Cape Leeuwin Freshwater Snail Monitoring Report

2009

Department of Environment and Conservation
South West Region

Prepared by Kimberly Onton, December 2009

Department of Environment and Conservation

CARING FOR OUR COUNTRY

SOUTH WEST CATCHMENTS COUNCIL

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Cover images: DEC staff taking water quality measurements at Canal Rocks; Cape Leeuwin Freshwater Snail observed under light microscope; DEC staff sorting through samples at Canal Rocks; Cape Leeuwin Freshwater Snail; DEC staff sorting through samples at Ellensbrook. Photos: Kim Onton, Chris Fleay and Jen Harrison (DEC).

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1 INTRODUCTION

The Cape Leeuwin Freshwater Snail *Austroassiminea letha* is currently listed on Schedule 1 (Division 6) of the Wildlife Conservation (Specially Protected Fauna) Notice 2006 as fauna that is rare or likely to become extinct, and is listed as Vulnerable on the IUCN Red List of Threatened Species (State of Western Australia, 2006; Clark and Spier, 2005; IUCN, 2009). The species is known from six sites between Cape Leeuwin in the south to Cape Naturaliste in the north, though according to the fossil record was once more widely spread throughout the Leeuwin-Naturaliste Ridge (Solem *et al*, 1982; Burbidge, 2004). It inhabits seepage films or splash zones alongside small freshwater streams and springs draining from limestone near the coast (Burbidge, 2004). The species grows up to 5.4 mm and is considered a short-range endemic as it is confined to small areas of discontinuous habitat and has poor powers of dispersal (Solem *et al*, 1982; Harvey, 2002).

Given the species' apparent requirement for freshwater, any activity that affects the amount or quality of water in streams and springs could detrimentally impact this highly restricted species. An understanding of the species' habitat requirements relating to hydrology, vegetation and soil substrate is required to better manage its habitat and threats to the species. In addition, a detailed study of the density of the snail populations at each site, and surveys of other potential sites along the south-west coast, are required to understand the current population parameters in order to detect changes over time. An understanding of the genetic diversity between occurrences is also important in determining whether distinct management units are required as the species' fragmented distribution and poor powers of dispersal can lead to evolutionary divergence. With increasing pressure along the south-west coast from development, plus the predicted effects of climate change, monitoring of the species and its habitat is critical in not only conserving the species but the habitat that it represents. This baseline information is critical to detect change over time and mitigate potential threats.

In 2007, the South West Catchments Council (SWCC) provided funding to the Department of Environment and Conservation's South West Region to: implement a monitoring program for the Cape Leeuwin Freshwater Snail, determine the threats to the species and carry out any required recovery actions. This project was extended from January to September 2009 to allow for additional sites to be surveyed and further genetic analysis to be undertaken. This report outlines the project objectives, methods and results to date and provides recommendations for future monitoring and management of the species and its habitat.

2 OBJECTIVES

1. Determine the abundance of the Cape Leeuwin Freshwater Snail at known occurrence sites.
2. Map the extent of Cape Leeuwin Freshwater Snail habitat at each occurrence.
3. Determine the microhabitat preferences of the Cape Leeuwin Freshwater Snail.
4. Investigate the presence of the Cape Leeuwin Freshwater Snail at additional sites along the south-west coast.
5. Determine the level of genetic isolation of each Cape Leeuwin Freshwater Snail occurrence and determine the need for taxonomic revision or management as distinct units.
6. Identify and quantify potential threats to the Cape Leeuwin Freshwater Snail at each occurrence.

3 METHODS

3.1 Study Locations

The Cape Leeuwin Freshwater Snail is known from six sites, for which four have had monitoring data collected (as one occurrence is on private land where access was not been permitted and the other was only discovered in the late stages of the project). The four monitoring sites are Canal Rocks, Ellensbrook, Cosy Corner and Cape Leeuwin (Fig 1).

