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Stormwater wetlands can function as ecological traps for urban frogs

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Running head: Frogs caught in ecological traps

ABSTRACT

Around cities, natural wetlands are rapidly being destroyed and replaced with wetlands constructed to treat stormwater. Although the intended purpose of these wetlands is to manage urban stormwater, they are inhabited by wildlife that might be exposed to contaminants. These effects will be exacerbated if animals are unable to differentiate between stormwater treatment wetlands of varying quality and some function as

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‘ecological traps’ (i.e. habitats that animals prefer despite fitness being lower than in other habitats). To examine if urban stormwater wetlands can be ecological traps for frogs, we tested if survival, metamorphosis-related measures and predator avoidance behaviours of frogs differed within mesocosms that simulated stormwater wetlands with different contaminant levels, and paired this with a natural oviposition experiment to assess breeding-site preferences. We provide the first empirical evidence that these wetlands can function as ecological traps for frogs. Tadpoles had lower survival and were less responsive to predator olfactory cues when raised in more polluted stormwater wetlands, but also reached metamorphosis earlier and at a larger size. A greater size at metamorphosis was likely a result of increased per capita food availability due to higher mortality combined with eutrophication, although other compensatory effects such as selective-mortality removing smaller individuals from low-quality mesocosms may also explain these results. Breeding adults laid comparable numbers of eggs across wetlands with high and low contaminant levels, indicating no avoidance of the former. Since stormwater treatment wetlands are often the only available aquatic habitat in urban landscapes we need to better understand how they perform as habitats to guide management decisions that mitigate their potential ecological costs. This may include improving wetland quality so that fitness is no longer compromised, preventing colonisation by animals, altering the cues animals use when selecting habitats, pretreating contaminated water prior to release, providing off-line wetlands nearby or simply not constructing stormwater treatment wetlands in sensitive areas. Our study confirms the potential for urban stormwater treatment wetlands to function as ecological traps and highlights the need for greater awareness of their prevalence and impact at landscape scales.

Keywords: *amphibians, behavior, constructed wetland, habitat selection, HIREC, metapopulation, predator avoidance, urbanisation*

INTRODUCTION

Urbanisation is changing the biological, chemical and physical characteristics of aquatic ecosystems (Walsh et al. 2005). Within urban areas, stormwater treatment wetlands are being created to provide ecosystem services such as flood control, and water filtration

and purification (Hammer 1989). These habitats are now ubiquitous and modify both the distribution and nature of aquatic habitats around many cities (Kentula et al. 2004). For example, in Oregon, USA, land-use changes from underdeveloped and agricultural to urban and residential led to almost all the palustrine emergent/open water wetlands being graded as either fair (46%) or poor (43%) quality (Kentula et al. 2004). As stormwater treatment wetlands often resemble natural wetlands and attract animals (Tixier et al. 2011, Hassall and Anderson 2015), they may provide valuable habitat in urban landscapes where most natural waterbodies have been lost or heavily degraded.

One of the primary functions of stormwater treatment wetlands, however, is to capture and treat contaminants (e.g. nutrients, sediments, heavy metals, pesticides) prior to release into receiving waters (Hammer 1989). Animals that inhabit them may thus suffer a range of deleterious effects (Sparling et al. 2004). For example, embryonic and larval amphibians were susceptible to stormwater wetland sediments, with short-term exposure often reducing survival and growth (Snodgrass et al. 2008, Gallagher et al. 2014). Contaminants common in stormwater wetlands can also cause maladaptive behaviours to develop *via* olfactory disruption (Tierney et al. 2010). For example, Cuban tree frog tadpoles do not avoid predator cues and are hyperactive following exposure to the herbicide atrazine (Ehram et al. 2016). These types of behavioural changes can indirectly reduce fitness by increasing the probability of predation (Broomhall 2004).

The impact of highly polluted stormwater treatment wetlands will be exacerbated if animals do not recognise them as poor-quality habitats, and they function as 'ecological traps'. An ecological trap occurs when there is differential fitness between available habitats, and animals show a preference (severe trap) or an equal-preference (equal-preference trap) for the habitat where fitness is lower (Robertson and Hutto 2006). Ecological traps are thus defined by impacts to individuals (Robertson et al. 2013), and so the identification of ecological traps requires information about how habitats impact the fitness (i.e. survival, reproduction) and habitat preferences of animals.

