) Temperature effects on exploratory behaviour and learning ability of invasive mosquitofish. *Aquatic Invasions* 14(3): 502–517, https://doi.org/10.3391/ai.2019.14.3.09

Received: 12 February 2019
Accepted: 2 July 2019
Published: 16 July 2019

Handling editor: Marian Wong

Abstract

The success of an invasive species depends on several factors. One of these is behavioural flexibility, which may promote invasions by allowing invaders to exploit novel foods, habitats and shelters. As behaviour can change rapidly it is an ideal first response to environmental change, including introduction and spread into a new habitat. Behaviour can be modified by learning, thus facilitating adjustment of existing behaviours to accommodate environmental conditions. Climate change is also likely to promote the introduction, establishment, and spread of aquatic invasive species. Temperature in particular is vitally important for ectotherms, directly affecting metabolic rates with consequent changes in behaviour. We evaluated the effects of temperature on the exploration tendency and learning ability of the western mosquitofish, *Gambusia affinis*. After acclimating fish to warm, medium and cool temperatures we showed that behaviour is affected by temperature, and overall warm-acclimated fish outperformed lower temperature fish. Mosquitofish showed remarkably rapid learning ability exhibiting a clear change within just three days. These effects also differed by fish sex and size. In general, larger females and smaller males were quicker to approach a novel object, while smaller females and larger males were quicker to find food. Warm-acclimated fish learned to adjust to a novel object more quickly and warm-acclimated males showed a faster response to food. In contrast, neither males nor females acclimated to warm temperatures appeared to learn to find the food and it was cool-acclimated females who were both faster to find food and showed the greatest improvement over time. Nonetheless, on both local and global scales, mosquitofishes' rapid learning ability and behavioural flexibility, which allows them to adjust to changing temperatures, is likely to augment their success as invaders in a warming world.

Key words: aquatic invasives, climate change, foraging, *Gambusia affinis*, innovation

Introduction

Ascertaining characteristics that make invasive species successful is a major goal in invasion research (e.g. Holway and Suarez 1999; García-Berthou 2007; Chapple et al. 2012). For example, successful ectothermic invaders often have "fast" traits, such as large and frequent clutches, at the establishment and spread stages of an invasion (Allen et al. 2017). Likewise, variability of traits is an important attribute of successful invaders (García-

Berthou 2007). Behavioural changes can occur very rapidly so are ideally suited as a first response to environmental changes, such as introduction and spread into a new habitat (Wright et al. 2010). Behavioural flexibility may thus promote invasions by allowing invaders to exploit novel foods, habitats and shelters (Duncan et al. 2003; Price et al. 2008; Wright et al. 2010). One aspect of behaviour that is rarely considered in conservation studies is learning (Berger-Tal et al. 2015). Learning can modify the behavioural repertoire through innovation and enables fine tuning of existing behaviours to accommodate environmental conditions (e.g. Wright et al. 2010). There is some evidence for greater learning ability in invasive species compared to native species, such as crayfish (Gherardi et al. 2002; Hazlett et al. 2002), birds (Sol and Lefebvre 2000) and crabs (Roudez et al. 2008), most often through innovation (Sol and Lefebvre 2000) or rapid learning (Roudez et al. 2008) in locating food.

Climate change is also likely to promote the introduction, establishment, and spread of aquatic invasive species (e.g, Rahel and Olden 2008). Climate change will modify the geographical distribution of fish species (Comte et al. 2013; Daufresne et al. 2009; van Vliet et al. 2013a), induce seasonal shifts in life cycle events (Daufresne et al. 2009) and enhance habitat disturbance (Chown et al. 2015) all of which can facilitate invasions (e.g. Walther et al. 2002; Chown et al. 2015; Fleming and Dibble 2015). Climate change may also modify the ecological impacts of invasive species by augmenting their predatory and competitive abilities (Rahel and Olden 2008). Temperature in particular is vitally important for ectotherms, regulating their distribution and fitness (Payne et al. 2016), and affecting competitive interactions (e.g. Taniguchi et al. 1998; Carmona-Catot et al. 2013). Temperature also directly affects the metabolic rate of ectotherms and metabolic rate is linked with changes in behaviour (Briffa et al. 2013; Norin et al. 2016), which may also benefit invasive species. The invasive crustacean, *Hemimysis anomala*, for example, is likely to cope better with warming temperatures than the functionally similar native, *Mysis salemaai*, as it increases its feeding rate in parallel with growing metabolic demand (Penk et al. 2016). However, how temperature may affect learning ability in ectotherms and the potential for this to enhance invasion success is yet to be addressed.

