

## Predation of two common native frog species (*Litoria ewingi* and *Crinia signifera*) by freshwater invertebrates

Natasha J. Wilson<sup>A,C</sup>, Jamie E. Seymour<sup>B</sup> and Craig R. Williams<sup>A</sup>

<sup>A</sup>Sansom Institute for Health Research, School of Pharmacy and Medical Sciences, University of South Australia, Adelaide, SA 5000, Australia.

<sup>B</sup>Queensland Tropical Health Alliance, James Cook University, Cairns, Qld 4878, Australia.

<sup>C</sup>Corresponding author. Email: [natasha.wilson@mymail.unisa.edu.au](mailto:natasha.wilson@mymail.unisa.edu.au)

**Abstract.** The primary aim of this study was to identify aquatic invertebrate predators of amphibian eggs and tadpoles in an area of South Australia. The presence and abundance of aquatic invertebrates was monitored at four field sites for a period of 5–6 months; this revealed notonectids, freshwater crayfish and odonates to be amongst the most common invertebrate predator types. The ability of these predators to consume eggs and tadpoles of the native Australian frogs *Litoria ewingi* and *Crinia signifera* was then investigated. Freshwater crayfish (*Cherax destructor*) were the most prolific consumers of frog eggs and tadpoles. The notonectid *Enithares woodwardi* significantly impacted tadpole survivorship for both species while *Anisops* sp. was less successful at capturing and consuming these tadpoles. Caddisfly nymphs (*Lectrides varians* and *Leptorussa darlingtoni*) reduced egg survivorship but not to the same extent as *C. destructor*. Unlike some predators, which prey upon particular life stages, freshwater crayfish are large, polytrophic omnivores that can act as important predators of both anuran eggs and tadpoles. Given that predation is a key source of mortality in juveniles, identification of likely common predators is useful for understanding the regulation of amphibian populations.

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### Introduction

Preying on eggs and larvae, aquatic invertebrates can be an important contributor to amphibian mortality (Smith 1983; Alford 1999; Cruz *et al.* 2006). Despite this, invertebrate predators are typically less well studied than their vertebrate counterparts (Cabrera-Guzmán *et al.* 2012). Aquatic invertebrate predators are diverse and include several feeding modes: from those with biting and chewing mouth parts (e.g. odonate nymphs) to those with sucking mouthparts used to ingest internal body fluids (e.g. hemipterans). Several aquatic invertebrates are reported to prey on amphibian eggs and/or tadpoles; in particular, odonate naiads, aquatic hemipterans, beetles and freshwater crayfish are commonly reported predators (Axelsson *et al.* 1997; Alford 1999). However, limited data are available describing invertebrate predation of eggs and tadpoles for native amphibians in southern Australia.

In total, 27 frog species have been recorded in South Australia (Tyler and Walker 2011). The most abundant and common frogs recorded for the State are the common froglet (*Crinia signifera*), the spotted marsh frog (*Limnodynastes tasmaniensis*) and the southern brown tree frog (*Litoria ewingi*) (Walker 2003). Each of these species has a different mode of egg deposition which may in turn influence rates of predator encounter (Magnusson and Hero 1991; Stebbins and Cohen

1995). *C. signifera* lays eggs attached to submerged vegetation in small clusters of up to 30 eggs (Williamson 1988; Williamson and Bull 1995). *L. tasmaniensis* deposits spawn in foam nests on open water while the eggs of *L. ewingi* are typically attached in large clumps to the submerged tips of vegetation hanging into the water (Tyler and Walker 2011). All three species may co-occur in ponds in southern Australia (N. J. Wilson, pers. obs.) yet may have differing levels of vulnerability to aquatic invertebrate predators.

One population of *C. signifera* located at Bridgewater in the Adelaide Hills was previously studied extensively across three breeding seasons, identifying several invertebrate predators. The most abundant predators of *C. signifera* tadpoles included naiads of two dragonfly species (*Aeshna brevistyla* and *Hemicordulia tau*), larvae of the damselfly *Austrolestes annulosus* and the larvae of the dytiscid beetle *Rhantus suturalis*, which also acted as an egg predator (Williamson 1988; Williamson and Bull 1999). Both *A. annulosus* and *R. suturalis* were restricted to preying on small tadpoles (Williamson 1988). Although capable of taking larger tadpoles, both *A. brevistyla* and *H. tau* showed a preference for small tadpoles (Williamson 1988). The naiad of the dragonfly *H. tau* was also shown to prey on the tadpoles of *L. tasmaniensis*, *L. ewingi* and *Pseudophryne bibronii* (Richards and Bull 1990a, 1990b; Peterson *et al.*

1992). Caddisfly (*Leptorussa darlingtoni*) nymphs have been identified as potential predators of frog eggs in southern Australia, with reports that they fed on frog spawn when limited resources were available (Jackson 1984). Aside from these few accounts, little information exists on likely common invertebrate predators of immature frog stages in southern Australia.

