

# The effects of suspended sediments on the tadpoles of two stream-breeding and forest dwelling frogs, *Mixophyes balbus* and *Heleioporus australiacus*

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## ABSTRACT

Tadpoles of two stream-breeding, forest-dwelling frogs (*Mixophyes balbus* and *Heleioporus australiacus*) were exposed to multiple short-term suspended sediment influxes, simulating storms. Three sediment concentrations were represented (control, low - 200 mg·L<sup>-1</sup>, and extremely high - 1655 mg·L<sup>-1</sup>) to approximate concentrations recorded in previous hydrological studies. Growth for both species and survivorship (individuals had lost a designated body mass) for *M. balbus* did not differ significantly between the three treatments over the 18-week experimental period. Survivorship of *H. australiacus* tadpoles was higher in the high sediment regime. More tadpoles had sediment on gill surfaces in sediment treatments than controls in both species. The occurrence of sediment on the gills did not increase with increased exposure to sediment. Tadpoles of *H. australiacus* appear to require a relatively low ( $\leq$  pH ~6.5) and constant water pH and the apparent advantage to tadpoles of *H. australiacus* of a high sediment load, probably reflects their sensitivity to pH rather than an affect of sediment *per se*. Tadpoles of both species are probably capable of surviving in streams that are subjected to brief periods of exposure to raised suspended sediments.

**Key words:** anurans, suspended sediment, conservation, management, forestry, survivorship.

## Introduction

Many human activities have caused reductions or elimination of many populations of amphibians, including frogs (Blaustein and Wake 1990, Pechmann and Wilbur 1994, White 1995). The frogs apparently most susceptible to decline on the east coast of Australia breed in streams and the adults are closely associated with stream habitats (Mahony 1996). The factors causing declines of frogs in general are not certain (Pechmann *et al.* 1991, Pechmann and Wilbur 1994), but forestry activities are a factor potentially impacting stream dwelling frogs in eastern Australia (Gillespie and Hines 1999, Hines *et al.* 1999, Lemckert 1999).

An increase in soil erosion is one impact of forestry practices that may affect frogs, as increased sediment concentrations potentially reduce the respiratory capabilities of tadpoles by fouling their gills (deMaynadier and Hunter 1995). Higher mean turbidity occurs in streams in logged, compared to unlogged catchments in New South Wales (Cornish 1980, 1981, 1983). The sources of increased sediment from timber harvesting include road construction, soil disturbance during tree-felling and post-logging burns (Beschta 1978; Campbell and Doeg 1989, Doeg and Koehn 1990, Cornish 2001). Erosion can be particularly important in south-eastern Australia because:

1. most of the rivers arise in forest, so forestry operations in uplands can affect a large proportion of the downstream habitats (Campbell and Doeg 1989).

2. erosion potential is frequently high due to a combination of steep slopes and high rainfall and the soils commonly have naturally high clay contents (Papadakis 1969, Rosewell 1993).
3. the extremely variable annual rainfall (Cremer 1990) makes it difficult to determine the optimal time for timber harvesting or road construction to minimise erosion because the rate at which suspended sediment enters streams is related directly to the frequency and intensity of storms (Olive and Rieger 1987).

The effects of logging on stream habitats can result in complex changes in communities of resident fauna over the short and long term (Campbell and Doeg 1989, Corn and Bury 1989). Small increases in sediments that contain large quantities of organic matter may actually increase stream productivity (Murphy *et al.* 1981), although some forestry practices can increase sediment loads in streams above thresholds considered to be detrimental to stream communities (Murphy *et al.* 1981). Long-term reductions in the densities of filter feeding invertebrates occur in streams receiving high concentrations of suspended sediment from forestry practices (Campbell and Doeg 1989), with the filter-feeding mechanisms and gills becoming clogged by inorganic particles, causing death (Lemly 1982, Campbell and Doeg 1989). Increased siltation can cause physiological and behavioural changes in adult fish and affect the development rate and survival of larval fish in the field (Berkman and Rabeni 1987) and

laboratory (Doeg and Koehn 1990). Stream-dwelling tadpoles respire using gills in the generally oxygen rich water (Feder 1981) and so may be affected similarly to fish, but we know of no laboratory studies on the impact of sediment on tadpoles. Deposited sediments may be able to impact upon tadpoles by affecting the periphyton used for food, leading to an apparent reduced food quality and/or availability and commensurate reduced growth rates (Gillespie 2002). How universal this effect may be is not known.