In this study, we evaluated the effects of temperature on the learning ability of one of the world’s 100 worst invasive species, the western mosquitofish, *Gambusia affinis* (Lowe et al. 2004), in Hong Kong. Mosquitofish were introduced into Hong Kong in the late 1940s (Dudgeon and Corlett 2004), where they threaten native freshwater taxa including several amphibian and fish species (Karraker et al. 2010; Tricarico et al. 2016) and have become the most prevalent of the introduced Poeciliidae (Tricarico et al. 2016). Mosquitofish are highly tolerant of a wide range of temperatures (Meffe et al. 1995; Lau 2014) and of temperature stress (Uliano et al. 2010). The behaviour of both *G. affinis* (Priddis et al. 2009), and the closely related eastern mosquitofish, *G. holbrooki* (Condon and Wilson 2006; Wilson et al. 2007; Lopez et al. 2018) is affected by temperature. Mosquitofish are thus an ideal model species for this study. We initially acclimated fish from a common source to warm, medium and cool temperatures. Neophilia is associated with increased exploration and innovation (Wright et al. 2010) so we first examined mosquitofish behaviour at different temperatures when presented with a novel object. We also assessed how quickly mosquitofish learned to adjust to the novel object. We predicted that mosquitofish would both approach a novel object quicker, and show faster learning, at warmer temperatures. Then, as most studies show learning and innovation in invasive species is related to foraging (e.g. Gherardi et al. 2002; Hazlett et al. 2002; Roudez et al. 2008) we evaluated mosquitofish ability to learn to locate a novel food source, predicting that learning would be quicker at warmer temperatures. Finally, as both sex (Carmona-Catot et al. 2013; Magellan and García-Berthou 2015) and size (Magellan and García-Berthou 2015) affect the behaviour of *G. holbrooki*, we assessed the interacting effects of these two traits on the exploratory behaviour and learning ability of *G. affinis*.

Materials and methods

Twenty one aquaria (60 cm × 40 cm × 40 cm) were covered on three sides with black plastic leaving the front clear. Each contained a 1 cm layer of gravel, an aquarium heater and an air supply, and was filled to 30 cm height with aged tap water. Six aquaria were pre-experiment stock aquaria, 3 were for post-experiment stock and 12 were for experiments.

Mosquitofish were collected from a well-established population in a pond in the University of Hong Kong campus (22.28°N; 114.14°E) in November 2015 when the ambient water temperature was around 23 °C. The fish were placed in the pre-experiment stock aquaria and allowed to acclimate to laboratory conditions for six weeks at ambient laboratory temperatures (23 °C ± 1 °C). After this, 7 aquaria (2 pre-exp, 1 post-exp and 4 exp) were designated for each of three temperatures, 23 °C, 27 °C and 31 °C. The cool temperature was dictated by ambient temperature while the warm temperature was selected as the highest temperature at which available aquarium heaters were reliable. These temperatures are all within the temperature range recorded at Lai Chi Wo wetlands, Hong Kong (K. Ho, *pers. comm.*) and mosquitofish's global temperature range. Temperatures were raised by no more than 3 °C per day to avoid stressing fish, so that the required temperature was achieved within a week. Fish remained at these temperatures to acclimate for a further 6 weeks. Throughout acclimation fish were fed flake fish food or defrosted frozen bloodworms (Chironomidae spp.) once or twice per day.

Six sets of feeding apparatus (feeding tubes: Figure 1) were also constructed. These consisted of a clear plastic dome, 6 cm tall and 10.5 cm

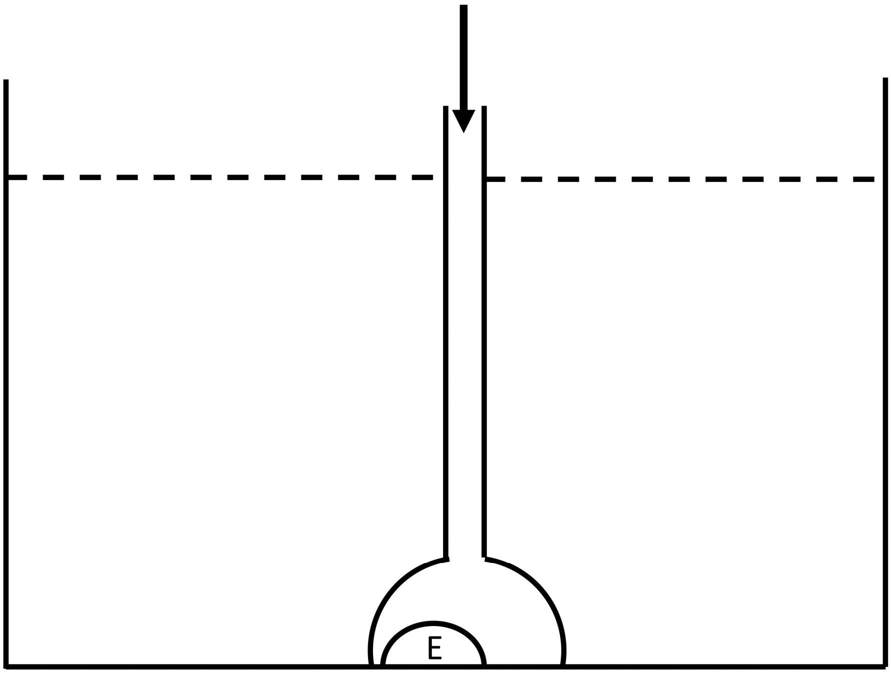

Figure 1. Schematic diagram of the experimental set-up (not to scale): dashed line represents water level, E is the entry to the feeding tube, the arrow shows where food was dropped. See main text for details.

diameter. A clear plastic tube, 2.25 cm diameter and 26 cm tall, was attached vertically to a hole in the top of the dome and a semicircular hole, 2.6 cm radius, was cut into the lower side of the dome. This construction meant that when the open base of the dome was pressed into the gravel lining an aquarium floor, the top of the tube protruded approximately 3 cm out of the water. Fish could thus see the food as soon as it was dropped into the tube from above, and as it travelled downwards to settle on the substratum, but to access food they had to swim through the semicircular opening into the dome, and in some cases up into the tube (Figure 1).