Compared with the invertebrate predators of the rest of the country, invertebrate predators of northern Australia have received a little more attention. Specifically, researchers have investigated the capacity of invertebrate predators to consume the toxic eggs and tadpoles of the invasive cane toad, *Rhinella marina* (= *Bufo marinus*) (Crossland 1998; Crossland and Alford 1998; Cabrera-Guzmán *et al.* 2012). *R. marina* is toxic to many predators, owing to a suite of bufadienolides (cardiac glycosides) present in the eggs, larvae and the parotid glands of adult toads (Hayes *et al.* 2009). Since its introduction to Australia in 1935 the dispersal range of this invasive pest has continued to expand (Urban *et al.* 2007; Tyler and Knight 2009). Projections of the maximal distribution of *R. marina* have been modelled using existing distribution and bioclimatic data and suggest that the risk of colonisation extends to southern Australia (van Beurden 1981; Urban *et al.* 2007). Establishing which anuran predators are the most common is of interest when considering the potential risks posed to these predators with range expansion of *R. marina*.

While the impacts of this noxious amphibian have been well studied for many vertebrate predators (e.g. Phillips *et al.* 2003; Crossland *et al.* 2008; Letnic *et al.* 2008; Hagman *et al.* 2009; Greenlees and Shine 2011) less research has been conducted on invertebrate predators (Crossland and Alford 1998; Punzo and Lindstrom 2001; Ward-Fear *et al.* 2010; Cabrera-Guzmán *et al.* 2012). Previous investigation of invertebrate predators of *R. marina* has produced mixed results. Several invertebrate groups, including crayfish, beetles, odonates and nepids are reported to consume immature life stages of *R. marina* without ill affect (Crossland 1998; Cabrera-Guzmán *et al.* 2012). In contrast, other invertebrate predators such as leeches have succumbed to toxin ingestion and have died after feeding on immature life stages of *R. marina* (Crossland and Alford 1998). Further, some predators, such as dytiscid beetles and belostomatid bugs, can successfully prey upon particular life stages but ingestion of other developmental stages has been associated with mortality, suggesting ontogenetic variation in toxicity (Crossland 1998; Crossland and Alford 1998). If the invertebrate predators of native frog eggs and tadpoles are known, then we can study these species further to determine their susceptibility to toxins present in *R. marina*.

The primary aim of this study was to identify invertebrate predators of eggs and tadpoles of native amphibians. This was achieved by first identifying invertebrate predators abundant at field sites where frogs were present, and then determining the relative rate of egg/tadpole consumption by these putative predators in the laboratory. We sought to identify which predators may potentially play important roles in regulation of amphibian populations. This study is an initial step in identifying common invertebrate predators of native amphibians to inform further studies of these predators for their ability to consume toxic eggs and tadpoles of *R. marina*.

## Methods

### Study sites

Four field sites were assessed for the presence of frogs and putative invertebrate predators. Two sites were located within metropolitan Adelaide, South Australia, namely a groundpool at Globe Derby Park (34°47'3.93"S, 138°35'12.39"E), and an artificial watercourse situated within the grounds of the Grange Golf Club in Seaton (34°53'4.80"S, 138°30'3.06"E). Outside of the city, a wetland in Milang (35°24'20.77"S, 138°58'37.80"E) and, finally, what would become the focal site for numerous frog egg collections, a pond at Bridgewater (35°0'13.24"S, 138°44'55.62"E) in the Adelaide Hills, were also studied.

Seaton, Milang and Globe Derby Park were all sampled monthly in 2011 from August to December inclusive whereas the Bridgewater site was sampled from May to October in 2012. These sites were selected as they represent a diversity of common habitat types in southern Australia. A decision was made to include the Bridgewater site, which was sampled at a later date, after it was identified as a site of high frog abundance. The waterbodies surveyed in this study ranged in size from 160 to 4500 m<sup>2</sup>.