We set up a series of laboratory experiments to measure the effect of increased concentrations of suspended sediment on growth, survivorship and the gills of two stream-dwelling tadpoles, the Southern Barred Frog *Mixophyes balbus* and Giant Burrowing Frog *Heleioporus australiacus*. These species were selected because both have declined in recent times (Gillespie and Hines 1999) and both occur in forested areas that are subject to logging and roading (Lemckert and Brassil 2003), activities that may elevate the concentrations of suspended sediment in streams used for breeding. Sediment enters streams primarily after storms (Olive and Rieger 1987) and subsequent settlement means that high concentrations of sediment are generally transient. Thus, we investigated the effects of transient sediment influxes on the health of tadpoles, using concentrations of sediment and rates of sediment introduction that simulate the short-term pulses of suspended sediment observed in field conditions.

## Materials and methods

### Collection and husbandry of tadpoles

Tadpoles of *M. balbus* (n=132) were collected from the Crawford River near Bulahdelah, NSW (33°22'30S; 152°06'30E) on 19<sup>th</sup> March 1997, and tadpoles of *H. australiacus* (n=159) were collected at Fawcett's Dam near Ourimbah, NSW (151°19'E; 33°17'S) on 4<sup>th</sup> April 1997. These tadpoles were transferred to the University of Sydney and initially each species was housed communally in one of two 400 mm × 190 mm × 230 mm glass aquaria.

Experimental work commenced on 12<sup>th</sup> April 1997, with 40 tadpoles from each species being assigned randomly to each of one of the three sediment treatments (=120 tadpoles per species) and placed individually into their experimental "tank". These tanks were round CR30 transparent plastic vessels (100 mm high, bottom inside diameter 90 mm and top inside diameter 115 mm) holding 500 ml of water. Water temperature was maintained at 18 °C with a 12L:12D photoperiod. Tadpoles were housed individually to eliminate any possible confounding effects from competition, crowding and/or waterborne growth inhibitors that could retard growth (Petranka 1989; Gromko et al. 1973; Semlitsch and Caldwell 1982). Half the water in each container was changed weekly. No sediment was added to the experimental treatments for 39 days to give the tadpoles time to acclimate to the new conditions. Tadpoles were fed boiled lettuce and spinach *ad libitum* and commercial fish food (Wardley™ Community Bites Mini Floating-sinking Food).

Water in which tadpoles were initially kept was prepared from milli-Q (reverse osmosis) water by adding sodium ions (Table 1) to a concentration of 1.5 mmol·L<sup>-1</sup> Na<sup>+</sup> (approximately the average value for water from the two collections sites of the tadpoles). Additional ions were added (Table 1) to approximate concentration in natural stream water in NSW (Cornish, 1978).

| Ion                               | Concentration (mmol·L <sup>-1</sup> ) |
|-----------------------------------|---------------------------------------|
| NaCl                              | 1.25                                  |
| NaHCO <sub>3</sub>                | 0.25                                  |
| KCl                               | 0.05                                  |
| CaCl <sub>2</sub>                 | 0.2                                   |
| MgSO <sub>4</sub>                 | 0.25                                  |
| Na(PO <sub>4</sub> ) <sub>2</sub> | 0.012                                 |

**Table 1.** The salt composition of water used to house the tadpoles in the first part of the study.

Approximately 40% of the tadpoles from both species failed to thrive in these conditions (continuous loss of body condition) during the first 39 days, with no obvious reason for the poor health. Consequently, these weak tadpoles were replaced in the experimental treatments with tadpoles from the communal aquaria. On day 39, when the first sediment was introduced to the containers, the "artificial" pond water was replaced with water from an 18,000 L outdoor fish pond which had a three chambered biological filtration system. In addition, a 70-90 mm length of a water plant *Anacharis* sp. (commonly named Elodea), was added to each container to provide a shelter site for the tadpole. The containers were placed onto black cardboard to provide a dark "substrate", and they were covered with lids (containing air holes) to reduce influx of any air-borne contaminants. Even with the pond water, the pH gradually rose from 7 to 7.4 between water changes. The addition of sediment to the high sediment treatments caused a significant decline in pH (mean 7.4 to mean 7.33;  $F_{(5,10)} = 131.509$ ;  $P < 0.001$ ,  $n=5$ ) compared to the control and low sediment treatments. Higher survivorship of "high sediment" tadpoles of *H. australiacus* early in the experiment led us to suspect that this species was sensitive to water pH. Therefore, over a four week period from five to nine weeks after the first sediment influx, Wardley Bullseye™ 6.5 Acid pH regulator (actually maintained about pH 6.7) was added gradually to the water for *H. australiacus* and Seachem Neutral Regulator™ (maintained a pH ~7.0) for *M. balbus*.

