EAZA Best Practice Guidelines for Polynesian tree snails (*Partula* spp)

Edition 1.0
Publication date June 2019

Partula Snail EEP Species Committee
Editor Dave Clarke, ZSL

EAZA Best Practice Guidelines for Polynesian tree snails (*Partula* spp)

Terrestrial Invertebrate Taxon Advisory Group
TITAG Chair: Mark Bushell, Bristol Zoo Gardens, Clifton, Bristol, BS8 3HA
TITAG Vice-Chairs: Tamás Papp, Chester Zoo, Moston Rd, Upton, Chester CH2 1EU. & Vítek Lukáš, Zoo Praha, U Trojského zámku 3/120, 171 00 Praha 7, Czechia.
EEP Co-ordinator: Paul Pearce-Kelly, ZSL
EEP Studbook keeper: Sam Aberdeen, ZSL

Edition 1.0 Publication date June 2019
(based on global Management Guidelines document Nov 2007 eds Pearce-Kelly, Blake, Goellner & Snider)

Editor Dave Clarke, ZSL
Citation - Clarke, D. (2019). EAZA Best Practice Guidelines for Polynesian tree snails (*Partula spp*) – *First edition.* European Association of Zoos and Aquariums, Amsterdam, The Netherlands.
We acknowledge the invaluable input of all Partula snail EEP Species Committee members, SSP colleagues and global participating Partula collections.

DOI: 10.61024/BPG2019PolynesianTreeSnailEN

Preamble
Right from the very beginning it has been the concern of EAZA and the EEPs to encourage and promote the highest possible standards for husbandry of zoo and aquarium animals. For this reason, quite early on, EAZA developed the "Minimum Standards for the Accommodation and Care of Animals in Zoos and Aquaria". These standards lay down general principles of animal keeping, to which the members of EAZA feel themselves committed. Above and beyond this, some countries have defined regulatory minimum standards for the keeping of individual species regarding the size and furnishings of enclosures etc., which, according to the opinion of authors, should definitely be fulfilled before allowing such animals to be kept within the area of the jurisdiction of those countries. These minimum standards are intended to determine the borderline of acceptable animal welfare. It is not permitted to fall short of these standards. How difficult it is to determine the standards, however, can be seen in the fact that minimum standards vary from country to country.

Above and beyond this, specialists of the EEPs and TAGs have undertaken the considerable task of laying down guidelines for keeping individual animal species. Whilst some aspects of husbandry reported in the guidelines will define minimum standards, in general, these guidelines are not to be understood as minimum requirements; they represent best practice. As such the EAZA Best Practice Guidelines for keeping animals intend rather to describe the desirable design of enclosures and prerequisites for animal keeping that are, according to the present state of knowledge, considered as being optimal for each species. They intend above all to indicate how enclosures should be designed and what conditions should be fulfilled for the optimal care of individual species.

Summary

These EEP guidelines have been based upon the previous BIAZA/WAZA Management Guidelines for the Welfare of Polynesian Tree Snails, originally compiled in 2005 by Paul Pearce-Kelly, Edwin Blake, Ron Goellner and Andy Snider. They have been extensively enlarged & updated, however remain the result of an extensive collaboration of many participating institutions, agencies and individuals. These guidelines have been able to draw from decades of experience of all programme colleagues involved in working with *Partula*, including field observations, husbandry and diet trials, epidemiological and genetic health investigations, questionnaires and group discussions. Although some species have been lost in the past, the result is a distillation of what we have been able to identify as producing best results for species currently in the programme.

The 2018 EEP Studbook of Partula snails records over 8,000 snails of 15 taxa being maintained in 10 participating EAZA institutions. Fortunately, most *Partula* species have similar management requirements, although exact details may vary at individual institutions. *Partula* snails are by no means easy animals to keep and require particular care. However, they are extremely rewarding to keep, and if basic requirements are fulfilled, they can do very well. Success of the captive programme has led to our ultimate aim of releasing animals back into the wild, which since 2015 has seen nearly 10,000 snails of 10 species return to Polynesia.