### Establishing the presence of adult frogs

Audio strip transects adapted from Heyer *et al.* (1994) were used to count and identify calling adult males for each of the study sites. The total perimeter of each waterbody was sampled. One of us (NJW) stood quietly and stationary for 3 min at an initial sampling point and made note of calling frogs. Each additional sampling point was situated 25 m from the previous one, walking along the edge of the waterbody. Owing to the varying sizes of the sites, the number of sampling points ranged from just one at Seaton to 13 at Milang.

### Evaluating presence and abundance of aquatic invertebrates, frog eggs and tadpoles

Tadpoles and aquatic invertebrates were sampled using a sweep net (260 × 180 mm frame, 0.3 mm mesh, Australian Entomological Supplies, Bangalow, NSW). For each location, a minimum of 10 sweeps, each 1 m in length, were performed following a protocol previously used by Gunzburger (2007). If additional species (i.e. not represented in any of the previous sweeps) were collected in the tenth sweep, additional sweeps were performed until the last 10% of sweeps netted no additional species (Heyer *et al.* 1994). The same researcher performed each sweep at all sites; sweep location was randomised using a random number table. Tadpole identification was assisted with a guide published by Anstis (2007).

All invertebrates collected in sweep nets were identified using Williams (1980). At a minimum, freshwater insects were classified to order. Due to the diversity and volume of organisms collected in field sampling, only invertebrates selected for predation trials have been identified to species level. Opera-house-style nets (700 mm L × 480 mm W × 190 mm H with two entry points that are both 75 mm in diameter) were baited with fresh meat and set for 16 h overnight to monitor waterbodies for crayfish.

The visual encounter technique was employed to search methodically for frog eggs at field sites. Each waterbody was searched along a series of transects that began at the margin and ran to the midpoint (or to a maximum depth of 45 cm). All frog eggs within 1 m of the transect were counted following Heyer *et al.* (1994).

#### Laboratory predation trials

Feeding trials were conducted in the laboratory to establish consumption of frog eggs and tadpoles by a subset of the putative predators identified in field surveys. Some predators identified in field surveys were not present across the time frame that frog eggs and tadpoles were sampled (and thus not available for study) or were not present in sufficient abundance to perform a reasonable number of replicate feeding trials. Pilot trials conducted with the nepid *Ranatra dispar*, the dragonfly *Adversaeschna brevistyla* (Aeshnidae), adult coleopterans, and larval ephemeropterans suggest that these predators consume frog eggs and/or tadpoles. However, due to insufficient replication, these data are not presented. All frog eggs, tadpoles and predators (listed in Table 1) for laboratory trials were collected in the field. Field collections were transported to the laboratory in an insulated container and sorted immediately. Frog eggs and tadpoles were identified to species (Anstis 2007) and then staged according to Gosner (1960).

All predators were fasted for 24 h before onset of feeding trials. Predators were always tested in isolation in individual experimental tanks; the number of replicates performed for each predator treatment are listed in Table 1. Feeding trials ran for 72 h following introduction of prey. Each predator was offered either 10 live eggs (Gosner (1960) Stage 1–20) or 10 live tadpoles (Gosner Stage 21–26) and prey consumption was monitored across the 72-h period. Control trials ( $n = 6$  for *L. ewingi* eggs, 11 with *L. ewingi* tadpoles, 7 for *C. signifera* eggs and 10 for *C. signifera* tadpoles) without predators were also conducted to determine baseline survivorship levels for eggs and tadpoles. Allocation of eggs and tadpoles to control and predator treatments was randomised.

All predators were tested in glass aquaria (100 mm L × 100 mm W × 200 mm H filled with 1 L aged tap water) except for crayfish, which were tested in 9-L containers (250 mm diameter) filled with 5 L of aged tap water. Trials were conducted in a temperature-controlled room set at 22°C with a 12 h light:dark cycle. Aquaria habitat was simplified and included a washed gravel substrate but no refuge.

As an extension of identifying invertebrates that prey on frog eggs and tadpoles, we also examined the rate of consumption to identify which predators could be considered major predators of frog eggs and/or tadpoles. We adopted the definition for a major predator as proposed by Portheault *et al.* (2007), that is, any predator consuming more than 50% of the eggs or tadpoles offered was considered to be a major predator. We considered that this would identify predators that may significantly affect survivorship of amphibian eggs and tadpoles in the field and thus may play important roles in population regulation.