### Experimental treatments

The three experimental treatments represented high (1655 mg·L<sup>-1</sup>), low (200 mg·L<sup>-1</sup>) and control (no sediment) concentrations of sediment based on published values for the Yambulla Research Catchment, in south-east NSW near the Victorian border (Olive and Reiger 1987), a forest where both species have been recorded. The sediment used in the experiment was collected from the B-Horizon of an exposed duplex soil at the site of collection of the *M. balbus* tadpoles. The sandy clay loam soil consisted of approximately 35-40% clay and fine sand

and had a pH of 5-5.5. Sediment with a low amount of organic matter was required for the experiment because high organic content can reduce the dissolved oxygen in water, killing aquatic biota (Hesse and Newcomb 1982; Garric *et al.* 1990). Organic content of the sediment ( $0.64 \pm 0.03\%$ ;  $n=3$ ) was measured using the Walkley assay (Walkley 1946). Sediment was dried in an oven at  $60^{\circ}\text{C}$ , ground to 2 mm using an EL 523-100 Rukuhia soil grinder and then finely ground using a mortar and pestle and sieved with a 0.125 mm sieve.

To simulate an influx of sediment, the tadpole was removed from its container, half the water was removed and replaced and the required volume of processed sediment was introduced into the tank and suspended by stirring. After one minute, the tadpole was returned to the container. A pilot study showed that  $65 \pm 0.8\%$  ( $n = 5$ ) of the sediment was deposited on the bottom of the containers after one minute and 85-90% of the suspended sediment had deposited within 24 hours. The final estimated sediment concentration after one minute was  $199 \pm 3 \text{ mg L}^{-1}$  and  $1675 \pm 14 \text{ mg}\cdot\text{L}^{-1}$  ( $n=10$ ) for the low and high sediment treatments respectively. Sediment influxes were repeated every three weeks to reflect the frequency of peak flow (storm) events in the Bulahdelah and Morisset districts between October 1994 and March 1997 (SFNSW, Unpublished Data). Tadpoles from all treatments were transferred to clean water after a week with sediment to simulate two weeks clear water that follows a week of sedimentation. The first sediment influx occurred on 21<sup>st</sup> May 1997 and the experiment continued for 18 weeks so that seven influxes of sediment were simulated. In the control, tadpoles were removed, half the water replaced and stirred and the tadpoles returned one minute later for monitoring.

Sixty tadpoles of each species (20 from each treatment) were measured over the 18 weeks. Survivorship of the tadpoles was recorded every 7th day with tadpoles being considered to be "dead" when they failed to grow and showed constant lethargy during the assessment period. Snout to tail length (STL) was measured every 14th and 28th day respectively (as per Semlitsch and Caldwell 1982) by placing each tadpoles into a clear plastic petri dish over a 2 mm grid sheet viewed through a dissecting microscope.

### Observation of gills

Eighteen tadpoles of each species were viewed and photographed under a Phillips 505 Scanning electron microscope (SEM). Six tadpoles from each species were selected randomly from the experimental treatments after each of the first, third and fifth sediment influxes. Thus, two tadpoles were available per treatment for each species for

this part of the study. The threatened status of these species precluded a larger sample size. Tadpoles were euthanased in a 3% chloral hydrate solution (Tyler 1976) for the 1st and 3rd sediment influxes and tricaine methane sulphonate (MS222-Sandoz; 1:6,000 w/v; McIndoe and Smith, 1984) for the 5th sediment influx. The reason for the change was that the tadpoles appeared to die in a more relaxed state in MS222. Tadpoles were preserved in Tyler's solution (Tyler 1976) until prepared for electron microscopy.

Gills were dissected from preserved tadpoles under a Leica stereo microscope MZ8 and the presence or absence of sediment in the gill chamber was recorded during each dissection. Gill arches from preserved tadpoles were prepared for the SEM using a simple cold-block procedure (Anon 1996) because specimen preparation was quick and avoided critical point drying, which may have dislodged sediment attached to the gill surfaces. Briefly, the gill arch was placed on a brass block, frozen into liquid nitrogen for approximately 45 seconds and viewed in the SEM as a standard dry specimen. Gills and samples of sediment were photographed on Ilford FP4 Plus ISO 125 film using the secondary detector in gamma mode at 10 or 20 kV or the back scattered detector at 20 kV. Presence or absence of sediment was scored from the micrographs.