The day before the start of a set of trials, three fish were added to each of the 12 experimental aquaria. An adult female and an adult male were taken from separate corresponding-temperature stock aquaria to ensure no familiarity between them. The third fish was intended to be juvenile, but inevitable maturation of fish over the acclimation and experimental periods meant that juveniles that had experienced the same acclimation protocol were limited. The third individual was therefore juvenile, male or female as available, with the proviso that they were always smaller than the selected adult fish and thus could easily be distinguished. Fish were fed and left overnight to settle.

The following day a food stimulus, consisting of 10 defrosted frozen bloodworms, was prepared before each trial. As bloodworms may vary considerably in size, we ensured each prepared stimulus was of approximately equal volume. As the objective was to provide equal visual and olfactory stimulus for fish in each trial, and not exactly the same amount of nutrition, stimuli were not weighed.

All observations were conducted in real time by a single observer. Each trial was initiated by placing a feeding tube in the center of an aquarium. The base of the dome was pushed into the gravel so that the feeding tube remained stable and there were no gaps between the base of the dome and the gravel. The semi-circular opening in the dome was always oriented so that it faced approximately 30° to the left of straight ahead (towards the front of the aquarium), allowing the observer, seated 1 m away, to easily see if a fish had entered the dome. Observations were conducted in 2 phases to assess responses to a novel stimulus (Phase 1) and a food stimulus (Phase 2).

For Phase 1 of each observation (novelty) fish were observed for 5 minutes, recording the time until each fish first approached any part of the feeding tube (approach), defined as being within one body length of the feeding tube and oriented towards it.

After 5 minutes, any fish that had entered the feeding tube were gently removed and the feeding tube was replaced in the same position and orientation. Phase 2 (food) of the observation was then initiated by dropping a food stimulus into the top of the feeding tube. The food was allowed to drift to the floor of the aquaria naturally which meant that occasionally one or two bloodworms stuck to the side of the tube but most bloodworms reached the floor. Ten minute observations began immediately recording "approach" as before. The time until first entering the feeding tube (1st entry), defined as when the front half of the fish was within the dome, was also recorded for each fish. Observations ended after 10 minutes but the feeding tube remained in place for a further 30 minutes to allow fish to explore and locate the food if they had not done so already. The feeding tube was then removed, any bloodworms that had been stuck to the inner surface were removed and the tube was rinsed in clean water.

Fish remained in their aquarium for 3 days, and the same protocol was repeated on days 2 and 3. No supplemental food was given during this time, as we wanted fish to associate the feeding tubes with food and the 10 bloodworms provided each day should have been sufficient for all fish within an aquarium. If any female gave birth during the 3 days of observations, that trial was abandoned and fish were removed to the stock aquaria. At the end of a set of trials, all fish were removed and measured using a ruler (total length to the nearest mm) and fish were placed in the previously prepared stock aquaria so that they were not reused. The entire 3 day trial protocol was repeated four times, using a subset of experimental aquaria for sets 3 and 4, resulting in 13 replicates for each temperature.

Statistical Analyses

All analyses focused on the large adult females and males only. The third fish, which may have been male, female or juvenile, was excluded 1) because the limited number of juveniles meant that some treatments contained very few individuals; and 2) to avoid the potential for any hierarchy, and therefore confounding, effects on the behaviour of the smaller males and females which were assumed to be subordinate (e.g. Matthews and Wong 2015). All analyses were carried out using SPSS v. 20.

First, to ensure that fish were evenly distributed across temperature treatments with respect to size we used a Generalized Estimating Equation (GEE: a special case of Generalized Linear Models (GLM) for non-independent data: see Magellan and García-Berthou 2015) with temperature (between subjects) and sex (within subjects) as the independent variables and fish length as the response variable (normal distribution, identity link function), and fish group included in the model structure.

Second, differences in approaching a novel object (the feeding tube) were examined using Phase 1 data in a GEE. The response variable, approach, was tested with assumed gamma distribution and log-link function so 1 was added to all scores to avoid any zeros which would be excluded from the gamma distribution. The independent variables were temperature (between subjects), day and fish sex (within subjects) and fish group was again included in the model structure. The inclusion of "day" enabled us to assess learning speed. Fish size was excluded from the previous analysis because the minimal overlap in size between females and males meant the inclusion of size would incorrectly increase the chances of a significant effect of sex. However, fish size may influence behaviour so female and male approach behaviour were examined in separate GEEs. The response variable was tested with assumed gamma distribution and log-link function as before and the independent variables were temperature and fish size (between subjects), and day (within subjects) with fish group included in the model structure.