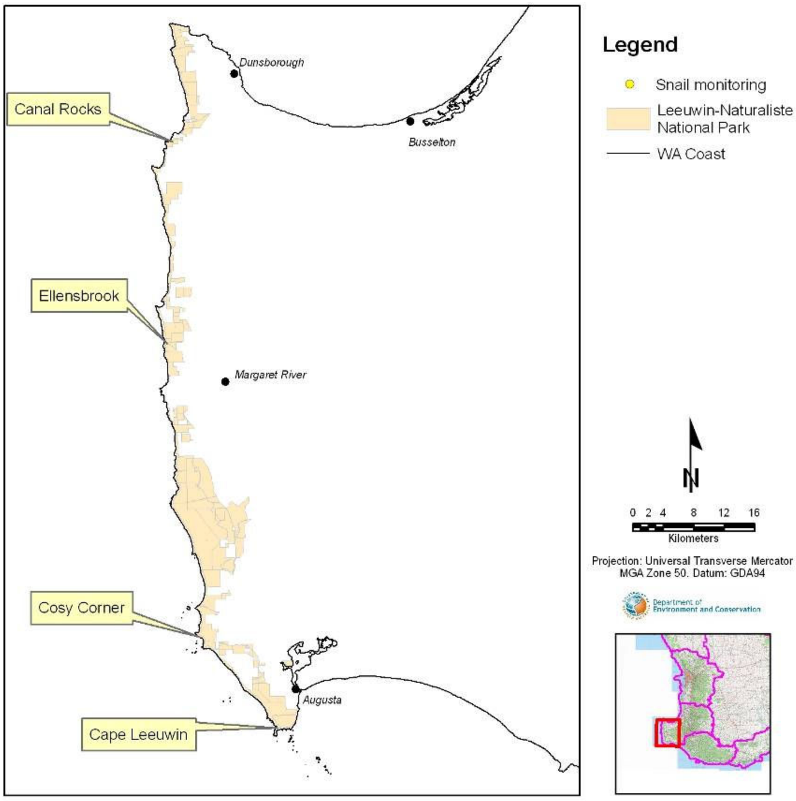


Figure 1: Map of south-west Western Australia showing Cape Leeuwin Freshwater Snail occurrences.

3.2 Gastropod abundance

Monitoring of the abundance of *A. letha* and was undertaken at each site. This involved establishing permanent monitoring stations in a stratified random manner across the sites based on habitat type and spatial considerations. A marker was installed and a GPS location was recorded for each station. Within the immediate vicinity of each station two 30cm x 30cm quadrats were sampled for gastropods. A scraping of soil and leaf litter to a depth of 1-2cm was collected from the quadrat and placed in a three-tiered sieve (consisting of 4mm, 2mm and 1mm sieves plus a collecting tray). Water from adjacent to the station was then poured into the sieve column and agitated to separate out the different sized specimens (Fig 2). Each sieve was then searched by eye and any gastropod species were extracted with forceps (Fig 3). The number of each species (dead and alive) was counted and recorded per sieve size (Attachment 1). Once counted, all sieved material and specimens were returned to the site, unless specimens were retained as vouchers for identification purposes. Vouchered specimens were sent to the Western Australian Museum for identification.

Note: Sampling stations at Cape Leeuwin were established in 2003 and resampled in 2005 by S. Slack-Smith (WA Museum) through a monitoring program for the Water Corporation. As monitoring data exists for this site, the DEC 2007-2009 monitoring program focused on the Canal Rocks, Ellensbrook and Cosy Corner occurrence sites only.

Figure 2: Images illustrating the site markers and the sieving process for Cape Leeuwin Freshwater Snail sample collection.

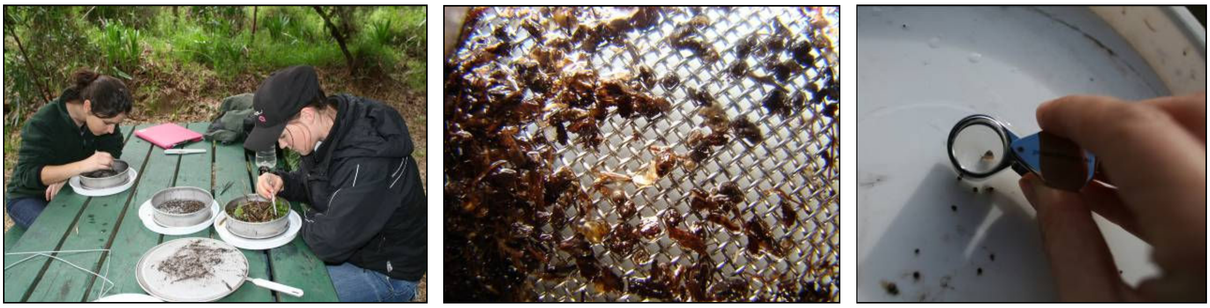


Figure 3: Images illustrating sieve sorting and specimen extraction for identification and counting.

3.3 Microhabitat

A temperature reading was taken with a digital thermometer at the soil surface of each quadrat. A photograph was taken of each station and a habitat description recorded, including vegetation species, soil characteristics, distance to surface water, storey type and cover (Fig 4; Attachment 2). Water quality monitoring was also undertaken at sites which held surface water. This involved using hand-held meters and probes to measure pH, electrical conductivity and dissolved oxygen. A water quality monitoring logger was also installed at the Ellensbrook site collecting ongoing conductivity, temperature and depth data. A floristic survey was also undertaken at monitoring sites by a DEC botanist.

Figure 4: Images of monitoring stations and sampling quadrats.

3.4 Habitat Extent

The extent of habitat use by *A. letha* at each site was mapped. This involved searching through soil throughout the site to determine the presence/absence of the species. An approximate boundary was delineated using a GPS and both minimum and buffered occurrence boundary maps were created using GIS.

3.5 Survey for New Occurrences

Based on an understanding of the basic habitat requirements of the Cape Leeuwin Freshwater Snail, several surveys for new occurrences were undertaken. This involved selecting sites with freshwater springs flowing from limestone near the coast. Visual assessment was undertaken along banks, in vegetation and in moist rock fissures.