The following agencies and IUCN SSC groups are also gratefully acknowledged, without whom the overall programme would not be possible: Direction de l'Environnement, gouvernement de la Polynésie Française; Délégation à la Recherche, gouvernement de la Polynésie Française; IUCN SSC Conservation Breeding Specialist Group; IUCN SSC Mollusc Specialist Group.

Also, personal thanks to those who helped refine the last details of this document.

Dave Clarke
May 2019

Table of Contents

Section 1: Biology and field data

Biology

1.1 Taxonomy

Phylum: Mollusca
Class: Gastropoda
Order: Stylommatophora
Family: Partulidae
Genera: *Partula*
Species *Partula* (100+)

These Best Practice Guidelines centre on the *Partula* genus, to which all the extant captive snails belong. The family includes two other genera, *Eua* (4 spp) and *Samoana* (25 spp), and although some species of the latter genus have been kept in captivity, they have proved very delicate and have not survived long term.

Common name: Polynesian tree snail is the general term, though some species in the genus live outside of the Polynesian realm. Some individual common names do exist, but they are virtually never used. On those islands where the snails played a cultural role (principally Huahine and Raiatea) the local Polynesian name for Partula was *areho* (ah-reh-ho). The general Polynesian name for shells is *pupu* (poo-poo).

A set of common names for extant species were approved in 2016, where possible using Tahitian translations of the scientific name. This list was created for those species current or recently in the captive programme, a concise and updated version is included here.

Partula common names for extant species					last edit 19 June 2019 TC/DC
As agreed at EEP Committee Feb 2016. Translations via Trevor Coote (with thanks to Djobrila Tiare)					
All to include the term tree snails, eg *Partula hyalina* = Poe tree snail.					
Partula species	**Common name**	**Tahitian**	**Pronounciation**	**Approximate meaning in Tahitian**	**Scientific name meaning**
affinis	**Marona tree snail**	marona	mahrona	brown	similar
garrettii	**Iareta tree snail**	iareta	yaretta	Tahitian pronounciation of Garrett	Named after Garrett
hebe	**Tapairu tree snail**	tapairu	tahpahiroo	beautiful youth of a goddess	hebe = Greek goddess of youth
hyalina	**Poe tree snail**	poe	pohweh	pearl	transluscent
mirabilis	**Navenave tree snail**	navenave	nahvehnahveh	marvellous	wonderful
mooreana	**Eimeo tree snail**	eimeo	eymayo	ancient name for Moorea	of Moorea
navigatoria	**Faatere tree snail**	fa'atere	fa-atehreh	navigation	
nodosa	**Niho tree snail**	niho	neeho	teeth	noduled
rosea	**Tarona tree snail**	tarona	tahrohna	rose coloured	rosy
suturalis	**Taamu tree snail**	ta'amu	ta'amu	banded	lined
ssp *strigosa*					grooved or ridged
ssp *vexillum*					standard, or feather
taeniata	**Parare tree snail**	parare	pahahreh	widespread	ribboned
ssp *nucleola*					core/central
ssp *simulans*					similar
tohiveana	**Tohiea tree snail**	tohi'ea	tohiya	current name for Tohivea	of Tohivea
varia	**Mauru tree snail**	mauru	mah'uru	many varieties	variable

Due to the intense genetic studies undertaken on Moorea in the Society Islands of French Polynesia, this is one of the few islands for which there exists a definitive taxonomy (Johnson et al 1992). Most contentious are those islands which were host to a number of different though similar species with many varieties, notably Raiatea and Tahiti in the Society Islands. Kondo (1968) was the standard taxonomic reference, though the latest species review is in Gerlach (2016).