Third, the differences in improvement in performance over time (learning) with food were investigated using Phase 2 data. The difference in time between approaching and entering the feeding tube was used as a proxy for learning, i.e. a smaller difference between these two scores over time implies that fish are learning that in order to obtain food once they have seen it, they have to enter the feeding tube. Only data for fish that both approached and entered the feeding tube were included as fish that did neither would obtain the same difference scores as fish that entered the feeding tube immediately on approaching it. The response variable, learning, was included in a GEE with assumed normal distribution and identity link function. The independent variables were temperature (between subjects), day and sex (within subjects) and fish group was included in the model structure. Finally, females and males were again examined separately. In this case, as only fish that both approached and entered the feeding tube were included, and fish performance improved over time (see below), the data for day 1 and 2 were too sparse for a realistic analysis. Therefore, only data for day 3 were used in two GLMs with the response variable, learning (normal distributions and identity link function), and temperature and size as independent variables.

Table 1. Results from the Generalised Estimating Equations for time until approach in the novelty part of the trials (Phase 1) for a) females and males together and b) females and males separately. Significant results are highlighted in bold.

a)

Effect	Wald χ^2	df	p
Temp	1.616	2	0.446
Day	65.688	2	**< 0.001**
Sex	17.540	1	**< 0.001**
Temp × Day	2.577	4	0.631
Temp × Sex	1.008	2	0.604
Day × Sex	9.497	2	**0.009**
Temp × Day × Sex	2.354	4	0.671

b)

	df	Sex			
		Females		Males	
		Wald χ^2	p	Wald χ^2	p
Temp	2	16.309	**< 0.001**	7.181	**0.028**
Day	2	6.098	**0.047**	2.928	0.231
Size	1	29.986	**< 0.001**	0.030	0.862
Temp × Day	4	12.213	**0.016**	11.555	**0.021**
Temp × Size	2	16.261	**< 0.001**	5.798	0.055
Day × Size	2	4.736	0.094	2.480	0.289
Temp × Day × Size	4	11.094	**0.026**	11.619	**0.020**

All data is available from the corresponding author upon reasonable request.

Results

There was no difference in the length of fish between temperature treatments (Wald $\chi^2 = 3.439$; df = 2; p = 0.179), but length differed significantly between the sexes (Wald $\chi^2 = 113.227$; df = 1; $p < 0.001$). Fish exhibited a range of responses to the presentation of both a novel object and food. Many fish readily approached the feeding tube and entered even without food, with some swimming up into the vertical part of the feeding tube. Some fish entered the feeding apparatus directly upon sighting food while others were more indirect, and still others seemed wary or indifferent. A few fish, particularly larger females, appeared to guard the entrance to the feeding tube.

During Phase 1 (novelty), approximately 64% of all fish approached the feeding tube over all days, with 34% approaching on day 1, 72% on day 2, rising to 86% on day 3. The proportion of fish approaching was more evenly distributed across temperatures with 57%, 71% and 63% approaching at cool, medium and warm temperatures respectively, across all days. The first analysis reflects these general patterns with the time to first approach differing significantly and interactively between sexes and over time (day) (Table 1a). For females, approach time generally decreased over time but there was no difference between fish acclimated to different temperatures. For males the decrease in approach over time is only apparent in warm-acclimated fish, with time to first approach increasing over time in the medium and cool temperature treatments (Figure 2).