#### Statistical analysis

Egg and tadpole survivorship in control and predator treatments was analysed with the Kruskal–Wallis test (owing to non-normally distributed data). Dunn's Multiple Comparisons *post hoc* test was used to compare prey survivorship for each predator treatment to survivorship in the predator-free control (Zar 1999). If a predator significantly impacted egg or tadpole survivorship of both frog species we used the Mann–Whitney test to determine whether the feeding rate differed with *C. signifera* and *L. ewingi* prey.

We also used the Mann–Whitney test to determine whether predators that reached major predator status were significantly larger than those predators that consumed fewer than 50% of tadpoles in feeding trials. Predator size was determined by measuring body length (BL), which was defined as the distance between the anterior of the head and the posterior of the terminal abdominal segment. Crayfish were excluded from this analysis because they were more than nine times larger than all other tadpole predators studied and hence would heavily bias the data. Finally, an additional Mann–Whitney test was used to determine whether BL was significantly different between the two notonectid predators studied. Statistical significance was set at  $\alpha = 0.05$  for all tests performed.

## Results

#### Putative invertebrate predator abundance

Notonectids and odonates were common to all waterbodies surveyed and ranked amongst the highest in terms of overall abundance compared with other predatory invertebrates (Table 2). Notonectid density was greatest at Milang, for which the mean catch ( $\pm$ s.e.m.) was  $24.2 \pm 17.1$  individuals per dip. Caddisfly larvae were particularly common at Bridgewater and were also sampled on occasion at both Milang and Seaton. The common yabby (*Cherax destructor*) was detected at all sites

**Table 1.** The number of field-collected invertebrate predators offered *Litoria ewingi* (LE) and *Crinia signifera* (CS) eggs and tadpoles in feeding trials (FT)

Predator	Life history stage	Body length (mean $\pm$ s.d.) (mm)	Feeding mode	$N_{(\text{egg FT})}$		$N_{(\text{tadpole FT})}$	
				LE	CS	LE	CS
<i>Anisops</i> sp. (Notonectidae)	Adult	6.1 $\pm$ 0.6	Pierce and suck	11	10	10	11
<i>Enithares woodwardi</i> (Notonectidae)	Adult	10.7 $\pm$ 1.5	Pierce and suck	19	6	12	8
<i>Lectrides varians</i> (Trichoptera)	Larvae	11.6 $\pm$ 2.5	Graze and shred	13	10	0	0
<i>Leptorussa darlingtoni</i> (Trichoptera)	Larvae	7.4 $\pm$ 1.5	Graze and shred	10	11	0	0
<i>Cherax destructor</i> (Decapoda)	Adult	104.1 $\pm$ 12.7	Bite and chew	10	10	10	10
Coenagrionidae (Odonata)	Nymph	9.2 $\pm$ 1.3	Bite and chew	0	0	0	6

except for Milang. Many of the invertebrates sampled in field site surveys were recognised by the researchers as strict herbivores and these species are not shown in Table 2.

*Anuran field records*

Three native species were recorded across the four field sites. *C. signifera* and *L. tasmaniensis* were detected at all sites at least once and *L. ewingi* was also recorded at the Bridgewater site (Table 3). Audio strip transects detected amphibians more frequently than did visual encounter egg surveys or tadpole dip netting, with adult frogs frequently detected while eggs and tadpoles were often absent (Table 3). The search time required to inspect each waterbody for eggs during the visual encounter

surveys varied between sites, ranging from ~20 to 90 min depending on egg visibility. Egg visibility was lower in deep or turbid water and at densely vegetated sites, increasing the search time required to complete the survey.