### Analyses

Data are expressed as mean  $\pm$  standard error. Survivorship and the presence of sediment in gills were compared between treatments using a Chi-squared test, with the control as the expected value. Means for STL were compared using one factor analysis of variance (ANOVA) or ANOVA with repeated measures, and *a posteriori* comparisons were performed by Tukey's HSD method using the SYSTAT 5.0 statistical package. Significance was assumed if  $P < 0.05$ .

### Results

At the time of collection, *M. balbus* tadpoles had a mean STL of  $22 \pm 2$  mm and were at stage 25 (Gosner 1960). The water pH was 7.0. Tadpoles of *H. australiacus* had a mean STL of  $28 \pm 4$  mm and were also at stage 25 when collected. Water pH was 6.5.

Conditioning of the water pH half way through the experiment appeared to influence the health of tadpoles, so data are described separately for both pre and post pH control conditions. Survivorship of *M. balbus* tadpoles was not different among treatments for both pre (weeks 1-9) and post pH controlled conditions (weeks 1-9 and 10-18 respectively; Table 2). Survivorship was high throughout for *M. balbus* with only six out of a total of 60 tadpoles having died (Table 2) and no differences existed among the different treatments. In contrast, 39 out of the total of 60 *H. australiacus* tadpoles were removed from experimental

**Table 2.** The number of tadpoles removed from the experiment due to ill health or death throughout the study.

| Weeks  | <i>Mixophyes balbus</i> |          |           | <i>Heleioporus australiacus</i> |          |           |
|--------|-------------------------|----------|-----------|---------------------------------|----------|-----------|
|        | Control                 | Low Sed. | High Sed. | Control                         | Low Sed. | High Sed. |
| 1 - 9  | 2/20                    | 0/20     | 2/20      | 12/20                           | 12/20    | 7/20      |
| 9 - 18 | 0/18                    | 1/20     | 1/18      | 4/8                             | 3/8      | 1/13      |
| 1 - 18 | 2/20                    | 1/20     | 3/20      | 16/20                           | 15/20    | 8/20      |

treatments. Mortality of tadpoles in the high sediment treatment (8 of 20) was about half that in the control (16 of 20) and low sediment (15 of 20) treatments (Table 2). Mortality in the control group was significantly different from that for the high sediment treatment ( $\chi^2=20.0$ ; 1 df;  $P < 0.001$ ), but not from that of the low sediment treatment ( $\chi^2=0.31$ ; 1 df;  $P > 0.50$ ).

The same pattern was evident when survivorship before and after pH regulation are considered separately. Survivorship was greater in the high sediment treatment compared to the control group both before ( $\chi^2=5.21$ ; 1 df;  $P < 0.05$ ) and after ( $\chi^2=9.31$ ; 1 df;  $P < 0.005$ ) regulation. Survivorship in the low sediment treatment was not significantly different either before ( $\chi^2=0.00$ ; 1 df;  $P > 0.99$ ) or after ( $\chi^2=0.50$ ; 1 df;  $P > 0.25$ ) regulation.

There were no significant differences among treatments in STL (ANOVA with repeated measures  $F_{(9, 18)} = 0.833$ ,  $p=0.661$ ; Figure 1) or SVL ( $F_{(4, 8)} = 1.144$ ,  $p=0.335$ ) in *M. balbus* or in *H. australiacus* ( $F_{(9, 18)} = 4.184$ ,  $p=0.083$ ; Figure 2; and  $F_{(4, 8)} = 0.938$ ,  $p=0.491$ ).

The gill structure of both species was similar, with paired gills anterior to the intestines on either side of the heart. Individual gill filaments of *M. balbus* tadpoles were larger and more convoluted than those of *H. australiacus*, which were smooth and more bulbous in shape (Figure 3).