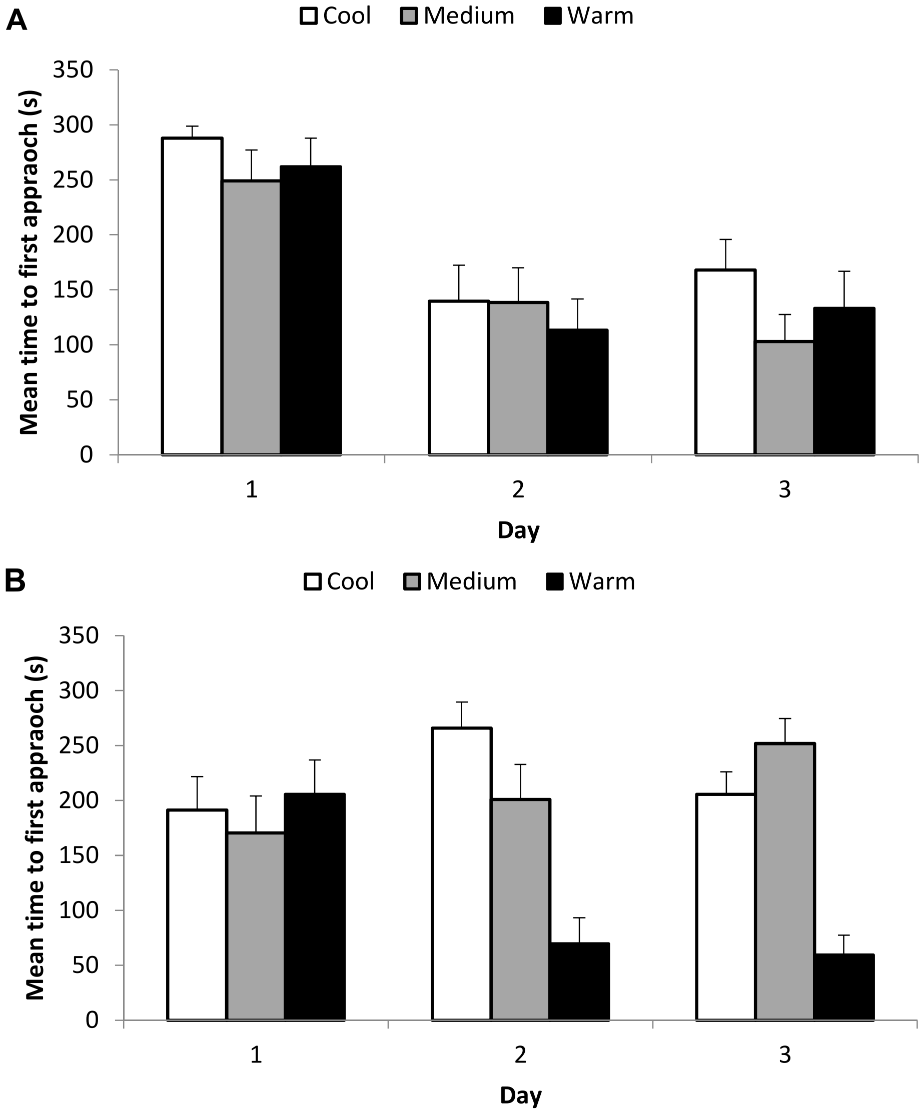

Figure 2. Changes in the mean time to first approach a novel object over time for females (A) and males (B). Error bars represent standard error.

Although temperature had no significant effect on performance when both sexes were examined together and size was excluded (Table 1a), when females and males were considered separately the effects of temperature were apparent. For females, almost all main effects and their interactions, particularly temperature, size and their interaction, were significant (Table 1b). There was no clear overall difference in approach speed with temperature, but approach time generally decreased with female size for cool and medium temperatures and remained approximately stable across female sizes at the warm temperature (Figure 3a, b, c). An overall decrease in approach time over the three experimental days is also evident (Figure 3). For males, in contrast, the main influence on performance was temperature with a significant interaction with both day and size (Table 1b). Approach times generally decreased with both increasing size and over time (Table 1b, Figure 3d, e, f). Males from the warm temperature treatment showed the most consistent trend for faster performance with increasing size, while males