Species and Red List assessments for French Polynesia (submitted by Trevor Coote, Partulid Global Species Management Programme, November 2007, and published 2009)

Genus *Partula*			
Island	**Species**	**IUCN category** (pub. 2009)	**Comment**
Society Islands (Leewards)			
Bora Bora	*P. lutea*	EX	
Huahine	*P. rosea*	EW	
	P. varia	EW	
	P. arguta	EX	
Raiatea	*P. faba*	EW	Now extinct as last captive died 2015
	P. fusca	EX	
	P. navigatoria	EX	Now to be reclassified EW due to misidentification.
	P. vittata	EX	
	P. radiata	EX	
	P. citrina	EX	
	P. imperforata	EX	
	P. formosa	EX	
	P. candida	EX	
	P. dentifera	EW	Now to be reclassified EX due to misidentification.
	P. callifera	EX	
	P. cedista	EX	
	P. auriculata	EX	
	P. robusta	EX	
	P. dolichostoma	EX	
	P. protracta	EX	
	P. leptochila	EX	
	P. labrusca	EX	
	P. dolorosa	EX	
	P. lugubris	EX	
	P. ovalis	EX	
	P. levilineata	EX	
	P. turgida	EX	
	P. remota	EX	

	P. atilis	EX	
	P. tristis	EW	Now to revert to EX due to misidentification.
	P. thalia	EX	
	P. rustica	EX	
	P. levistriata	EX	
	P. cuneata	EX	
	P. crassilabris	EX	
	P. garrettii	EX	Now to revert to EW due to misidentification.
	P. hebe	EW	Only subspecies *P. h. bella* survives
Tahaa	*P. faba subangulata*	EX	Extinct on this island. See *P. faba*, Raiatea
	P. planilabrum	EX	
	P. sagitta	EX	
	P. bilineata	EX	
	P. umbilicata	EX	
	P. eremita	EX	
Society Islands (Windwards)			
Moorea	*P. mooreana*	EW	
	P. suturalis	EW	Two subspecies included
	P. suturalis vexillum	-	
	P. suturalis strigosa	-	
	P. taeniata	CR	Three subspecies included
	P. taeniata elongata	-	
	P. taeniata simulans	-	(Would be EW)
	P. taeniata nucleola	-	(Would be EW)
	P. tohiveana	EW	
	P. mirabilis	EW	
	P. aurantia	EX	
	P. exigua	EX	
Tahiti	*P. jackieburchi*	EX	
	P. otaheitana	CR	
	P. affinis	CR	One known population surviving.
	P. cytherea	EX	
	P. nodosa	EW	
	P. producta	EX	
	P. filosa	EX	
	P. clara	CR	
	P. hyalina	VU	
Austral Is			
Tubuai	*P. hyalina*	VU	Tahiti endemic transported by man as part of the trade in shell jewellery
Rurutu	*P. hyalina*	VU	"
Raivavae	*P. hyalina*	VU	"

Rimatara	*P. hyalina*	VU	"

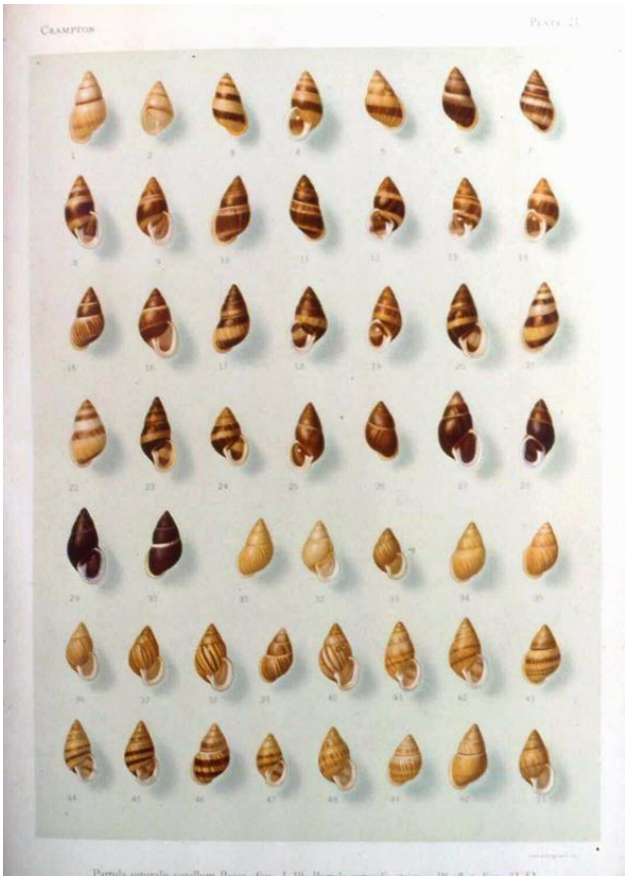
Species summary		**As of 2009**	
	EX	43	
	EW	11	
	CR	4	
	EN	-	
	VU	1	
	DD	-	
Totals		59	(= species from French Polynesia only)

Important note: Since the categorisations were last published, a number of species have been reclassified (Gerlach 2016) and these changes will be absorbed into the next revised assessments.