Sediment was observed on gills of both species with both the SEM and dissecting microscope (DM) (Table 3), with some of the sediment in large clumps resembling a mass of particles in mucus. Sediment occurred more frequently on

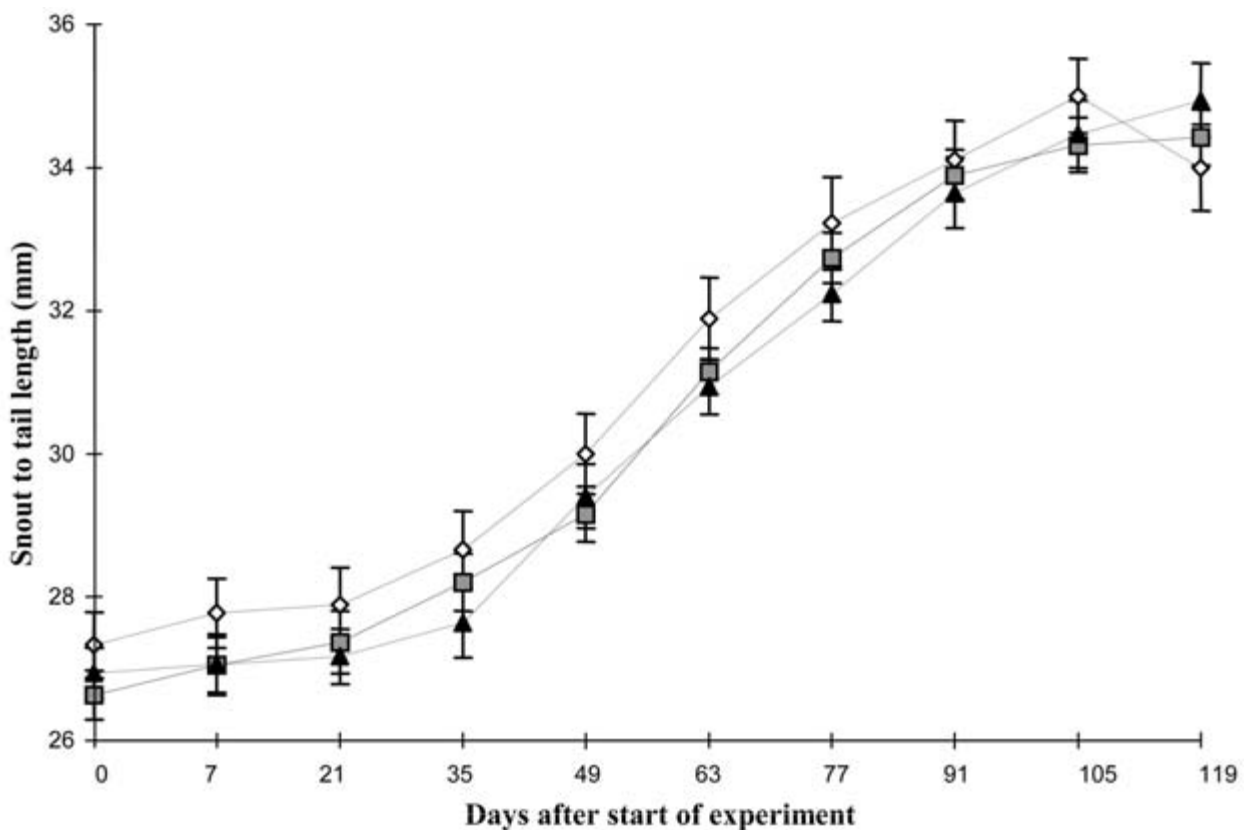
gills of *M. balbus* from the high sediment treatment than the control and low sediment treatments at all sediment influx times ( $\chi^2$  test;  $P < 0.001$ ). There was no difference in the occurrence of sediment in control and low sediment treatments.

Sediment occurred on gills of *H. australiacus* equally frequently in the high and low sediment treatments and significantly more than the control ( $\chi^2$  test;  $P < 0.001$ ). The number of times tadpoles had been subjected to elevated sediment did not cause any increase in the number of tadpoles with sediment in gill structures in either species (Table 3).

## Discussion

### Survivorship

The two species in this study showed obvious differences in survivorship and growth. Tadpoles of *Mixophyes balbus* survived well in all sediment concentrations, indicating that they are capable of surviving brief periods of exposure to high concentrations of suspended sediment. Tadpoles of *Heleioporus australiacus* differed in their survivorship among treatments, but not as initially predicted. Survivorship increased with increasing sediment concentrations. This was attributed to an indirect effect of sediment through pH changes to the water, rather than a direct effect on the tadpoles. As for *M. balbus*, *H. australiacus* tadpoles appear capable of tolerating the direct physical and physiological impacts of increased sediment influxes.