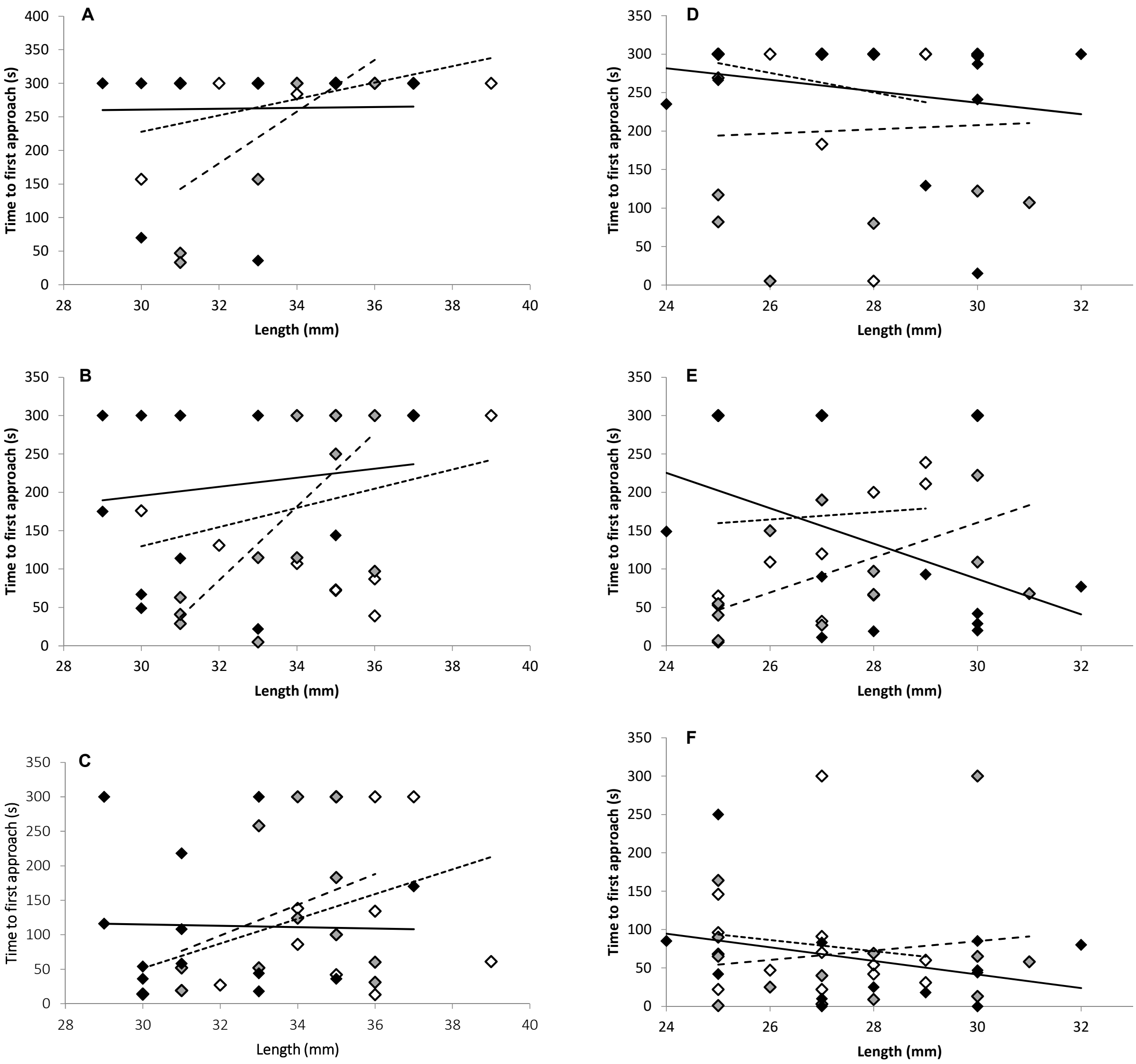

Figure 3. Time to first approach a novel object as a function of size for females (A, B, C) and males (D, E, F) on Days 1 (A, B), 2 (C, D) and 3 (E, F) respectively. Trendlines are added for illustration: dot – cool temperature; dash – medium temperature; solid – warm temperature.

in the medium and cool temperature treatments showed a weaker negative, and in some cases even positive, association between approach time and size (Figure 3).

In Phase 2 (food) 37% of all fish both approached and entered the feeding tube over all days. This was biased towards day 3 with 25%, 39% and 48% of fish approaching and entering on days 1, 2 and 3 respectively. For temperature treatments 39%, 38% and 35% approached and entered in the cool, medium and warm temperatures respectively.

When both sexes were considered together, the significant interaction between sex and day for learning is illustrated by an overall trend for a smaller difference between first entry and first approach over time, with males most clearly showing this pattern while females increased the time between approaching and entering the tube on day 2 (Figure 4). For females, fish in the cool temperature treatment showed the fastest response, while for

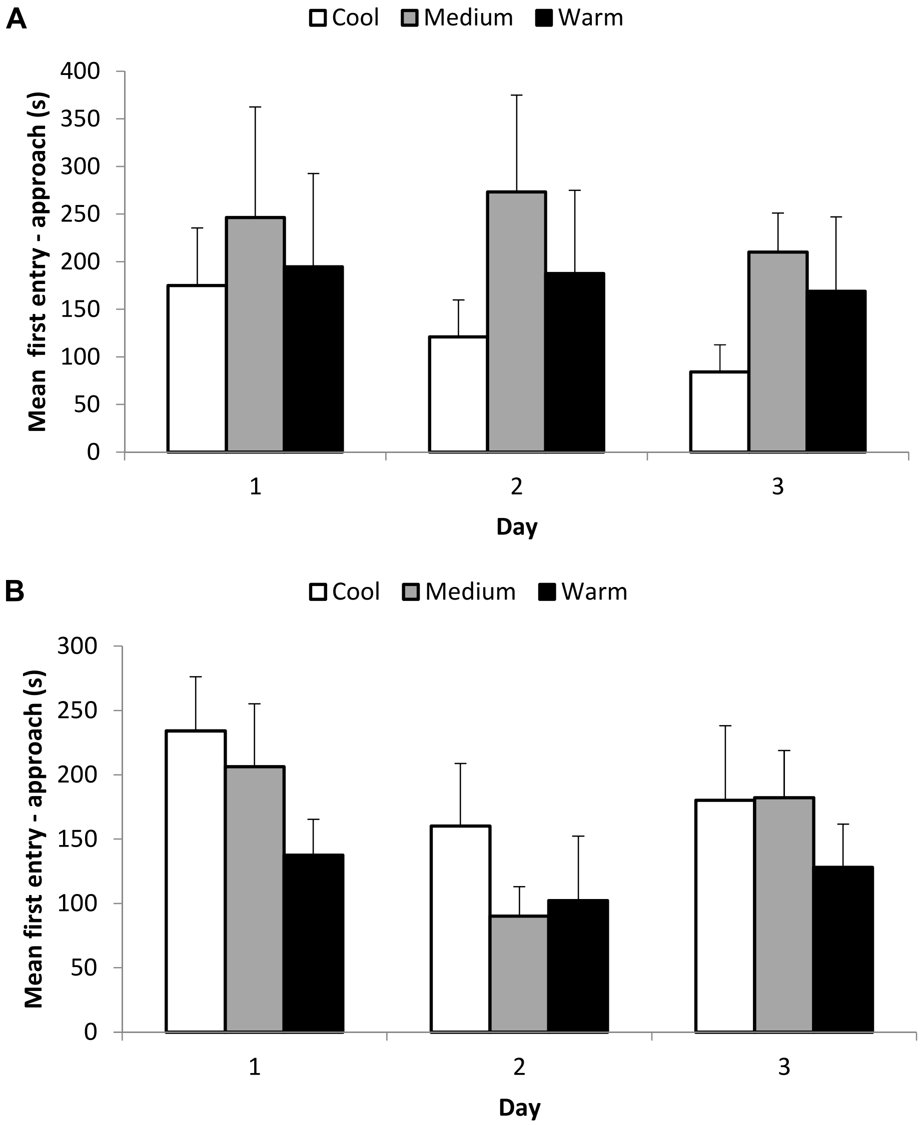

Figure 4. Differences in learning (entry time – approach time) with temperature over time for females (A) and males (B). Error bars represent standard error.

males, the warm-acclimated fish showed the fastest response but this did not seem to improve over time. When females and males were examined separately, the relationship between performance and size became increasingly positive with an increase in temperature for both sexes (Table 2b, Figure 5). Larger females took longer to enter the feeding tube in all temperature categories, and particularly at the warm temperature, while larger males entered the feeding tube quicker at the cool and medium temperatures but slower at the warm temperature.