3.6 Data Management

All abundance and microhabitat data collected was entered onto data sheets stored at the DEC South West Regional office (Attachments 1 & 2). All data was also entered into the Cape Leeuwin Snail Microsoft Access database located on the DEC South West Region server. All photographs taken are digital and are stored on the DEC South West Region server. Reports and shapefiles are located on the DEC South West Region server and are sent to the DEC Species and Communities Branch.

3.7 Genetic Analysis

An analysis of the molecular systematics of the Cape Leeuwin Freshwater Snail was undertaken in 2008. Up to 15 *A. letha* specimens were collected from each site and were preserved in 100% ethanol-filled vials. The specimens were sent to Helix Molecular Solutions for analysis. Specimens were insufficient for a sound analysis (DNA was difficult to extract in most specimens), therefore a further 65 live specimens were collected and transported to the laboratory where further DNA extractions were made by Helix Molecular Solutions staff. DNA sequences were used to determine genetic (haplotype) diversity[1], effective population size[2] and the evolutionary relationships among the isolated snail occurrences. A specific report outlining genetic analysis methods and results was prepared by Oliver Berry and Terrie Finston of Helix Molecular Solutions (2009), and an extract of results is provided in this report. DNA sequences were deposited on Genbank.

3.8 Threat Analysis

Potential threats to *A. letha* were identified and quantified at each site.

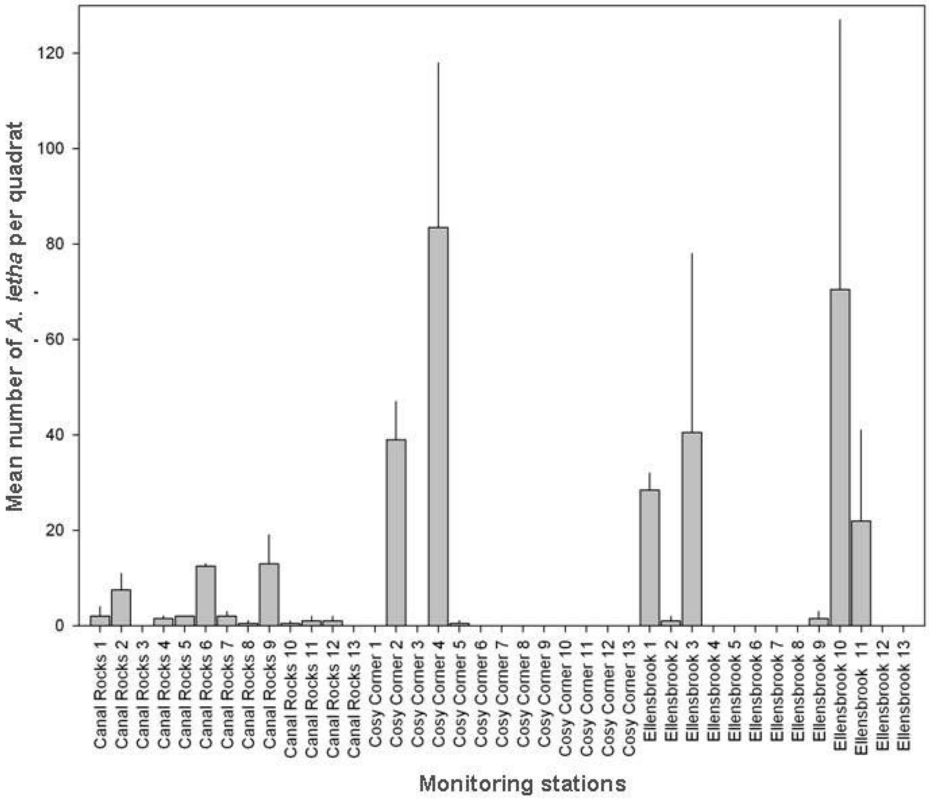
[1] Haplotype diversity is a measure of the number and frequency of haplotypes within a population.
[2] Effective population size represents a long-term average size that a population with idealised demographic parameters (including equal reproductive contribution per generation and equal sex ratios) would need to be to retain the observed genetic diversity (Avise 1994).