Consistent with the literature, *L. ewingi* eggs (mean clutch size ± s.d., 44.9 ± 32.3) were almost always found on the submerged tips of vegetation that was overhanging the pond, such as reeds/rushes (Tyler and Walker 2011). *C. signifera* attached eggs to submerged vegetation either individually or in small clumps (4.6 ± 4.8) while *L. tasmaniensis* deposited eggs in foam nests (91.0 ± 37.2). It was decided to focus on predator interactions with *C. signifera* and *L. ewingi* as the eggs and tadpoles of these frogs were by far the most abundant. Bridgewater became the focal field site for collection of

**Table 2. Abundance (mean per dip ± s.e.m.) and seasonality of putative invertebrate predators at field sites in South Australia as determined by sweep netting**

Order	Globe Derby Park		Milang		Seaton		Bridgewater	
	Per dip	Present	Per dip	Present	Per dip	Present	Per dip	Present
<i>Insecta</i>								
Coleoptera	3.3 ± 1.7	Aug–Nov	2.7 ± 3.2	Aug–Dec	0.5 ± 0.5	Aug–Nov	<0.1	Jul–Aug
Ephemeroptera	–	–	–	–	2.6 ± 6.5	Aug	<0.1	Jul–Aug
Hemiptera (Nepidae)	–	–	–	–	–	–	0.17	May–Aug
Hemiptera (Notonectidae)	2.8 ± 1.8	Aug–Nov	24.2 ± 17.1	Aug–Dec	19.0 ± 30.5	Aug–Nov	12.8	May–Aug
Hemiptera (other)	7.0 ± 2.4	Aug–Nov	2.3 ± 3.4	Sep–Dec	–	–	–	–
Odonata	2.0 ± 0.9	Aug–Oct	4.2 ± 5.0	Aug–Nov	4.0 ± 4.4	Aug–Nov	0.2	May–Aug
Trichoptera	–	–	0.2 ± 0.4	–	0.3 ± 0.8	Sep	7.2	May–Aug
Total insects	15.1		33.6		26.4		20.4	
<i>Mollusca</i>								
Gastropoda	5.8 ± 2.9	Aug–Nov	0.1 ± 0.2	Oct	9.0 ± 7.1	Aug–Nov	<0.1	May–Aug
<i>Crustacea</i>								
Decapoda <sup>A</sup> ( <i>C. destructor</i> )	Present		–		Present		Present	

<sup>A</sup>Presence determined by visualisation of individuals in water, and/or moults, and/or present in traps, and/or juveniles in sweep nets.

**Table 3. Detection of frog eggs (E), tadpoles (T) and calling adult males (A) for each of the four sites**  
CS, *Crinia signifera*; LT, *Limnodynastes tasmaniensis*; LE, *Litoria ewingi*

Month (2011)	Globe Derby Park			Milang			Seaton			Bridgewater		
	E	T	A	E	T	A	E	T	A	E	T	A
Aug. 2011	–	–	CS: 2 LT: 1	–	–	CS: 3	–	–	CS: 10			
Sep. 2011	CS: 9 LT: ~200	–	CS: 1	–	–	CS: 1	LT: 107	–	CS: 2			
Oct. 2011	–	–	CS: 1 LT: 1	–	–	CS: 1 LT: 2	LT: 82	–	CS: 1 LT: 2			
Nov. 2011	–	–	CS: 1	–	–	–	–	–	CS: 1			
Dec. 2011	–	–	–	–	–	–	–	–	–			
May 2012										CS: ~100 LE: ~500	LE: ~50	LE: 4
Jun. 2012										CS: 20 LE: ~190	CS: 4 LE: 17	CS: 2 LE: 6
Jul. 2012										CS: 82 LE: 167	CS: 9 LE: 12	CS: 2 LE: 4
Aug. 2012										CS: 48 LE: 128 LT: ~200	CS: 7 LE: 21	CS: 3 LE: 1 LT: 2

both invertebrate predators and amphibian prey for laboratory trials.

#### Predation on amphibian eggs

Notonectids (*Anisops* sp. and *Enithares woodwardi*) did not significantly affect egg survivorship for either species (Fig. 1). Although never reaching statistical significance, exposure of both *L. ewingi* and *C. signifera* eggs to the caddisfly larvae *Lectrides varians* and *L. darlingtoni* caused a 5–10% reduction in egg survivorship compared with controls (Fig. 1). The freshwater crayfish (*C. destructor*) was a major predator, significantly reducing egg survivorship of both species. Egg survivorship was reduced to 13% and 26% for *L. ewingi* ( $\chi^2=27.82$ ,  $P=0.001$ , Dunn's  $Q=3.15$ ) and *C. signifera* ( $\chi^2=19.94$ ,  $P=0.001$ , Dunn's  $Q=3.21$ ) respectively (Fig. 1). Prey identity was not a significant factor in rate of egg consumption by *C. destructor* ( $z=0.76$ ,  $P=0.456$ ).