1.2 Morphology

It was the extraordinary polymorphism in shell size, shape, colour and banding patterns, and direction of coiling that attracted scientists to study the underlying genetic mechanisms of inheritance in the Partulidae. There is a wide range of intraspecific polymorphism in many species.

Shell length varies from around 12mm for species such as *Samoana decussatula* (Hiva Oa) to almost 30mm in *P. calypso* (Palau). Shells can be thin and translucent as is found in many species of the genus *Samoana* and two or three species of *Partula*, or thicker and more robust as in most species of *Partula* and *Eua*, and a few *Samoana*. Their shape ranges from long and slender to squat and globose. Shells can also vary in chirality, the majority being dextral (coiling to the right when viewed from above) but some species are sinistral (left 'handed') with a few having both left and right-handed shells in the same species. Chirality can be an isolating mechanism in evolution, and many species were seen to be actively evolving hence their scientific interest.



Variation in *Partula suturalis* (including chirality) from Crampton monograph 1932

Adult snail live weights can vary from around 1.9 grams in *Partula tohiveana*, one of the larger species, to only 0.5g in *P. garrettii,* one of the smallest.

Among approximately 60 species of partulids from the Society Islands, 29 have banded morphs, but only 5 out of the approximately 70 species of partulids beyond the Society Islands have banded morphs (Johnson et al 1992). Crampton (1932) designated 11 colour varieties and 11 banding varieties among Moorean *Partula*. Polymorphisms also exist in direction of coiling (on Tahiti and Moorea), mantle colour, proteins and mitochondrial DNA and these are reviewed in Johnson *et al* 1992.

1.3 Physiology

There appears to be little published information on the physiology of the Partulidae, though Kondo and Burch (1979) have made anatomical studies on genitalia, ostensibly for taxonomic purposes.

1.4 Longevity

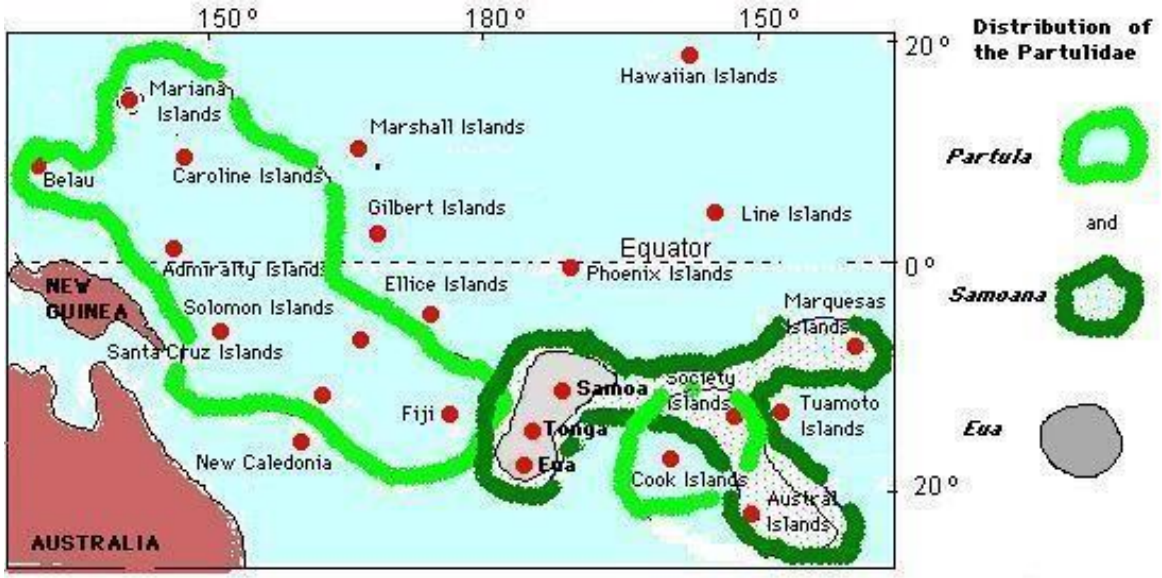
Partula are relatively slow-growing, long-lived and slow-reproducing land snails. *P. taeniata* from Moorea lives for at least 5 years after achieving maturity (Murray and Clarke 1984) and one individual was recorded as living for 17 years in the laboratory (Johnson *et al* 1992).