Discussion

The exploration behaviour and learning ability of invasive mosquitofish is affected by temperature. Overall, warm-acclimated fish outperformed medium- and cool-acclimated fish, but effects are not clear cut. Links between temperature, metabolic rate and behaviour in ectotherms (e.g. Briffa

Table 2. Results from the Generalised Estimating Equations (a) and Generalised Linear Models (b) for the difference between approach and 1st entry to the feeding tube during Phase 2 for a) females and males together, b) females and males separately. Significant results are highlighted in bold.

a)

Effect	df	Wald χ^2	p
Temp	2	2.193	0.334
Day	2	3.122	0.210
Sex	1	1.585	0.208
Temp × Day	4	0.321	0.988
Temp × Sex	2	10.755	**0.005**
Day × Sex	2	3.936	0.140
Temp × Day × Sex	4	2.446	0.654

b)

Effect	df	Females		Males	
		Wald χ^2	p	Wald χ^2	p
Temp	2	11.478	**0.003**	11.974	**0.003**
Size	1	15.323	**< 0.001**	6.737	**0.009**
Temp × Size	2	12.275	**0.002**	11.819	**0.003**

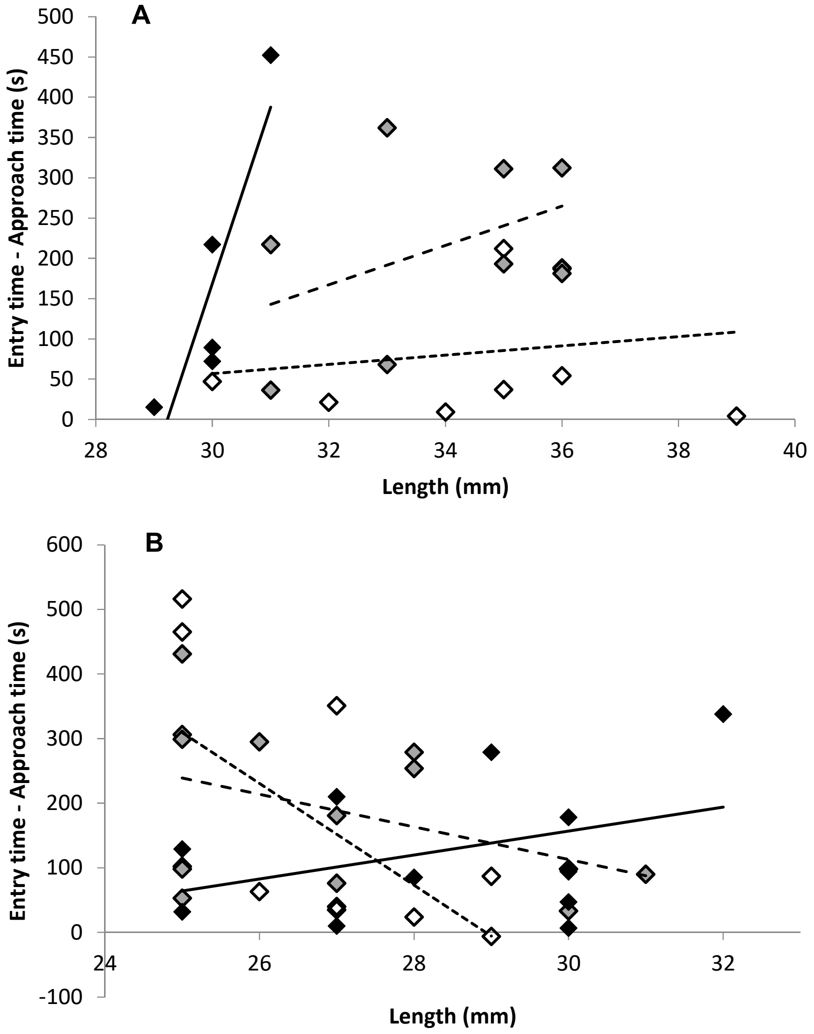

Figure 5. Differences in learning with temperature and size on day 3 only for females (A) and males (B). Note the negative y-value in Figure 5B: this results from a single male who never approached the food but reversed into the feeding tube while watching a female. White, grey and black dots represent the low, medium and high temperature treatments respectively. Trendlines are added for illustration: dot – low temperature; dash – medium temperature; solid – high temperature.

et al. 2013; Norin et al. 2016) mean some influence of temperature on behaviour is likely. Higher energy requirements at elevated temperatures suggest fish will be more motivated to find food. Higher temperatures may also facilitate an increase in function. Both male (Wilson 2005) and female (Condon and Wilson 2006) *G. holbrooki* have higher burst swimming speeds when acclimated to higher temperatures, and a similar effect may have contributed to the faster response to food in warm temperature males. However, it was cool temperature females who were both faster to find food and showed the greatest improvement over time. Phenotypic diversity may influence individual performance with variations in baseline metabolism affecting responses (Norin et al. 2016) and individual behaviour may be less predictable at higher temperatures (Briffa et al. 2013). Moreover, although mosquitofish in Hong Kong maintain their physiological performance over a wide range of temperatures (Lau 2014), peak performance varies depending on the population, the specific behaviour examined (e.g. Wilson 2005; Condon and Wilson 2006; Carmona-Catot et al. 2013) and individual traits such as sex and size (Carmona-Catot et al. 2013; Magellan and García-Berthou 2015).