#### Predation on tadpoles

*L. ewingi* (mean  $\pm$  s.d. SVL  $3.3 \pm 0.4$  mm) and *C. signifera* (SVL  $3.7 \pm 0.7$  mm) tadpoles were offered to the same notonectid and crayfish predator species that were offered eggs. Additionally, we also offered *C. signifera* tadpoles to coenagrionid damselfly nymphs. As depicted in Fig. 2, *Anisops* sp. was observed to consume some *L. ewingi* tadpoles, reducing survivorship to 86% compared with the control. However, *Anisops* sp. did not significantly impact survivorship of either anuran. In contrast, *E. woodwardi* was a major predator of tadpoles ( $\chi^2=32.11$ ,  $P=0.001$ , Dunn's  $Q=4.53$ ). *Enithares woodwardi* consumed all available *L. ewingi* tadpoles in feeding trials. A significant reduction in *C. signifera* survivorship was also observed: just 21% of tadpoles survived exposure to *E. woodwardi* predators ( $\chi^2=40.84$ ,  $P=0.001$ , Dunn's  $Q=3.66$ ) (Fig. 2). Prey identity did influence tadpole consumption rates by *E. woodwardi*:

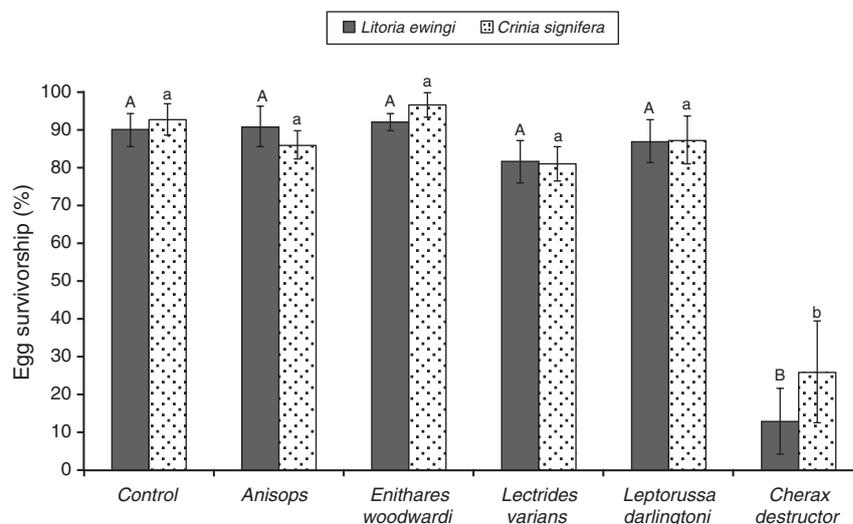
significantly more *L. ewingi* tadpoles were consumed than *C. signifera* tadpoles ( $z=3.04$ ,  $P=0.002$ ). *C. destructor* was again a major predator and always consumed all available tadpoles, regardless of prey species (*L. ewingi*: Dunn's  $Q=4.74$ ; *C. signifera*: Dunn's  $Q=4.94$ ). Coenagrionid damselfly nymphs consumed some *C. signifera* tadpoles but did not significantly reduce survivorship when compared with the control group. Surviving tadpoles in the damselfly treatment often sustained tail damage, indicating that they escaped following capture by a predator. Tadpoles in the control group were never observed to have any tail damage.