In general, they can be said to take about 1 year to reach maturity and live up to 10 years, although this can vary with species.

Field data

1.5 Conservation status/Zoogeography/Ecology

The snail family Partulidae, endemic to many of the volcanic islands of the South Pacific, have an enormous geographical range, from Palau, east of the Philippines, to the Marquesas Islands, 8500 km away to the west. Yet there are only three genera throughout: *Eua* (4 species) in Tonga and Samoa, *Samoana* (25 species) from east of Fiji to the Marquesas, and *Partula* (100+ species) from Belau to the Austral Islands. The epicentre of their evolutionary radiation is in the Society Islands of French Polynesia, where more than half of the recognised species were found, though their taxonomy has only recently been largely clarified (Gerlach, 2016). They are small, relatively inconspicuous, inhabitants of the forested slopes of many South Pacific volcanic islands, existing on the stems, trunks and undersides of leaves of many species of plant. There is evidence of microhabitat partitioning where species coexisted (Murray et al. 1982). Most species live at lower elevation, especially along the forested slopes of valleys, but there are a few high altitude species and some are found almost to the coast.



Natural distribution of Partulidae: *Eua* (black line), *Samoana* (dark green line), *Partula* (light green line). From Cowie 1992.

Despite some localised habitat loss, the biggest threat to the Partulidae has been from introduced predators, in particular the Rosy wolf snail *Euglandina rosea* and more recently the New Guinea flatworm *Platydemus manokwari*. As a result, most surviving species are IUCN Red listed as Critically Endangered (CR) or Extinct in the Wild (EW), see species list in 1.1 for full details. An integrated conservation breeding plan has been running since 1994 (Mace et al 1998).

History of the conservation programme

Following widespread extinctions of partulid tree snails due to the misguided introduction of *Euglandina* (Clarke et al, 1984) the International Partulid Conservation Programme was established in 1986 (see Appendix 1 for conservation status table). Since its inception, the International *Partula* Breeding Programme has been the driving force for developing in-region conservation initiatives aimed at mitigating the effects of introduced predators.

Although eradicating *Euglandina* or *Platydemus* is probably unrealistic, any future conservation measures will be heavily reliant upon having a detailed understanding of the predator dynamics in these island ecosystems. Current conservation measures are therefore focusing on clarifying the degree of spread, population fluctuations, interspecific and ecological dynamics. Although *in situ* measures trialled the creation of *Euglandina* exclusion reserves, a strategy pioneered by the Partulid Programme and further evaluated on Hawaii, these have not proved practical long term and anyway would be ineffective against *Platydemus*.

From the mid-1980s the Partulid Programme's in-region conservation efforts have developed through five distinct but integrated phases, which can be summarised as follows:

Crisis rescue phase (1985 – 1996)
This initial phase involved a succession of fieldwork (largely implemented and supported by programme member institutions) in French Polynesia on most of the Society and Marquesas Islands predominately aimed at rescuing populations of threatened partulid species before *Euglandina* swept through their range valleys. This crisis necessitated the establishment of an international breeding programme whereby 25 partulid taxa were taken into management facilities in North America and Europe. This programme continues to maintain a large number of *Partula* taxa in closely managed breeding groups.