Although the potential for stormwater wetlands to function as ecological traps has been discussed previously (Tilton 1995, Brand and Snodgrass 2010, Hale et al. 2015a, Sievers et al. 2018), and while we have clear evidence that these habitats can reduce individual fitness (e.g. Snodgrass et al. 2008, Gallagher et al. 2014), we currently lack the

necessary information on habitat selection preferences to confirm this. For taxa that lack post-hatching parental care, the selection of oviposition sites is a critical decision that directly affects offspring fitness, and ultimately, population dynamics (Reich and Downes 2004). In particular, strong selective forces should be acting on oviposition behaviour when larvae are unable to leave unsuitable sites (Rudolf and Rodel 2005, Resetarits and Silberbush 2016). These decisions can be influenced by a range of cues, such as the presence of predators (Resetarits and Wilbur 1989) and pesticides (Vonesh and Kraus 2009), and so amphibians can respond adaptively by preferring predator-free or unpolluted conditions. However, as stormwater wetlands are often the only available breeding habitat within urban landscapes, oviposition-site selection is likely to involve choices between wetlands that all are contaminated to varying degrees, complicating the decision-making process.

Distinguishing when stormwater wetlands are functioning as ecological traps is important as they represent ubiquitous and potentially valuable aquatic habitat within urban areas. If traps are present, they will draw animals from higher- to lower-quality stormwater wetlands, impacting the likelihood of both local and regional population persistence (Donovan and Thompson 2001, Hale et al. 2015b). Understanding how traps form and their consequences to animals has thus become an emerging management challenge. In this study, our overarching goal was to test if stormwater wetlands can function as ecological traps for frogs. While ecological trap studies often compare habitats that are hypothesised to be traps with unaltered (e.g. 'reference') habitats, here we examined responses to pairs of stormwater wetlands of differing quality, which better reflect the reality of urban landscapes. We experimentally replicated the conditions of heavily and, in comparison, mildly contaminated wetlands in a mesocosm experiment to quantify impacts on the fitness and behaviour of tadpoles, coupled with a field-choice experiment to examine the oviposition preferences of breeding frogs. We hypothesized that: (1.) tadpole survival and size/mass at metamorphosis would be lower within more contaminated wetlands, and (2.) their ability to respond adaptively to predator cues would be reduced. If urban stormwater treatment wetlands are ecological traps, we predicted that females would not avoid wetlands where offspring fitness was lower. We discuss our results in the context of managing stormwater wetlands that are ecological traps to

facilitate the persistence of aquatic animals in urban landscapes.

METHODS

Study species

Marsh frogs (Genus *Limnodynastes*, Fitzinger, 1843) are ground-dwelling frogs native to Australia, New Guinea and the Torres Strait Islands. We focused on the spotted marsh frog *Limnodynastes tasmaniensis* (Günther, 1858) and the striped marsh frog *Limnodynastes peronii* (Duméril and Bibron, 1841), two species common in south-eastern Australia that are known to occupy and breed in urban wetlands (Hamer and Parris 2011, Ficken and Byrne 2013). Nocturnal call surveys confirmed the presence of both species at all study sites.

Study sites

As of 2015, nearly 500 wetlands had been constructed in Melbourne, with more than 80% of those built in the last two decades alone (Hale et al. 2015a). To examine frog responses to stormwater wetland quality, we selected six stormwater wetlands that differed in the level of contamination within the Greater Melbourne Region, Australia. We selected three pairs of wetlands each containing one hypothesized high quality (HQ) and one hypothesized low quality (LQ) site, based on sediment and water quality data collected by the Centre for Aquatic Pollution Identification and Management (Marshall et al. 2016, Sharley et al. 2017). In particular, LQ and HQ status was based on the presence and concentrations of contaminants (heavy metals and pesticides; Table 1) that could have an effect on amphibian fitness (Egea - Serrano et al. 2012).