#### Influence of predator size

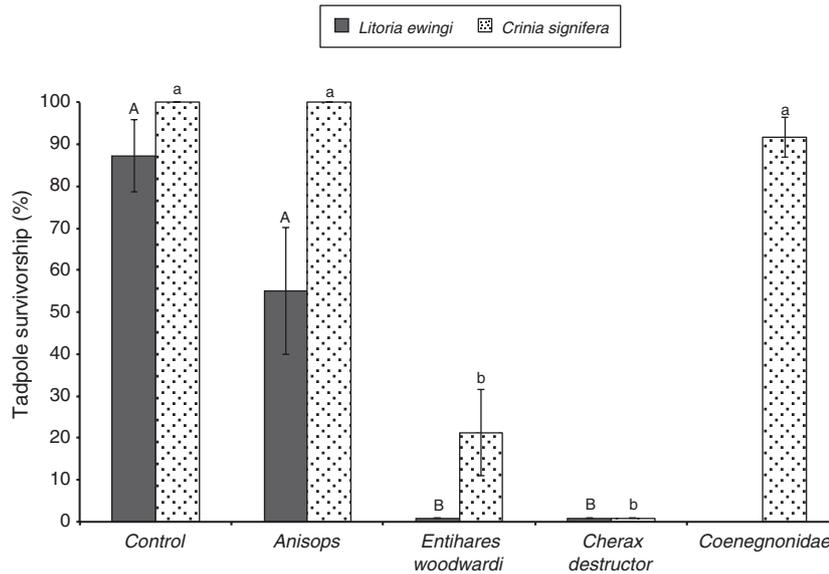
Predators that consumed more than 50% of offered tadpoles were significantly larger (as measured by BL) than predators that consumed less than 50% of tadpoles; this held true when crayfish were excluded from the analysis ( $z=3.68$ ,  $P=0.002$ ). There was a significant size difference between the two notonectid groups studied here: *Enithares woodwardi* were substantially larger than the *Anisops* predators (BL:  $10.7 \pm 1.5$  and  $6.1 \pm 0.6$  mm respectively) (Table 1) and this difference was statistically significant ( $z=8.88$ ,  $P \leq 0.001$ ). This size difference was of interest given the predation rate differences between them.

#### Discussion

Field surveys of the study sites, which represented a range of habitat types from natural groundpools to artificially constructed wetlands, identified the presence of three frog species: *C. signifera*, *L. ewingi* and *L. tasmaniensis*. Frog eggs and tadpoles were most abundant at the Bridgewater site, which had one of the lowest total number of putative predators per dip in sweep surveys. In comparison, a high number of putative predators were recorded for Milang, where frog eggs and tadpoles were never detected. This result is tentatively suggestive that



**Fig. 1.** Mean survivorship of *Litoria ewingi* and *Crinia signifera* eggs following exposure to predators in laboratory feeding trials. Error bars depict the standard error of the mean (s.e.m.) for each treatment. Significant differences between egg survivorship of control and predator treatments is denoted with uppercase letters for *L. ewingi* and lowercase letters for *C. signifera*.



**Fig. 2.** Survivorship of *Litoria ewingi* and *Crinia signifera* tadpoles following exposure to predators in laboratory feeding trials relative to the control. Error bars depict the standard error of the mean (s.e.m.) for each treatment. Significant differences between tadpole survivorship of control and predator treatments is denoted with uppercase letters for *L. ewingi* and lowercase letters for *C. signifera*.

predator density may be an important predictor of egg and tadpole abundance for a given site. However, extrapolation from field data based on four sites should be considered cautiously.

Freshwater crayfish had the highest, per individual, consumption of amphibian eggs and tadpoles and were the only predators studied that preyed on both life stages. Crayfish are large invertebrates and have been identified by others (Momot 1995; Nyström *et al.* 1996) as keystone predators in freshwater environments, particularly in habitats lacking predatory fish. However, considering the high abundance of notonectids observed at field sites, and their size relative to crayfish, these aquatic hemipterans are also likely significant predators of young tadpoles/hatchlings in field settings. Collectively, notonectids may very well apply higher predation pressure than freshwater crayfish. Yet, unlike notonectids, freshwater crayfish are capable of preying on older, larger tadpoles (N. J. Wilson, unpubl. data). Comparison of the predation pressure exerted by crayfish and notonectids is beyond the scope of this paper and warrants further investigation.

Body length appeared to be an important determinant of tadpole predation success for notonectids. *Enithares woodwardi* was a major predator and significantly affected tadpole survivorship while the smaller *Anisops* notonectids were less successful despite observed attempts to strike tadpoles (N. J. Wilson, unpubl. data). The short BL of *Anisops* may limit their ability to overpower tadpole prey: size can be a limiting factor in predation success and has been described previously for notonectid predators (Cronin and Travis 1986; Streams 1994).

Notonectids did not prey on anuran eggs. This observation is consistent with the literature, which indicates that notonectids rely on vibration to detect and localise prey and thus mobility is a prerequisite of prey items (Murphey and Mendenhall 1973).