**Figure 1.** The snout to tail lengths (STL) of tadpoles of *Mixophyes balbus* subjected to three different sediment concentrations. Values are mean  $\pm$  SE. The open diamonds are the control, grey squares are the counts for the low-sediment treatments, and the black triangles are the high sediment treatments.



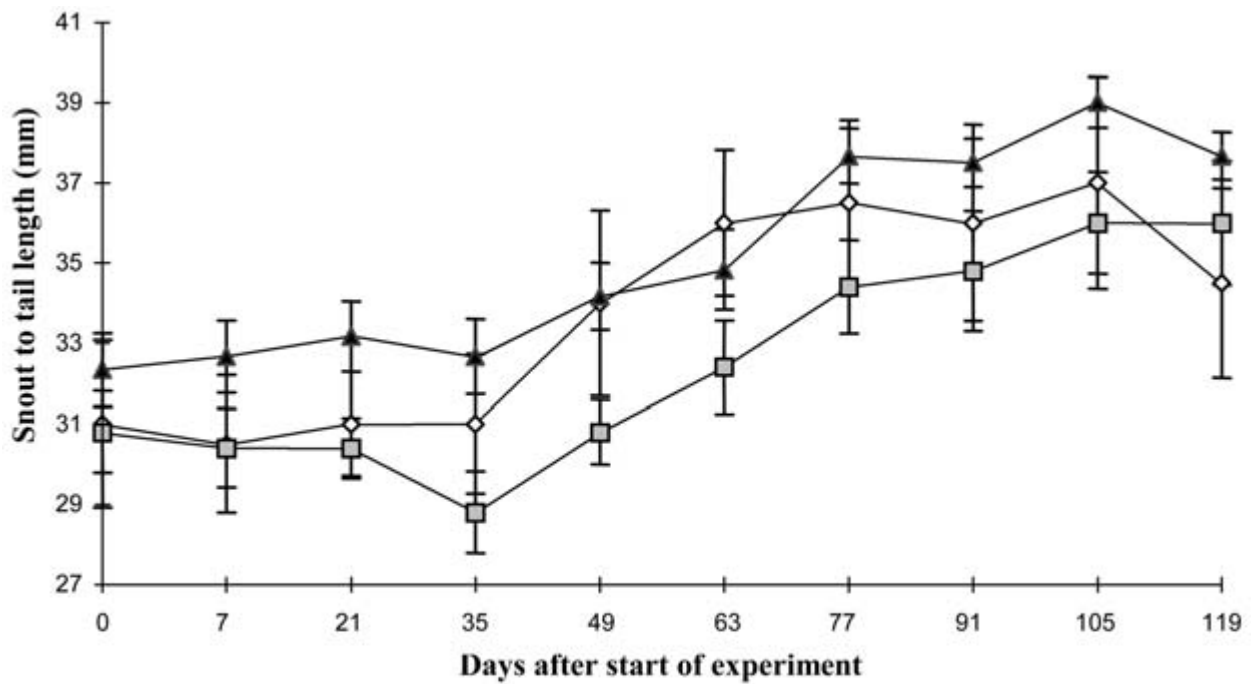


Figure 2. The snout to tail lengths (STL) of tadpoles of *Heleioporus australiacus* subjected to three different sediment concentrations. Values are mean  $\pm$  SE. For symbols, see figure 1.

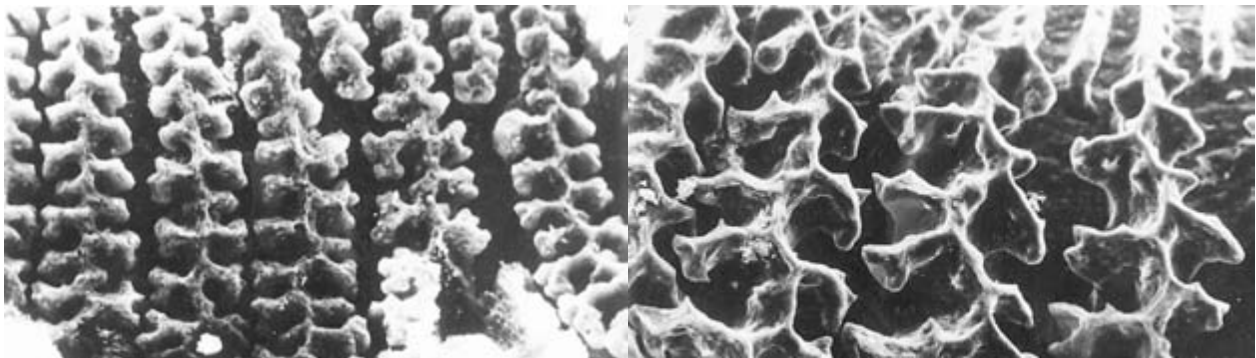


Figure 3. SEM of the gills of a) *Mixophyes balbus* and b) *Heleioporus australiacus*.