Mesocosm design

The experiment included the six source wetlands, each replicated eight times in a completely randomized design. We established 48 circular mesocosms (150L, 875 mm diameter x 310 mm deep) at The University of Melbourne, Burnley Campus

(37°49'47.17" S, 145°01'28.64" E). Each mesocosm was filled with 120 L of water filtered through a 100 µm sieve, 2 L of fine sediment filtered through 64 µm mesh, and 4 L of unfiltered, autoclaved sediment from the respective source wetland in September 2016, and covered with 50% shade-cloth to keep out predators. Filtered sediment was added to allow the establishment of natural benthic communities, and unfiltered, autoclaved sediment to provide benthic structure; both methods also removed predatory invertebrates (Pettigrove and Hoffmann 2005). We collected zooplankton from an on-campus wetland using a 50 µm plankton net, with samples diluted and added to each mesocosm in 0.2 L aliquots after screening to exclude predators. Mesocosms were allowed to settle and mature for 4 weeks. We conducted fortnightly 25 L water changes using water from the respective source wetland to more accurately simulate the conditions of the source wetland for the duration of the experiment. Evaporated water was replaced with aged tap water or natural rainfall so that depth was kept consistent.

We collected ten spotted marsh frog egg masses from a wetland (37°37'08.06" S, 145°00'10.67" E) created for the endangered growling grass frog *Litoria raniformis* (Keferstein, 1867). Each egg mass was hatched in an individual 2L beaker in the laboratory, and we transferred two-day-old tadpoles into the mesocosms. Each mesocosm received ten tadpoles on the 16th October 2016, one from each of the ten egg masses to remove any confounding genetic/parental effects.

Fitness responses

Tadpoles were removed from mesocosms as they began to metamorphose (during tail resorption; Stage 45; Gosner 1960) and placed in individual 0.5 L plastic containers until complete tail resorption (Stage 46). We then recorded days to, length at, and mass at metamorphosis, and the incidence of any physical abnormalities (e.g. missing or malformed limbs, scoliosis). Survival was calculated as the number of tadpoles reaching metamorphosis relative to the number of tadpoles released into each mesocosm (after accounting for those removed for the predator avoidance trials; see Appendix S1: Fig. S1).

Periphyton, water and sediment analysis

After the final tadpole metamorphosed (80 days), we collected periphyton, water and sediment samples from each mesocosm. To estimate food availability, we scraped all attached periphyton from a 5 x 10 cm section of the mesocosm wall with a razor blade, following the removal of the water. Samples were oven dried at 60°C for 24 h, weighed, inserted into a blast furnace at 475°C for 18h, and re-weighed to calculate ash-free dry-weight (AFDW). We measured ammonia, nitrite, nitrate, phosphate and total alkalinity using an eXact® Eco-Check photometer (Industrial Test Systems, Inc, USA), and pH and conductivity using a WP-81 meter (TPS, Brisbane, Australia). We air-dried and filtered sediments to 1 mm, and metal concentrations were quantified by TrACEES at The University of Melbourne (Appendix S2; www.chemicalanalysis.unimelb.edu.au).

Predator avoidance trials

We harvested odours from dragonfly larvae – a ubiquitous and voracious tadpole predator commonly used in predator detection and avoidance experiments (Carlson and Langkilde 2013) – and conducted predator avoidance trials in a choice tank with three spatial zones delineated by lines drawn on the tank: near predator (10 cm section), a middle zone (30 cm) and an away from predator zone (10 cm; see Appendix S3 for detailed experimental design). Between each trial, the choice tank was rinsed with de-ionised (DI) water, towel dried and re-filled with DI water for the next trial. Trials were run with tadpoles (42 days old) from two source wetland pairs, as insufficient tadpoles were available from the third due to high mortality. We used two tadpoles from each mesocosm, except for the second pair, where two mesocosms from each source wetland did not contain enough tadpoles (Appendix S2: Fig. S1). For each source wetland, we conducted 12 replicates of each of four treatments: control (i.e. aged tap water) vs control, visual cues vs control, olfactory cues vs control, and visual and olfactory cues combined vs control. Therefore, we tested tadpoles from the first pair three times and tadpoles from the second pair four times, with no single tadpole experiencing the same treatment type more than once.