Although they did not consume large quantities, caddisfly larvae appeared to have some impact on egg survivorship; this is a significant observation given that, to our knowledge, only one other report of consumption of amphibian eggs exists for Australian caddisflies (Jackson 1984). Survivorship of frog eggs in our feeding trials with *L. darlingtoni* scarcely deviated from the control, yet *L. darlingtoni* were previously reported to feed on frog eggs when food resources were limited (Jackson 1984). Given this, it is possible that consumption rates in feeding trials could increase with an extended fasting period. The lack of reports on caddisflies as amphibian predators within Australia is in contrast to the situation in North America and Europe, where caddisfly larvae are commonly reported to consume amphibian eggs (e.g. Murphy 1961; Stout *et al.* 1992; Majecki and Majecki 1996, 1998; Gall and Brodie 2011).

Damselfly nymphs did not prey significantly on tadpoles in this study. We suspect that the short BL of the damselfly nymphs used in feeding trials limited predation success. It has been previously noted that damselfly nymphs are successful at preying only upon small tadpoles (Williamson 1988). Tail injuries in surviving tadpoles were common (N. J. Wilson, unpubl. data) and suggest that damselfly nymphs were able to capture tadpoles but unable to overpower them.

The consumption rates reported here cannot reasonably be directly translated to estimates of predation in field settings and may better be thought of as an estimate of maximum rate of consumption. One limitation to this investigation is that predators were not provided with alternative prey. Provision of alternative prey may influence the consumption rates observed for these frog eggs and tadpoles. Predators do not typically feed indiscriminately and provision of alternative prey can influence consumption rates (Gunzburger and Travis 2005). Alternative

prey, particularly in the form of macroinvertebrates, were present at the field sites surveyed and likely influence prey selection in the natural setting. One avenue of further study, which would further understanding of predator choice behaviour, would be to redesign the feeding trials as choice experiments and offer frog eggs or tadpoles in combination with an alternate prey that was abundant at field sites.

These feeding trials were conducted in small aquaria that lacked refugia and in no way mimic the habitat complexity seen in natural waterbodies. The simplistic nature of the laboratory set-up likely forces prey into closer contact with predators than would be encountered under normal circumstances in the field and probably increases capture rates (Alford 1999). Habitat structure, such as the presence of vegetation, has been shown to influence the outcome of predator–prey interactions. In one study, predation on tadpoles by dragonfly naiads and belostomatids was high in the absence of any vegetation (Tarr and Babbitt 2002). Another study demonstrated that tadpole survivorship was greater with high vegetation cover compared with lower levels of vegetation cover (Babbitt and Tanner 1997).

However, the provision of greater habitat complexity in this study may not have necessarily increased prey survival. Previous work has showed that provision of vegetation appears to offer no survival advantage against crayfish predators (Figiel and Semlitsch 1991). In some instances, vegetation has been shown to facilitate predation and thus reduce tadpole survivorship. Vegetation was reported to provide crayfish a framework for greater access to the upper regions of the water column as well as places to hide and wait for prey in one study (Davis *et al.* 2012).

Our research focus was to identify invertebrate predators of common and abundant tadpoles. In doing so, we aimed to identify predators that would likely encounter the eggs and tadpoles of the cane toad *R. marina* should the invasion front reach southern Australia. Although not specifically reporting on the same species investigated in this study, freshwater crayfish, odonate larvae and notonectids collected from within the present dispersal range of *R. marina* in northern Australia have been reported to consume immature developmental stages of *R. marina* (Crossland 1998; Crossland and Alford 1998). This suggests that, if encountered, the crayfish, notonectids and damselfly nymphs identified at field sites investigated in this study may attempt to prey on *R. marina* eggs and/or tadpoles. Future research could assess how these southern Australian predators fare against *R. marina* eggs and tadpoles in comparison to their northern (*R. marina* exposed) counterparts.

This work does not definitively describe the relative impact of different predators on amphibians. However, by identifying the most likely invertebrate predators and characterising their capacity for predation at an individual level, we have advanced our understanding of the role of such predators feeding on immature frogs in southern Australia. The freshwater crayfish (*C. destructor*) was a very efficient predator in laboratory trials, although it is not as numerous at field locations as notonectid hemipterans. Both may play important roles as regulators of amphibian assemblages. This study may have important implications for the design and management of wetland habitats and could inform conservation programs to assist with the protection of locally threatened frog species. An important future direction would be to research predation of the eggs and tadpoles

of rarer frogs; these predators could have a significant impact on the survival of small or fragmented frog populations.

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