Differences between sexes in performance is expected. Sexual dimorphism (e.g. Magellan and García-Berthou 2015), contrasting motivations in reproduction (Kokko and Jennions 2008) and even sex specific vulnerability to predation (Magurran 2000) suggest selection should also favour differences in exploration and learning. In a close relative of mosquitofish, the guppy, *Poecilia reticulata*, females learned to discriminate between shoals of different sizes faster than males (Lucon-Xiccato et al. 2016), whereas males learned shape discrimination faster (Lucon-Xiccato and Bisazza 2016). In another study on guppies, the ability to complete a novel foraging task varied with body size as well as sex (Laland and Reader 1999), and in this study too effects of temperature and sex were modified by body size. In general, larger females and smaller males were quicker to approach a novel object, while smaller females and larger males were quicker to find food. Three related factors may explain this difference. First, mosquitofish form sex specific hierarchies with larger individuals usually achieving dominance (Chen et al. 2011). Second, mosquitofish tend to be consistent in their behaviour (Cote et al. 2010). Third, male mosquitofish are ardent in pursuit of mating opportunities and target larger, more fecund females (Bisazza et al. 1989). It may be that during the novelty part of trials, larger females, that were used to being dominant, were able to approach a novel object, but males were more interested in mating; whereas when food was present, the associated presence of large males excluded larger females who tried to avoid male harassment.

Consistency in behaviour has other implications for the findings of this study. Animals often exhibit personalities, demonstrated as individual consistency in behavioural traits (Conrad et al. 2011). Such traits may also be affected by environmental conditions. For example, on exposure to temperature alterations, bold and shy rainbow trout, *Oncorhynchus mykiss*, showed less extreme boldness and shyness respectively (Frost et al. 2013). These traits may also be associated with other behaviours. For example, bold individuals may out-perform shy individuals during foraging as they cover more ground, accumulate information more rapidly and make more frequent food discoveries (Ólafsdóttir and Magellan 2016). In this study, although a correlation between exploratory tendency (boldness or shyness) and foraging ability was initially expected, unfortunately it was not possible to assess this as the fish that approached the novel object in Phase 1 and those that found food in Phase 2 were not necessarily the same individuals. This may explain why warm-acclimated fish of both sexes did not appear to learn to find the food although they did learn to adjust to a novel object. However, this remains an interesting possibility for further investigation, perhaps using alternative learning tasks.

At a population level, fish in almost all conditions improved their performance over time indicating that learning occurred. Given that trials occurred over just three days, this demonstrates a remarkably rapid learning ability. Feeding innovations and rapid learning of novel foraging situations are beneficial for invasive species (e.g. Hazlett et al. 2002; Roudez et al. 2008), especially during the initial stages of invasion (Wright et al. 2010). Moreover, fish learning improves with more repetition (Bisazza et al. 2014), and this learning ability can be extrapolated to other situations such as learning to distinguish predator odours (e.g. Gherardi et al. 2002) or novel toxic prey (e.g. Wijethunga et al. 2016). Mosquitofishes' reputation for outcompeting native species (Pyke 2008) is thus not surprising.

We have shown here that exploration tendancy and learning ability in mosquitofish is affected by temperature. Higher temperatures are generally associated with an increase in performance though the results are complex, and affected by time, sex and body size. This may, in part, reflect the relatively low sample size in this study, particularly for the second (food) phase of the trials. The fish in this study came from a common source and were acclimated to their test temperatures. However, behavioural flexibility is just one aspect of plasticity. Over longer time scales, plasticity may also be expressed developmentally, and genetically via selection over generations (Wright et al. 2010) which may also be affected by temperature. Indeed, *G. holbrooki* mature earlier at a smaller size at high temperatures (Meffe 1992) and show inheritance of temperature tolerance (Meffe et al. 1995). Mosquitofish are notoriously difficult to eradicate (Pyke 2008; Ruiz-Navarro et al. 2013) but their sperm storage ability means just a single female can found a population, as demonstrated in the closely related guppy (Deacon et al. 2011). They also have very wide physiological temperature tolerance (Lau 2014) and one of the widest reproductively active temperature ranges for ectotherms (Wilson 2005). On a local scale, Hong Kong water temperatures are predicted to rise by 0.6–1.2 degrees over the 21st Century (van Vliet et al. 2013a, b) or even up to 3.0 degrees in some warming scenarios (van Vliet et al. 2013b), and for some areas of the world, including many already invaded by mosquitofish, the predicted rise is even greater (van Vliet et al. 2013a, b). The rapid learning ability of mosquitofish, together with their behavioural flexibility which allows them to adjust to changing temperatures, is likely to augment their success as invaders.

Acknowledgements

KM was funded by a University of Hong Kong "Earth as a Habitable Planet" postdoctoral fellowship. Sincere thanks to Marian Wong and 2 anonymous reviewers who gave their valuable time to help improve our manuscript.

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