We placed a tadpole in the centre of a choice tank and allowed it to acclimate to conditions for 2 min, which allowed time for the olfactory cues to reach the centre of the tank (i.e. tadpole; sensu Ward and Mehner 2010). The cylinder holding the tadpoles was then lifted with a rope and pulley, and we recorded the 5 min trial using a GoPro Hero 3+

(see Appendix S3). We calculated the proportion of time spent in the zone away from the predator cues relative to the time spent in the zone with the cues, based on recording the position of tadpoles every 30 seconds. Following trials, these tadpoles were humanely killed.

Oviposition site preference

We monitored oviposition preference concurrently with mesocosm experiments so that water and sediment quality would be as comparable as possible. We deployed three pairs of 100 L pre-formed, rock-style fibreglass ponds around each of the six source wetlands used in the mesocosm experiment. Each pond-pair simulated each wetland-pair used previously by reciprocally adding sediment and water from source wetlands. This provided breeding frogs with a choice between two source wetland conditions with differing contaminant loads. We deployed pond-pairs approximately 10–40 m from the wetland edge, with the two ponds in a pair consistently deployed 50 cm apart in the same orientation. Pond-pairs were separated from each other by at least 50 m. Similar to the mesocosm experiments, we conducted fortnightly water changes.

Within each pond, we added a potted, semi-aquatic plant abundant at the source wetlands to encourage oviposition (foliage approximately 30 cm above water level; *Carex* sp.). We inspected ponds for egg masses at least weekly over the spring/summer breeding period. Although mesocosm and predator avoidance experiments were conducted with spotted marsh frogs, we could not exclude the possibility that some egg masses laid in the oviposition experiment belonged to striped marsh frogs, given that species identifications are challenging, and we did not have the capacity to raise tadpoles from every mass to confirm species identity. We removed and photographed egg masses, and emptied and refilled any pond containing eggs with sediment and water from the respective source wetland to avoid confounding future oviposition decisions.

Statistical analysis

Mesocosm experiment

We analyzed survival, mass and length at metamorphosis, development (stages per day), total periphyton dry weight, ash-free fry weight of periphyton (AFDW), water quality,

and the concentration of heavy metals in sediments using linear models. For each model, we fitted treatment (HQ or LQ) and location (source wetland pair) as fixed effects. Ideally, here and for the below analyses, location would have been fitted as a random effect, but due to low replication, we would be unable to accurately estimate variances. Instead, including it as an interacting fixed effect allows us to estimate how consistent the treatment effect is across wetland pairs. Data were log-transformed for all variables except survival to meet the assumptions of normality and homogeneity of variances.

Predator-avoidance trials

To examine responses to predator cues, we used linear mixed models with location (source wetland pair), treatment (HQ or LQ) and cue (control, visual, olfactory and visual + olfactory) and a treatment by cue interaction fitted as fixed effects, and tadpole ID nested within mesocosm, and site (source wetland) fitted as random effects. We estimated credible intervals for the fixed effects used in figures. In addition, we examined if the proportion of time spent away from predators for the olfactory-cue-only trials was related to the sediment concentration of metals within mesocosms using linear regression. Because the concentrations of different metals were highly correlated, we calculated a heavy metal quotient, calculated as the sum of the observed concentrations divided by the probable effects level from MacDonald et al. (2000) using arsenic, chromium, copper, lead, mercury, nickel and zinc.

Oviposition-site preference experiment

We assessed oviposition-site preference using Chi-squared tests of the conditions (HQ or LQ ponds) eggs were laid into and the conditions (HQ or LQ source wetland) ovipositing frogs came from as factors, with each egg mass treated as a single data point. The number of eggs per egg mass was analysed using a linear model with treatment and location fitted as fixed effects.

We assessed normality and homogeneity of variances before all analyses using Q-Q and residual plots, respectively. We performed analyses in R 3.2.2 (R Development

Core Team 2015), using the lmerTest package (Kuznetsova et al. 2015) to fit mixed effects models.

RESULTS

Fitness responses

All measured parameters were significantly different between HQ and LQ treatments (Table 2). Based on model estimates, tadpoles raised in HQ treatments exhibited on average 42.0% higher survival, took 5.9 days longer to reach metamorphosis, and were 0.29 g lighter and 2.5 mm shorter than those raised in LQ treatments (Fig. 1; Table 2). No physical abnormalities were observed in any of the tadpoles.