4 RESULTS

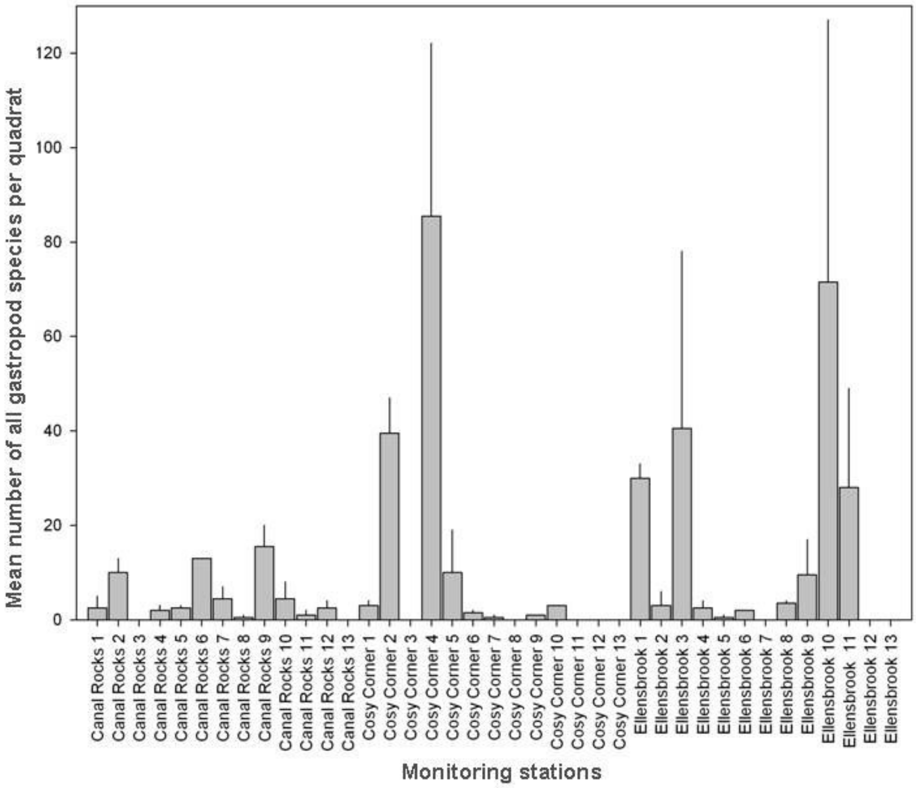
Note: Results presented on A. letha, Oxychilus and total gastropod abundances refer to live snails unless stated otherwise.

4.1 Gastropod abundance

Analyses of variance (ANOVA) revealed significant differences in both the abundance of *A. letha* (P = 0.013) and total abundance of gastropods (P = 0.019) across all individual monitoring stations, however no significant difference was observed between the sites as a whole (*A. letha* abundance P = 0.350; total abundance P = 0.270) (Fig 5). Ellensbrook and Cosy Corner had greater *A. letha* abundance than Cosy Corner, however the high variance likely reduced the statistical power suggesting that greater sampling replication may have been required (Fig 5a).



a)



b)

Figure 5: Mean abundance of a) *A. letha* and b) all gastropod species across monitoring stations for three snail monitoring sites (Canal Rocks, Cosy Corner and Ellensbrook).

The predatory snail *Oxychilus* was present at all stations within the Ellensbrook site with a significant variation in the abundance of *Oxychilus* across stations (P = 0.014). With the exception of station 11, *Oxychilus* were most abundant at sites where *A. letha* were either absent or present at low densities (Fig 6). This may suggest different habitat requirements for each species or the detrimental impacts of predation by *Oxychilus* on *A. letha*. However sites with a high abundance of predated *A. letha* shells did not have particularly high *Oxychilus* densities (Fig 6).

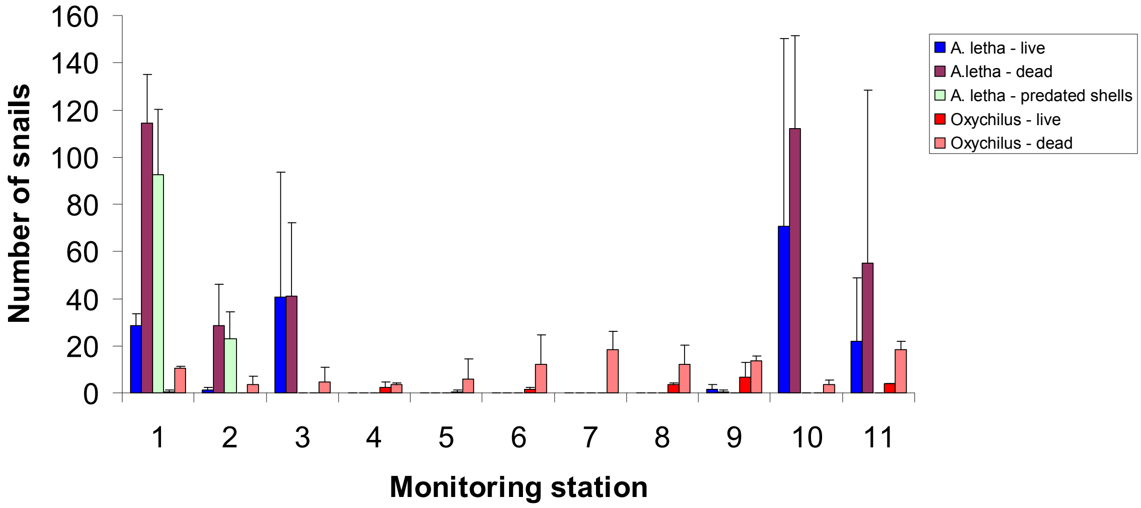


Figure 6: Mean abundance of *Oxychilus* species across Ellensbrook monitoring stations.