Investigating predator spread and practical protection (1994 – 2003)
The second phase (again led largely by programme member institutions) involved a series of more intensive surveys on the principal range islands, to determine the extent of predator invasion and its impact on the native mollusc fauna. This work, carried out in close collaboration with a wide range of partners in the local community, confirmed that 15 of the rescue-collected *Partula* species were extinct over their natural range.

This phase also saw the development, testing and construction of predator exclusion partulid reserves (measuring 20 x 20 m^2) in forest natural range habitat. Throughout this period the reserve strategy remained the most practical measure to address the *Euglandina* threat to the surviving endemic species. It could be used to ring-fence surviving wild populations, or to re-establish species lost from their natural range using the EEP and SSP breeding programme populations. Two such reserves (the world's smallest wildlife reserves) were constructed on Moorea and Tahiti, with a further two erected on O'ahu in the Hawaiian Islands using the model developed on French Polynesia.


Moorean snail reserve in 1996 (Dave Clarke/ZSL)

Developing a conservation management strategy for the region (2003 – 2014)
If the first two phases were a combination of crisis rescue collections and associated predator impact research, this third and most significant phase addresses the future conservation requirements of the region through the development of a formal regional conservation management strategy for the French Polynesian government.
Since the mid 1990's Dr Trevor Coote has been the principle investigator of the endemic snail extinctions and developing the potential for predator exclusion reserves in French Polynesia. This sustained conservation effort has forged a conservation alliance with local communities and key government agencies. These include la Direction de l'Environnement de Polynésie Française, la Délégation à la Recherche, le Musée de Tahiti et ses Isles, welfare and artisan NGOs, landowners and individuals living in and using forest habitats in key endemic species areas. In 2003 the French Polynesian Environment Ministry (Direction de l'Environnement de Polynésie Française) and the International Partulid Conservation Programme commenced an ongoing collaboration to develop, fund and implement a conservation management strategy for the region's endemic tree snails and their associated montane forest habitat. An intensive set of field survey work resulted in the *Conservation Action Plan for the long term protection of French Polynesia's last surviving populations of endemic tree snails of the genera Partula, Samoana and Trochomorpha* (Coote 2005). The implementation elements of the Action Plan were being realised through the ongoing collaborate efforts of the FP Environment Ministry and the International Partulid Conservation Programme and the Action Plan revised annually to take account of developments.

Active re-introduction (2015-2019)
The fourth phase is the process of the actual field reintroductions. In 2012 the first predator exclusion reserve was constructed in Te Faaiti Valley on Tahiti. At that time this was the only strategy on the table for the reintroduction into the wild of *Partula* species maintained in the breeding programme. However, this strategy was quickly superseded and then discarded in the light of important observations in the field. The first was the discovery of surviving populations of *Partula clara* in Tahitian chestnut trees (mape) in valleys invaded by *Euglandina*. This offered the possibility of large trees acting as refuges from predation, their dry, dusty trunks acting as considerable deterrents to what were basically ground predators in moist habitat. When the first stock of three species of *Partula* were released into the reserve in 2015 after a three-year delay, a simultaneous control experiment of releasing *Partula nodosa* directly into *mape* trees in another valley took place.

The second important observation was that the snails released into the reserve quickly died out in testing conditions but those released into the *mape* trees rapidly dispersed into the higher branches, with apparently low mortality and newborns observed. In contrast, it was evident that, in addition to its failure to keep the snails alive, the almost prohibitive consumption of resources in terms of cost and labour required, its inaccessibility and impracticability to be extended to other locations on Tahiti (and certainly onto other islands) meant that the strategy of releasing *Partula* directly into trees was the only realistic option available.

Over the space of four years there have been six shipments from Europe containing nearly 9,000 individuals of 11 species and one subspecies (12 taxa) of *Partula* that have now been released onto three of the four target islands. The releases have all been followed up by intensive and regular monitoring. Although the snails have, in the main, quickly dispersed into the trees where they were released, evidence from dead shells suggests that mortality has been low. A few individuals (colour-marked by year) of some species have been recorded the following year, and some early recruitment observed. The one negative aspect was the unexpected loss of at least half of the three species released onto Raiatea due to predation by *Platydemus manokwari*. Subsequent research showed that the flatworm was also present at every release location on Tahiti and Moorea but had little apparent impact. It is most likely that the invasion on Raiatea was more recent and surveys on this island and Huahine did demonstrate that density of flatworms on these islands was indeed more severe.