Periphyton, water and sediment analysis

Periphyton was approximately three times more abundant in LQ mesocosms (Table 2; Fig. 1). Water from LQ mesocosms also had higher pH, conductivity, total alkalinity, and phosphate levels (Fig. 2; Appendix S4: Fig. S1). Although ammonia, nitrite and nitrate were often detected in LQ treatments, levels were below the detection limit for most HQ treatments (Appendix S4: Fig. S1). LQ mesocosms also had significantly higher concentrations of nine of the analysed metals (Fig. 2; Appendix S4: Table S1, Fig. S2).

Predator avoidance trials

Control trials with no stimuli indicated no inherent biases in terms of preference for one side of the tank (mean proportion of time to left side (\pm 95% CI); HQ: 0.48 ± 0.14 ; LQ: 0.54 ± 0.15). There was a significant two-way interaction between treatment and cue type (Table 3), as tadpoles from the HQ treatment avoided olfactory cues from predators, but those from LQ conditions did not (Fig. 3). Tadpoles from both treatments did not respond to visual cues in isolation, but did when both visual and olfactory cues were provided simultaneously (Fig. 3). Further, there was a significant, negative correlation between the proportion of time tadpoles spent away from predator olfactory cues and the heavy metal quotient for mesocosm sediments (Table 4; Fig. 4).

Oviposition site preference

In total, 23 egg masses were laid into both high- and low-quality ponds, with no effect of the quality of the site they were laid at, or the conditions they were laid into ($\chi^2_1 = 0$, $p = 1$). The number of eggs per egg mass (Mean \pm SE) was also similar for masses laid into different treatment conditions (HQ: 1146 ± 74 ; LQ: 1102 ± 62 ; $F_{1,42} = 0.51$, $p = 0.47$).

DISCUSSION

We provide the first empirical evidence that urban stormwater treatment wetlands can be ecological traps for frogs. Tadpoles had lower survival and were less responsive to predator cues when raised in more polluted conditions, but breeding adults laid comparable numbers of eggs at sites regardless of habitat quality.

Amphibians are often considered sensitive to habitat modification and contamination (Blaustein et al. 1994, Sievers et al. 2018), and survival differed most for the two source wetland pairs with the greatest differences in environmental conditions (Appendix S5: Fig. S1). Heavy metals (Lefcort et al. 1998), pesticides (Egea-Serrano et al. 2012), hydrocarbons (Jelaso et al. 2002) and nitrogen (Marco et al. 1999) can all reduce amphibian survival, and a combination of contaminants can have even greater impacts (Relyea 2009). Metal concentrations within the mesocosms closely matched those from initial wetland sampling, so pesticide and hydrocarbon levels were also likely to have been higher within LQ treatments, further contributing to reduced survival. In addition, we would have likely observed even stronger fitness reductions if we also examined the survival of eggs and embryos given they can be very susceptible to contaminants (Brand et al. 2010).

Survival was lower in LQ treatments, but tadpoles were quicker to metamorphose, and larger when they did. This was likely a compensatory effect, with high early mortality of tadpoles resulting in survivors having more food less crowding. Greater food resources within low-quality treatments may also have been due to the increased phosphates and nitrogen levels enhancing periphyton growth (Smith 1982).

A recent meta-analysis showed that while survival and reproductive success

frequently differ between altered and reference wetlands, metamorphosis-related measures do not (Sievers et al. 2018). These measures can be influenced by other factors such as hydroperiod, predation and contamination (Edge et al. 2016), and can vary considerably between species and study systems (Earl and Whiteman 2015). Many studies have reported conflicting findings on the effects of pollution on metamorphosis-related measures, for example, in Ontario, Canada, tadpoles exposed to copper suffered reduced survival rates but increased growth rates (Leduc et al. 2016). However, the size differences observed here may still be ecologically important, and future studies should measure a suite of traits including post-metamorphic survival, growth and reproductive success to determine the full impact of environmental conditions on fitness.