Table 3. The number of tadpoles in which particles were observed in the gills of *M. balbus* and *H. australiacus* under the dissecting microscope (DM) and the scanning electron microscope (SEM) at the time of one of three sediment influx treatment.

| Species                | Sediment Treatment | n | Control |     | Low Sediment |     | High Sediment |     |
|------------------------|--------------------|---|---------|-----|--------------|-----|---------------|-----|
|                        |                    |   | DM      | SEM | DM           | SEM | DM            | SEM |
| <i>M. balbus</i>       | 1                  | 2 | 0       | 0   | 0            | 0   | 1             | 2   |
|                        | 3                  | 2 | 1       | 1   | 0            | 0   | 2             | 0   |
|                        | 5                  | 2 | 0       | 0   | 1            | 1   | 2             | 2   |
|                        | Total              | 6 | 1       | 1   | 1            | 1   | 5             | 4   |
| <i>H. australiacus</i> | 1                  | 2 | 0       | 0   | 2            | 2   | 0             | 2   |
|                        | 3                  | 2 | 2       | 1   | 1            | 2   | 1             | 2   |
|                        | 5                  | 2 | 0       | 0   | 2            | 2   | 2             | 2   |
|                        | Total              | 6 | 2       | 1   | 5            | 6   | 3             | 6   |

The apparent positive influence of pH on *H. australiacus* tadpoles was not predicted, but it is of particular interest. The apparent insensitivity *M. balbus* to the range of water pH found in the laboratory relates well to the likely natural variation in pH in the variety of environments in which they occur (Barker *et al.* 1995; Lemckert and Morse

1999). In contrast, tadpoles of *H. australiacus* showed high sensitivity to water pH. *Heleioporus australiacus* is a specialist of the Sydney Sandstone formations, the source of individuals used in this study (Robinson 1993). The soils derived from this rock formation are generally acidic and so are the adjacent waters. Hence, *H. australiacus*

have tadpoles that are selected to cope with relatively low and constant pH and be disadvantaged by more neutral pHs. The laboratory set-up originally did not consider changes to water pH over time, which was an artefact of the experimental treatments. The pH of water in the field was 6.5. Our regulation of pH in the laboratory treatments to 6.7 was sufficient to greatly improve survival of the tadpoles. More field measurements and laboratory experiments are required to confirm that this species is dependent on lower pH waters.

The unexpected influence of pH on the tadpoles revealed a potentially important phenomenon. Tadpoles of *M. balbus* are apparently less sensitive to changes in water pH than tadpoles of *H. australiacus*. Even small changes in pH may have a detrimental affect on *H. australiacus*. Susceptibility to changes in water chemistry, such as pH, is common in other aquatic biota (Chutter 1969, Bruton 1985, Quinn *et al.* 1992, Newcombe and Jensen 1996) and may result in changes to species composition of animals in freshwater habitats. Changes in stream pH due to forestry activities have not been recorded and it is possible that the size of “natural” water bodies (compared to our laboratory experiments) buffers any potential pH changes (P. Cornish, State Forests of NSW, pers. comm.).

Tadpoles of both species survived exposure to concentrations of suspended sediment typical of a heavily logged catchment during peak flow. Changes in water chemistry brought about by the suspended sediment in the water (Bruton 1985, Beattie and Tyler-Jones 1992, Ryan 1991, Erskine 1996) may have an adverse impact in sensitive species, such as *H. australiacus*. The continued high mortality in the control and low sediment treatment, compared to the high sediment treatment after pH was regulated in the experiment, identifies that early exposure to high pH may have long term consequences on surviving tadpoles. An alternative explanation is that the pH change was not the only factor that caused the mortality of the *H. australiacus* tadpoles. Detailed investigation of the natural pH of waters inhabited by tadpoles of *H. australiacus*, together with laboratory studies of the influence of pH on this species, are required to fully explain our results.