Release of *Partula hyalina* on Tahiti 2016 (Dave Clarke/ZSL)

Post-release (2020-)

Smaller releases of *Partula* will continue into the near future but further surveys to determine the status of *Playdemus* on both Raiatea and Huahine will be carried out in 2019 before any releases onto those islands will take place later in the year. A workshop initiated by the French Polynesian environment department is planned for August in 2019 to clarify the respective roles of both the zoo community and the local government in the future of *Partula* reintroductions, continual monitoring and hopefully, reestablishment.

1.6 Diet and feeding behaviour

Wild partulids are principally detritivores, feeding on a range of different decaying plant material from both endemic and introduced species. Some species are associated with specific plant habitats, whereas others are herbivorous generalists. On Moorea, *Hibiscus tiliaceous* (*purau* in Tahitian) is the species most often associated with *Partula* feeding habits. Many species were found living in stands of Climbing pandanus *Freycinetia impavida* (‘*ie’ie*)*,* which traps leaf litter from the *Hibiscus* canopy. In comparison, *Partula rosea* from Huahine were usually found high in the leaf whorls of Screwpine trees *Pandanus tectorius* (*fara*), with no direct upper canopy. The snails remain fastened to leaves during dry periods but emerge to feed and mate when it rains, mostly at night.

The chemical components of the dried outer stalk layer of field-collected *H. tiliaceous* have been analysed in the laboratory, and gave 9.6% moisture, 6.4% ash, 1.8% protein and 1.1% fat, 24.4% crude fibre, with carbohydrates assumed by difference to be 56.3% (PNL report, 1991). A wider range of partulid associated Polynesian plants have also been analysed.

Gerlach did a study in 2014 looking at the gut contents of alcohol specimens collected from Moorea in the 1960’s. Samples from 8 species indicated that they could be placed in four main ecological groups, with some overlap: detritivores, omnivores, plant grazers and fungal feeders. The surviving species in the captive programme (*tohiveana, mooreana, taeniata, suturalis* and *mirabilis*) mainly fitted the detritivore group.

One species on Moorea, *Partula exigua,* was the only known partly predatory species of *Partula*, eating other snails (Johnson et al, 1993). It is possible that larger snails do sometimes eat newborns, perhaps mainly to re-ingest calcium (as evidenced by otherwise unexplained disappearance of newborns in containers in captivity).

1.7 Reproduction

Most species are cross-fertilising hermaphrodites, with self-fertilisation relatively rare. The overall rate of selfing of *Partula taeniata* in the wild was estimated from allozyme studies to be only about 2%, though this could reach 20% in the first group of young (Murray and Clarke 1976a). In *P. suturalis* from Moorea selfing occurred throughout life at a rate of about 2%. In *P. gibba* of Saipan, where self-fertilisation is the normal reproductive strategy, crossing has, nevertheless, still been detected.

1.7.1 Gestation period and offspring size/number

The family is named after Partula – the Roman goddess of birth. All species of partulid are ovoviviparous, with 1-2 mm newborns growing to adulthood in as little as 3-6 months (Johnson et al. 1993b), although larger species are known to produce young up to 5mm in shell length and can take up to a year to mature. Usually only one baby is produced, but occasionally twins. Several young may be present at different stages in the oviduct, with a gestation period of approximately 3 months, giving an average reproduction rate of 1 birth

per month (Murray & Clarke 1984). Reproduction is year round but may be influenced by wet season/dry season, although this is generally minimal (Trevor Coote, pers comm).

Note that eggs are occasionally produced, but these are normally infertile developments with no visible embryo, or remnants of egg shell may be seen around a presumed premature birth.

1.7.2 Developmental stages to sexual maturity

Growth from newborn to adult is continuous, with shell shape changing dependant on the species. All stop growing in size at adult, with most species (particularly in *Partula*) forming a thickened lip around the shell opening defining maturity.