Behaviour also differed between treatments. Tadpoles raised in more contaminated conditions did not avoid olfactory stimuli from predators, which will likely further reduce survival rates within low-quality wetlands. The attraction towards predator olfactory cues by tadpoles raised in the most contaminated conditions mirrors similar responses observed in Cuban tree frog tadpoles after exposure to the pesticide atrazine (Ehram et al. 2016). Interestingly, when both visual and olfactory cues were presented in concert, tadpoles raised in LQ treatments avoided predators (unlike when purely visual cues were provided when no response was observed). This result may be due simply to chance although it is also possible that there is some synergism occurring between the two sensory modalities (Hale et al. 2017). More work is needed to explore if this is a biologically meaningful result, and if so, its underlying cause.

Predator avoidance was negatively correlated with levels of the heavy metal quotient. Copper, lead and zinc were the three metals most correlated with the heavy metal quotient, and all three can affect predator avoidance of frogs (lead and zinc: Lefcort et al. 1998, copper in combination with elevated temperatures: Hayden et al. 2015). This interesting trend, and the fitness impacts of these altered behaviours, warrants further investigation.

Stormwater wetlands typically contain a mixture of contaminants, often at individual concentrations below those deemed biologically relevant (Allinson et al. 2017). Therefore, although frogs can discriminate and avoid unfavorable conditions (e.g. Vonesh and Buck 2007), their ability to do so may be reduced when they are exposed to

complex mixtures of contaminants. Consequently, females may lay eggs in sites that are highly polluted in multiple ways, rather than them being redirected to higher quality sites elsewhere, with implications for offspring fitness, and local and regional population persistence.

Habitat selection must be studied using an appropriate design to reflect the natural behaviour of the focal species. We offered breeding frogs a simultaneous choice between high- and low-quality conditions, an approach that is appropriate for urban stormwater treatment wetlands that are often made up of multiple, connected, sections, constructed in a deliberate sequence to maximise capture and treatment of contaminants (Lawrence and Breen 1998). Consequently, within a single stormwater wetland network, breeding amphibians may be able to choose between areas of differing contaminant concentrations, with particular sections (e.g. sediment settlement ponds) more likely to act as ecological traps. While research into ecological traps is still in its infancy, the discovery and apparent prevalence of traps is starting to increase along with recognition of the significant problem they pose for wildlife (Hale and Swearer 2016, Robertson and Chalfoun 2016, Hale and Swearer 2017). Regardless of how frogs are selecting sites, our results suggest that contaminated urban stormwater treatment wetlands are capable of reducing juvenile fitness, have the capacity to attract breeding frogs, and ultimately threaten population viability because of their capacity to function as ecological traps.

Management implications

Stormwater treatment wetlands can play an important role in supporting wildlife in urban areas (Hassall and Anderson 2015, Hill et al. 2017) but we need to know when there are conflicts between their intended function as stormwater treatment devices and the habitat requirements of animals. For example, when do wetlands that are ecological traps become detrimental to amphibian populations, and how can we manage this risk?

We highlight three potential options for managers. First, we may be able to actively manage wetlands to ensure they have suitable environmental conditions for animals. For example, remediating sediment to remove contaminants can improve habitat for wildlife, leading to greater amphibian richness and abundance (Stevens et al. 2002). Pre-treatment of runoff using hybrid filtration, deep sump catch basins, settling devices,

vegetated filter strips and sand filtration can also reduce contaminant inputs (Johir et al. 2010). A regular maintenance regime though has its limitations due to unknown pollutant accumulation rates, intermittent pollution events, and the substantial costs associated with intensive management regimes (Weiss et al. 2007).

The second option is to discourage behaviours (Greggor et al. 2014) that lead to frogs colonizing wetlands that are traps. Designing or modifying wetlands so they have steep banks and edges, for example, may make sites less attractive than those with the gently sloping, vegetated banks that suit most amphibian species. Likewise, removing fringing vegetation from wetlands eliminates a cue used by many amphibians to select oviposition sites (Egan and Paton 2004). These strategies are possible but require detailed knowledge of habitat selection behavior (e.g. which cues are used), and thus they have rarely been used (although see Robertson 2012, Greggor et al. 2014).