4.2 Microhabitat

Of the environmental habitat parameters measured, distance to a surface water source was the only parameter that showed a significant correlation through a log transformed regression analysis (R^2 = 29.1, P = 0.002) (Fig 7). *A. letha* were most commonly found less than one metre from a surface water source. Observations noted that *A. letha* were not commonly found on banks adjacent to brooks with rapid water flow but were abundant adjacent to freshwater streams and springs. This suggests that close proximity to a slow-flowing spring water source is a likely habitat requirement for the species. Nested ANOVA and regression analyses tested the relationships between *A. letha* abundance and environmental parameters including water quality, soil texture and moisture and vegetation cover (over and understorey) and species composition and revealed no significant relationships (R^2 = 0 - 6.3%, P = 0.088 – 0.922).

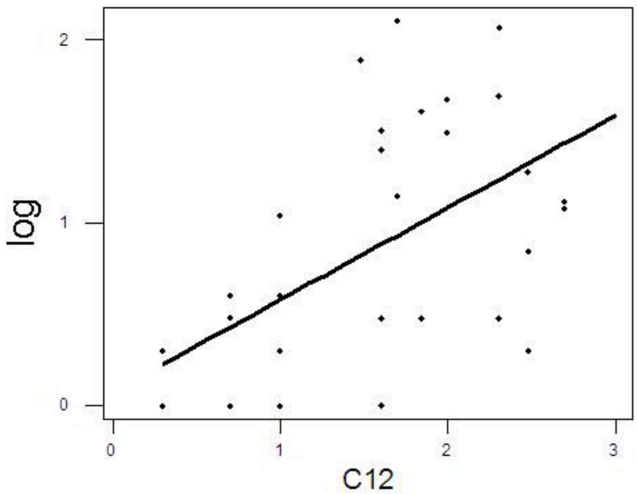


Figure 7: Regression correlation (log transformed) between abundance of *A. letha* and distance to a surface water source.

Cape Leeuwin Freshwater Snail sites varied in vegetation species composition (Table 1). The Canal Rocks site was dominated by kikuyu (*Pennisetum clandestinum)* and buffalo grass (*Stenotaphrum secundatum*) forming a very dense understorey with little or no overstorey. The grasses often grew over shallow running water and appeared to stabilise soil in areas where it would otherwise be washed away. Ellensbrook monitoring sites had considerably dense overstorey cover dominated by peppermint trees (*Agonis flexuosa*). Introduced species, such as arum lily (*Zantedeschia aethiopica)*, comprised much of the understorey with some sedge cover provided by spreading swordsedge (*Lepidosperma effusum*). The Cosy Corner site was dominated by open heath on the elevated areas of exposed granite where soil was generally shallow with no overstorey cover. The lower areas had greater overstorey cover provided by peppermint trees and significant shading from the tall cliff face. Coastal sword sedge (*Lepidosperma gladiatum*) was a dominant understorey species in the lower-lying areas.

Table 1: Dominant plant communities at Cape Leeuwin Freshwater Snail monitoring sites, May 2008. (Vegetation survey: Andrew Webb)

<u>**CANAL ROCKS**</u> Closed grassland with some low heath dominated by *Pennisetum clandestinum**, *Stenotaphrum secundatum**, *Centella asiatica*, *Rhagodia baccata* and *Apium prostratum* with a greatly reduced and declining sedge layer of *Lepidosperma gladiatum* and *Juncus krausii*. Fringed by *Melaleuca lanceolata*, *Spyridium globulosum* and *Acacia cochlearis* closed tall scrub.
<u>**ELLENSBROOK**</u> **EB01** *Hakea oleifolia, Leucopogon parviflorus, Rhadinothamnus anceps, Acacia saligna, Billardiera heterophylla* open heath, over *Juncus kraussii, Baumea preissii, B.juncea, Lepidosperma effusum* sedges and *Romulea rosea*, Centella asiatica, Plantago lanceolata*, Paspalum dilatatum** herbs and grasses. **EB09** *Agonis flexuosa* low closed forest, over *Trymalium floribundum, Callistachys lanceolata, Boronia alata* open heath, over *Rhagodia baccata, Hibbertia cuneiformis* low shrubland, over mixed herbs and sedges including *Zantedeschia aethiopica*, Hypocharis glabra*, Cyperus tenuiflorus*, Paspalum dilatatum*, Microlaena stipoides, Geranium solanderi, Sonchus oleraceus*, Lotus sp.** and *Dichondra repens*. **EB10 & 11** *Agonis flexuosa* low closed forest, over *Rhagodia baccata, Pteridium esculentum, Leucopogon australis, Spyridium globulosum* very open shrubland, over *Dichondra repens* and annual weeds including annual grasses, *Cerastium glomeratum** and *Isolepis marginata*.* **EB12** *Agonis flexuosa* low closed forest, over *Spyridium globulosum* shrubland, over very dense grasses *Pennisetum clandestinum*, Stenotaphrum secundatum** with the sedges of *Baumea juncea* and *Lepidosperma effusum*.
<u>**COSY CORNER**</u> **CC01, 03, 04, 05, 06, 09, 10** Exposed granite rocks with an open low heath in areas of soil dominated by *Pimelea ferruginea, Thryptomene saxicola, Eutaxia obovata* and *Acanthocarpos preissii* over the sedge and grass species of *Schoenus nitens* and *Poa poiformis*. The small creekline at this site is dominated by species including *Agonis flexuosa, Leucopogon parviflorus, Melaleuca huegelli, Boronia alata* and *Acacia littorea*. **CC02, 08 & 07** *Agonis flexuosa* low closed forest, over *Rhagodia baccata, Olearia axillaris, Spyridium globulosum, Clematis pubescens* open low heath, over *Lepidosperma gladiatum, Ehrharta erecta*, Poa poiformis* sedge and grasses.