## Growth

Growth was not affected significantly by the sediment treatments for either species. This study was designed to simulate natural concentrations, duration and timing of sediment influxes, although we could not simulate flow that keeps sediment suspended longer in the field (Doeg and Koehn 1990) than in our experiment. Increased concentrations of suspended sediment may affect growth by reducing food availability or feeding efficiency of aquatic biota through smothering and reduced visibility (Bruton 1985, Gillespie and Hines, 1999). Suspended sediment will reduce light penetration in streams resulting in reduced photosynthesis and consequent total biomass. Such effects were absent in the experiment, where there was a rapid deposition of sediment and the regular addition of food. The impact of deposition of sediment on photosynthesis and production in habitats of *M. balbus* and *H. australiacus* requires further investigation in the field.

## Gills

Gas exchange occurs across the skin, gills and, in most cases, the lungs of aquatic tadpoles (Burggren and West 1982, Boutilier *et al.* 1992). As tadpoles grow, gills and then lungs become more important (Burggren 1984). Tadpoles of both *M. balbus* and *H. australiacus* have well developed gills, suggesting that they depend on gills to exchange oxygen, and possibly carbon dioxide, with the water to meet their minimum respiratory requirements. Thus, a decrease in the efficiency of gills of stream-dwelling species like *M. balbus* or *H. australiacus* may have an adverse impact on their respiration, which would reduce their activity and their ability to feed.

More sediment attached to the gills of tadpoles in the high sediment treatments than the controls for both *M. balbus* and *H. australiacus*, with a higher incidence of sediment in the gills of *H. australiacus* from the low sediment treatments than *M. balbus*. Thus, tadpoles of *H. australiacus* are more susceptible to contamination of their gills by sediment than are *M. balbus* tadpoles, probably because the smooth branched gill filaments of *H. australiacus* collect sediment more easily than the shorter and wider gill filaments of *M. balbus*.

We could not quantify the extent of clogging of individual gills. Most sediment particles were sparsely distributed on gills, or in the form of large clumps of sediment. These clumps, that resembled mucus with sediment particles trapped in it, occurred within the cup-like gill chambers. Mucus probably is secreted from gill filaments in an attempt to clear sediment particles from the gills. Elimination of foreign particles from gills with mucus is common in many species of fish (Dietrich and Schlatter 1989, Erard Le Denn and Ryckaert 1990, Martens and Servizi 1993), although we are unaware that it has been described in tadpoles. The presence of sediment trapped in mucus suggests that these species can clear the gills of sediment. The exposure in our experiment did not exceed their capacity to clear the gills. The lack of any effect of sediment exposure on the growth of either species also suggests that sediment in the gills did not occlude gas exchange to the extent of reducing growth. Finally, the incidence of sediment particles on gill structures did not increase with the increased number of times (1, 3 and 5) that tadpoles were exposed to sediment influxes, suggesting that the gills are readily cleared.

## Conclusions

This study has provided the first, albeit preliminary, analysis of the influence of suspended sediment on two species of threatened stream-dwelling tadpoles in NSW. Short-term sediment influxes into water bodies do not directly threaten tadpoles from these two stream breeding species. If a threat exists from sediment suspension, it is most likely to occur where sediments are regularly re-suspended over an extended period of time. “Splash” (unbridged) crossings of streams, such as are often used by logging trucks (as well as by other vehicles), may lead to high siltation rates when vehicular traffic is frequent. In a logging operation that lasts several weeks, a crossing will be used up to a dozen times a day (C. Slatyer, Australian Heritage Commission pers. comm.), which may result

in more continuously-suspended sediment and in turn lead to a greater impact on the respiration or feeding of tadpoles. Frequently-used public roads may represent an even more regular source of sediment "stirring". Roads form the most important source of sediment introduction into streams in forests (O'Shaughnessy *et al.* 1995, Cornish 2001), suggesting again that "splash" crossings may be an important future research area.

Erosion control guidelines, at least in New South Wales, have become far more stringent for all activities than were available in the forestry studies reviewed by Doeg and Koehn (1990) and may result in less erosive material entering streams than naturally enter streams in "old growth" forests

(Cornish 2001). These guidelines increase the protection required around streams and other water bodies where slopes are steeper and/or the soils more prone to erosion events and so should, in general, minimise any possible increases in suspended sediment. Any short-term and direct threats to tadpoles from increased suspended sediment that might once have been present should today be less. The experimental approach in this study has identified its value for work on amphibians and we encourage others to pursue this line of investigation. There are many other species and conditions, such as fire and drought followed by floods, that are amenable to the experimental approach with this important group of forest dwellers.

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