When improving habitat quality or reducing the likelihood of frog colonization is unfeasible, wetlands may not meet the dual objectives of stormwater treatment and suitable habitat for animals. The final option then would be to reduce the effects of wetlands that are ecological traps via landscape-planning. For example, we may be able to consider strategically planning where wetlands are being constructed in the landscape (e.g. away from other wetlands), or create nearby off-line habitats that are suitable for animals.

Ecological traps are an emerging conservation issue (Robertson and Chalfoun 2016). Identifying the potential for habitats to function as ecological traps – as we have done here – is important for minimizing the harm and realising the ecological potential of human-altered habitats. We need to better understand how a wider range of stormwater wetlands perform as habitat, particularly in terms of the broader landscape-level consequences when some stormwater wetlands are functioning as ecological traps. When traps exist, we recommend identifying the factors responsible for reduced fitness, determining the specific cues animals rely on when selecting habitats, and assessing the potential to manipulate these cues to eliminate the trap (Hale et al. 2015a). This information can then be used to determine if managing the local effects of traps is possible, or whether we need to take a more landscape-based approach. Ultimately, given the growth in stormwater treatment wetlands in cities around the world, minimizing the

ecological costs these artificial habitats can impose will be critical for sustaining wildlife in urban environments. This will require a greater understanding of the net effect of urban development on the availability of stormwater and natural wetlands within the landscape, and the effect of these changes on local and regional animal populations. This knowledge will be an important component of managing biodiversity in urbanized landscapes.

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Supporting Information

Additional supporting information may be found in the online version of this article at <http://onlinelibrary.wiley.com/doi/10.1002/eap.xxxx/supinfo>

Data Availability

Data available from Figshare: <https://doi.org/10.4225/49/5a8f946ddcb4e>

Tables

Table 1. Initial sediment quality data used to select and dichotomize the wetland pairs into high and low quality (HQ and LQ, respectively). All data collected by the Centre for Aquatic Pollution Identification and Management (www.capim.com). Note: Asterisk indicates lowest detection limit.

	Pair 1		Pair 2		Pair 3	
	HQ	LQ	HQ	LQ	HQ	LQ
	Lynbrook Estate Wetland	Chandler Rd	Woodlands Lake	Cheltenham Rd	Yarrabing Wetland	Ringwood Lake
<i>Heavy metals (mg kg-1 dry weight)</i>						
Aluminium	21100	16200	23800	20000	7480	9570
Antimony	2.5*	5	2.5*	2.5*	17	9
Arsenic	16	39	17	37	2.5	10
Barium	90	430	130	150	40	60
Beryllium	0.5*	0.5*	1	1	0.5*	0.5*
Boron	25*	25*	25*	25*	25*	25*
Cadmium	0.5*	2	0.5*	2	0.5*	0.5*
Chromium	45	73	43	48	12	21
Cobalt	10	19	10	13	2	3
Copper	72	299	45	113	14	40
Iron	25400	29300	26300	28000	6360	14300
Lead	29	176	24	283	13	48
Manganese	109	187	91	136	79	55
Mercury	0.1	0.4	0.1	0.4	0.05*	0.05*
Molybdenum	1*	13	8	2	1*	1*
Nickel	34	159	27	36	4	10
Selenium	2.5*	2.5*	2.5*	2.5*	2.5*	2.5*
Silver	1*	1*	1*	1*	1*	1*
Strontium	24	38	46	37	8	11
Thallium	2.5*	2.5*	2.5*	2.5*	2.5*	2.5*
Tin	2.5	48	2.5	11	2.5	2.5
Titanium	180	430	40	370	40	90
Vanadium	49	47	80	58	16	27
Zinc	606	3790	299	2390	138	597
HMQ	3.7	17.1	2.7	10.8	0.8	2.7
<i>Pesticides (ppm)</i>						
Bifenthrin	136	68.4	2	37.2	2.5	13
Diethyltoluamide	4.8	2.5	17.6	1*	20	15
Diuron	165	20	6	22	3	91
Fenamiphos	69.6	10	20	10	5	15
Permethrin	49.2	209	10	39.4	5	32.8

Prometryn	5	2.5	5	5	2.5	27
Pyrimethanil	1	7.6	4	1	1	5
Triclosan	69.6	155	20	80.8	10	60
Trifloxystrobin	2.5	2.5	5	2.5	20	5
Other						
Moisture						
Content	4.3	3	8	4.6	2.8	2.3
Total Organic						
Carbon	4.22	4.29	4.19	4.25	6.14	3.9
TPH	1940	21810	500	5420	1690	1170

Table 2. Output from analysis of variance (ANOVA) of linear models for the mesocosm experiment with treatment ('high' or 'low' quality conditions) and location (wetland pair) fitted as fixed effects. Significant p-values are in boldface. Asterisks indicate measures that were log-transformed to meet the assumptions of normality and homogeneity of variance.