*Denotes introduced species.

4.3 Habitat Extent

Habitat maps were created for each of the monitoring sites incorporating a minimum and buffered occurrence boundary (or polygon). Due to the sensitive nature of this threatened species locality information, the maps are not provided in this report, however can be requested from the DEC South West Region.

4.4 New Occurrences

In 2007, a new occurrence of *A. letha* was discovered at Canal Rocks extending the species range north by approximately 30km. In September 2009, a site highlighted by Hayley Rolfe of the Cape to Cape Catchments Group as a potential tufa site was inspected. The site near Boodjidup Creek was found to be inhabited by *A. letha*. This site is within the known species range and extends the number of sites known to be inhabited by the species to six. A site at Contos Spring was also inspected for the species but was found to be unsuitable and no *A. letha* were observed.

4.5 Genetic Analysis

Genetic analysis provided evidence for both historic and recent gene flow between the four *A. letha* populations examined and do not support the need for taxonomic revision of the species (Berry and Finston 2009). Mitochondrial DNA sequences were obtained from 31 of the 83 specimens and 21 unique DNA sequences (haplotypes) were recovered. Ellensbrook was the only site with sufficient sampling effort to determine an effective population size of 1954. This is surprisingly high given the apparently small and relictual nature of the Ellen Brook population.

One haplotype was shared between sites (Cosy Corner and Leeuwin Swamp), but in all other cases haplotypes were unique to each site. No well-supported monophyletic lineages corresponding to individual sites or groups of sites were recovered (Fig 8). The level of genetic differentiation between snails from different sites was generally higher than that between snails within sites. However, there was extensive overlap in the levels of differentiation within and between sites (Fig 9).