Measure	Factor	df	MS	F	<i>p</i>
Survival	Treatment	1	0.180	5.10	0.030
	Location	2	0.087	2.40	0.104
	Treat × Loc	2	0.085	2.30	0.111
	Residuals	42	0.036		
*Mass at metamorphosis	Treatment	1	0.034	12.61	0.001
	Location	2	0.006	2.24	0.120
	Treat × Loc	2	0.006	2.09	0.137
	Residuals	40	0.003		
*Length at metamorphosis	Treatment	1	0.022	14.20	<0.001
	Location	2	0.006	3.97	0.027
	Treat × Loc	2	0.004	2.53	0.093
	Residuals	40	0.002		
*Developmental rate	Treatment	1	0.015	13.45	<0.001
	Location	2	0.036	3.18	0.052

	Treat × Loc	2	0.027	2.43	0.101
	Residuals	40	0.011		
*Periphyton	Treatment	1	0.0034	9.85	0.003
	Location	2	0.0005	1.35	0.270
	Treat × Loc	2	0.0001	0.40	0.673
	Residuals	42	0.0003		
*Ash-free dry-weight	Treatment	1	0.0022	10.30	0.003
	Location	2	0.0003	1.50	0.235
	Treat × Loc	2	0.0002	0.84	0.440
	Residuals	42	0.0002		

Table 3. Output from analysis of variance (ANOVA) based on a generalized linear mixed model for the predator avoidance trials with pair (source wetland pair), treatment (high- or low-quality) and cue (control, visual, olfactory or both) fitted as fixed effects (plus the interaction between treatment and cue), and site (source wetland) and tadpole nested within mesocosm fitted as random effects. P-values were calculated based on Satterthwaite's approximations, and significant values are boldface.

Factor	NumDF	DenDF	MS	F	p
Pair	1	1	0.02	0.15	0.765
Treatment	1	1	0.24	1.63	0.422
Cue	3	144	0.37	2.52	0.061
Treatment × Cue	3	144	0.61	4.16	0.007

Table 4. Output from analysis of variance (ANOVA) based on a linear model for the predator avoidance trials with treatment (high- or low-quality) and the heavy metal quotient (HMQ) fitted as fixed effects. Significant p-values are boldface.

Factor	df	MS	F	p
Treatment	1	19.53	7.19	0.012
HMQ	1	26.61	9.80	0.004

FIGURE LEGENDS

Fig. 1. Bivariate plots of model estimates (\pm SE) for survival and developmental rate, and length and periphyton dry weight based on linear models with treatment (high- or low-quality) and location (wetland pair) fitted as fixed effects.

Fig. 2. A subset of the sediment heavy metal concentrations (mg/kg), and the conductivity (microsiemens; μ S) and phosphate (PO_4 ; ppm) concentrations within mesocosm water. $N = 24$ per treatment. Asterisks indicate significant differences between treatments at $\alpha = 0.05$. For visualizations of all measured sediment and water variables see Appendix S5: Fig. S1.

Fig. 3. Mean ($+95\%$ CI) model estimates from a linear mixed effects model for the proportion of time spent in the 'away from predator' zone relative to the 'near predator zone' in the choice tank for both low-quality (LQ) and high-quality (HQ) treatments. The dashed line shows a one-to-one ratio between treatments.

Fig. 4. Relationship between the heavy metal quotient (log-transformed) of mesocosm sediments and predator odour avoidance behaviour of tadpoles. The heavy metal quotient is based on probable effect concentrations from MacDonald et al (2000). Grey smoothing represents standard errors based on output from general linear models using the predict function in R.