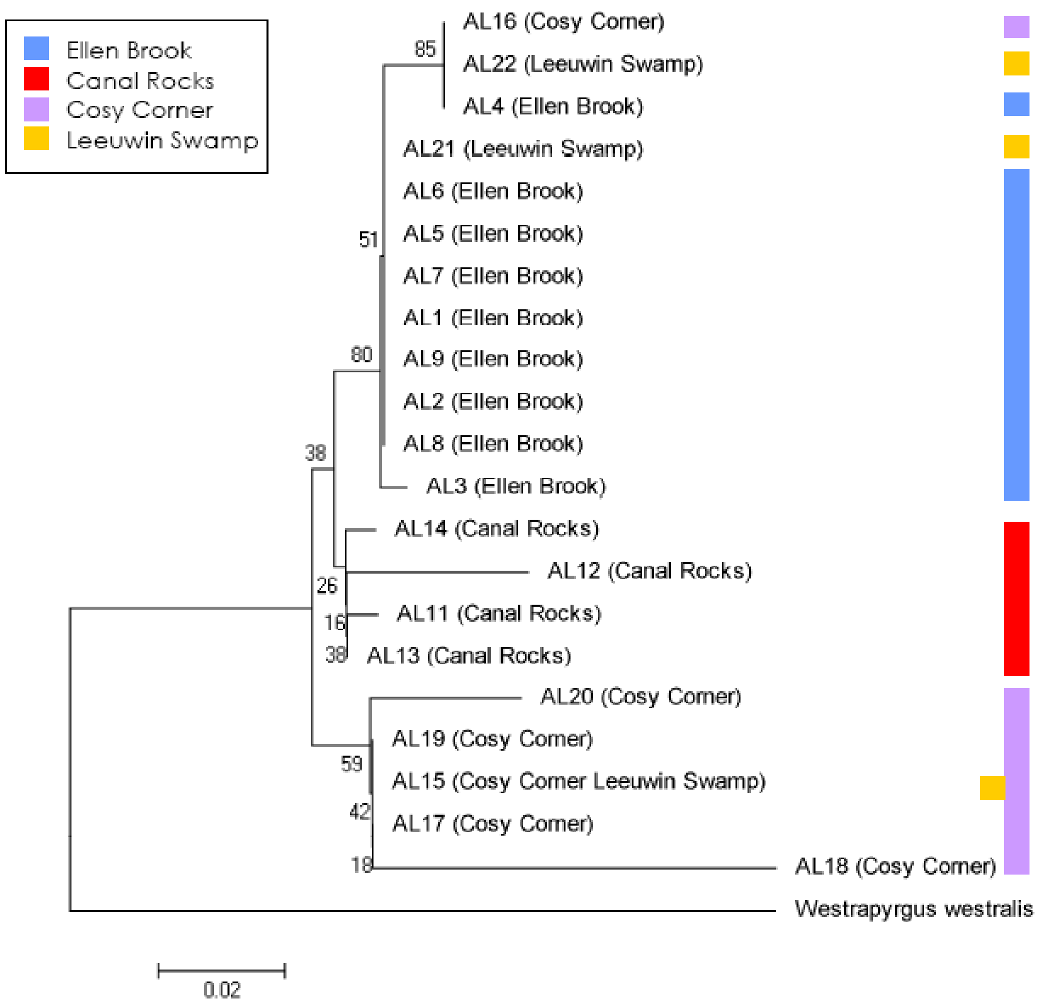


Figure 8: Phylogenetic tree summarising the evolutionary relationships among mitochondrial haplotypes recovered from *Austroassiminea letha* specimens. The evolutionary history was inferred using the Neighbour-Joining method and uncorrected nucleotide distances. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (500 replicates) is shown next to the branches. The tree is drawn to scale, with horizontal branch lengths representing the genetic distance (divided by 100) between each haplotype. Note that in most cases snails from each site group together but the bootstrap support for these groupings is weak.

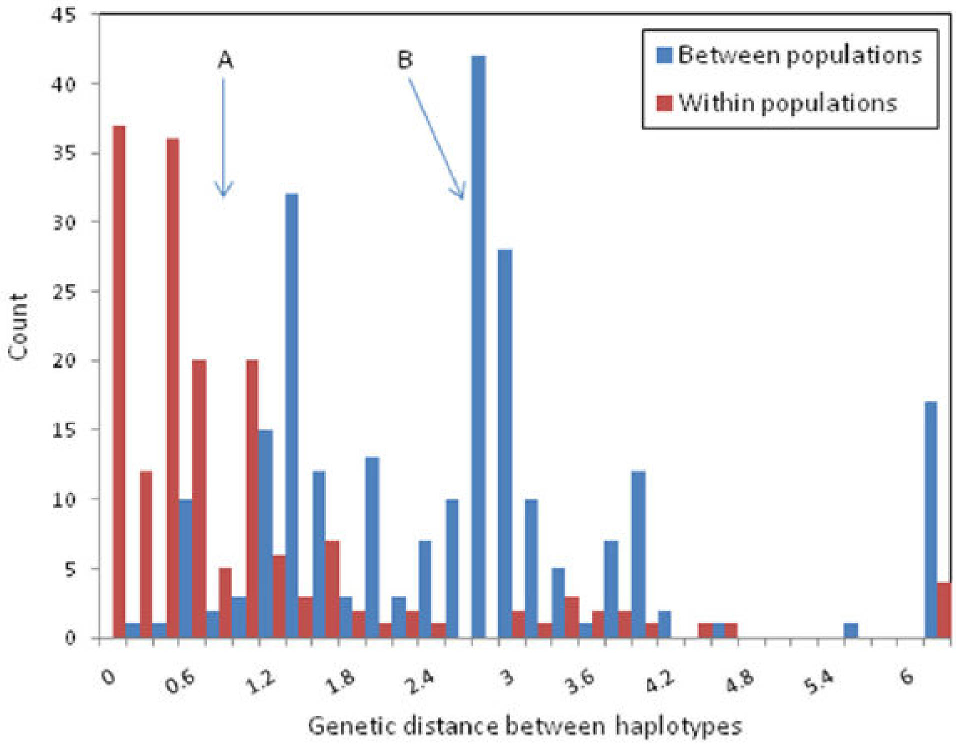


Figure 9: Histogram of corrected genetic distances between unique DNA sequences (haplotypes) obtained from *Austroassiminea letha* specimens. A and B indicate the average within and between population genetic distance respectively.

The lack of phylogenetic signal combined with the sharing of closely related haplotypes between populations indicates that *A. letha* has historically and recently exchanged migrants. The divergent haplotypes recovered within sites are indicative of historically larger effective population sizes, which support geological evidence of a more widespread distribution in the past (Solem et al 1982).

4.6 Threat Analysis

Potential threats at each site were identified and ranked according to their likelihood of occurrence and significance of impact (Table 2). The greatest threats were identified as any reduction in water availability, either through extraction or diversion and reduced rainfall as a consequence of climatic changes. The presence of the freshwater seeps and springs appears to be a critical attribute to suitable habitat for *A. letha*. Several *A. letha* sites are also heavily infested with weeds, such as kikuyu grass, arum lilies and figs. Often these weeds appeared to be providing suitable habitat for *A. letha*, however weed management needs to be considered for greater biodiversity conservation. If weed control is to be undertaken at *A. letha* sites, a staged approach with minimal chemical application is recommended to avoid detrimental impacts to *A. letha* populations though direct disturbance (trampling, loss of suitable habitat due to weed extraction and loss of vegetation cover) or toxicity through application of chemicals. A guidance document has been prepared for the DEC Blackwood District identifying considerations when undertaking disturbance activities, such as weed control, at Cape Leeuwin Freshwater Snail sites.

Table 2: Ranking of potential threats to Cape Leeuwin Freshwater Snail occurrences.

THREATS	SITES				
	Canal Rocks	Ellensbrook	Boodjidup Creek	Cosy Corner	Cape Leeuwin
Reduced water availability (reduced rainfall, extraction)	Moderate	Moderate		High Very small volumes of surface water available at this site. Snails keep moist in rock fissures.	High A decline in surface water and an increase in terrestrial vegetation encroaching on the swamp has been observed (S.Slack-Smith, pers. comm.)
Water quality decline	Moderate High viticulture pressure within catchment	High Higher nutrient levels recorded at this site.	Moderate High viticulture pressure within catchment	Low	Low
Fire	Moderate	Moderate	Moderate	Low	High
Development	Low All occurrences are within Leeuwin-Naturaliste National Park				
Predation	Low	High Predatory snail *Oxychilus* observed at this site.	High Predatory snail *Oxychilus* observed at this site.	Low	Low
Disturbance through weed control	High Site has previously been sprayed with herbicide to control kikuyu grass across track to spring	High Local conservation volunteers/National Trust/DEC wish to undertake weed control at this site. Requires strategic approach so as not to introduce chemicals and create unsuitable habitat	Moderate This site has been targetted by local conservation volunteers who wish to undertake weed control	Low	Low
Climate change	High	High	High	High	High

5 CONCLUSION & RECOMMENDATIONS

The Cape Leeuwin Freshwater Snail's distinctly fragmented, isolated populations suggest very specific and restricted habitat requirements. Freshwater seeps and springs derived from limestone near the coast are features of all known *A. letha* occurrences and this criteria guided the successful discovery of two new populations including a range extension. It is possible that a number of other sites along the coast are also inhabited by *A. letha* which should be further explored and considered when management decisions are made at freshwater spring sites. This study found weak correlations between other environmental factors and *A. letha* abundance, possibly due to the scale of the parameters measured. The presence of a freshwater flow or seepage remains the only indicator to suggest the potential presence of the species. A finer scale measurement of snail habitat including quantitative soil moisture and organic content and sediment size is suggested to better understand *A. letha* habitat requirements, as the soil provides both habitat and a food source (as *A. letha* is detritivorous). This monitoring program has facilitated the collection of baseline information on snail density and habitat parameters required for monitoring change over time and managing threats to this endangered, short-range endemic species.

Although evolutionary divergence between the four sampling sites was modest and not sufficient to warrant taxonomic revision, populations or groups of populations below the level of species can still warrant independent conservation attention if they show evidence of a long history of evolutionary independence and ecological differences. Although gene flow is restricted between *A. letha* populations, it has occurred in the deeper past - most likely when the distribution was more continuous (poor phylogenetic resolution, approximately during the past hundreds of thousands of years to early Pleistoncene), as well as the recent past (similar haplotypes shared between populations; approximately the past tens of thousands of years) The genetic data does not provide evidence that each site represents a separate evolutionary significant unit .

The following are recommendations regarding ongoing *A. letha* monitoring and management.

- Undertake baseline monitoring of Cape Leeuwin Freshwater Snail abundance and habitat at Boodjidup Creek and Deepdene occurrences which were unable to be fully surveyed through this project.
- Continue monitoring of the Cape Leeuwin Freshwater Snail at known sites to monitor population trends, noting any significant changes to habitat including vegetation and water quality and volume. To maximise efficiency of monitoring, subset samples focusing on *A. letha* and *Oxychilus* only could be collected. This might entail only counting specimens in one or two sieve sizes. Quantitative measurements of soil moisture, organic content and sediment size should also be collected to complement the snail abundance data.
- Continue liaison with organisations interested in weed management at Cape Leeuwin Freshwater Snail sites to ensure weed eradication techniques have a minimal impact. A document was prepared for the DEC Blackwood District in September 2009 outlining recommendations and considerations for managing disturbance activities such as weed removal at Cape Leeuwin Freshwater Snail sites.
- Consider the potential presence of the Cape Leeuwin Freshwater Snail when making management decisions affecting confirmed and potential habitat for this species, such as sites incorporating freshwater springs and seepages.

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Attachment 1: Cape Leeuwin Freshwater Snail density monitoring data sheet.

Date: Site Name: Time start: Time finish: Page of

Recorder: Other participants: Air Temperature at start: °C Air Temperature at Finish: °C

Station	Snail Species	Sieve Size (mm)	Count		Substrate (i.e. Veg, soil, water, rock)	Associated Vegetation Spp.	Distance to water (cm)	Temperature at ground level (°C)	Specimen Collected		Comments
Quadrat			Alive	Dead					✓ ×	#	

Notes:

Attachment 2: Monitoring station habitat description record sheet.

STATION	LATITUDE	LONGITUDE	QUADRAT	SOIL CHARACTERISTICS	VEGETATION SPECIES	VEGETATION HEIGHT (m)	OVER-STOREY (%)	TYPE	UNDER-STOREY (%)	TYPE	NOTES
e.g CC01	*-33.12345*	*115.12345*	*A*	*Moist, humic, dark brown*	*Agonis flexuosa; Lepidosperma gladiatum*	*7 over; 2 under*	*95*	*Tree (A.flexuosa*	*100*	*Sedge (L.gladiatum)*	
			B								