In captivity four developmental stages were defined at the start of the formal breeding programme, particularly when it was important to monitor growth and keep separate generations. These categories are also used to help identify age of wild specimens. These are generally identified as follows, although they do depend upon species size –

Development stage	Detailed explanation
Newborn – **Any snail below 5mm in shell length.**	Freshly born snails can vary in size within species but particularly across species. As individual snails cannot normally be identified, 'newborn' is important in capturing all snails which are close to neonate.
Juvenile – **Snails with shells of 5mm or over, but not yet definable as Subadult.**	Once 5mm or above all snails have shown some development growth beyond newborn. The juvenile stage captures any snails between newborn and close to reproductive stage.
Subadult – **Snails of close to adult size for the species, but with no shell lip.**	It has been important to identify snails reaching maturity, but not yet able to breed, particularly when keeping separate generations. Size of subadult is entirely linked to expected adult size of each species, so can vary from 10mm – 30mm. It is a short stage but helps define large juveniles close to reaching adult stage.
Adult – **Snails with a visible lip on the shell.**	Fully mature *Partula* snails usually have a thickened edge to the shell opening, and the shell stops growing in size. This is the only reproductive stage. Snails are classed as adult as soon as the lips starts flaring out

	from the whorl, as full thickening of the lip is usually very quick (within 1 week).

Partula mirabilis (l-r) adult, subadult, juvenile, newborn


Underside of *Partula tohiveana* adult (left, with lip) and subadult (no lip)

Although generally this applies to all species, there are some caveats. For example, when this system was first implemented, all captive species did not give birth to young above 5mm in shell length. This changed when *P. faba* was first collected, as this large species was able to produce young of around 5mm (although sadly this species has not survived long term).

Also, some species have a lip which does not flare outwards that visibly but is clearly thickened when viewed from the underside of shell aperture – examples are *Partula navigatoria* and *P. garrettii*.

1.8 Behaviour

1.8.1 Activity and locomotion

In dry weather partulid snails seal themselves to their substrate but emerge and are more active after rain, especially at night, when they tend to move from resting underneath leaves to actively foraging.

Experiments have determined the rate of dispersal of *Partula taeniata* at a mean of 2.8m (SD 1.6m) movement from the sites of their original capture (Murray and Clarke 1984) after 5 years. The maximum recorded gene flow measurements of 10 m after 1 year, and 27 m after 13 years have been obtained. These measurements confirmed the hypothesis of very small neighbourhoods - approximately 200 m^2. Accelerated range expansion has occurred through the establishment of populations by rare long-distance movements (passive dispersal) (Nichols and Hewitt, 1994). Recent re-introductions to the wild have shown strong upward dispersal after release (Coote et al 2019).

1.8.2 Predation

Natural predators of Partulid snails would have been few but included native birds and lizards. Polynesians used to collect the snails for ornaments such as necklaces known in Tahitian as '*hei*', but there was no evidence of this significantly affecting wild populations. The introduction of alien predators has been the main recent threat (see 1.5).

1.8.3 Social and sexual behaviour

Snails of several species of *Partula* used to be found in very high adult numbers in suitable habitat before the impact of predators, with densities of 20 per metre (Johnson et al 1993). In contrast, some *Samoana* would only be seen individually or in small numbers, however this may be linked to them being more montane. Young stages were rarely seen, especially newborns, which are obviously harder to observe but also either hide effectively in foliage or were more elevated.

Where different species were found in the wild, they were rarely found together, with the expectation that each had some subtle preference for niche usage. A maximum of three sympatric species were found on Moorea (Johnson et al, 1993).

Mating behaviour is similar to other terrestrial snails, though partulids do not fire 'love-darts' as is observed in many other families. Courtship behaviour has been described by Lipton (1979) and Lipton and Murray (1979).

Section 2: Management in Zoos and Aquariums

Partulid species and sub-species maintained in the International conservation breeding programme (as of January 2018):

Taxon and original island of collection	Year of original field collection	Origin of stock
French Polynesia		
Tahiti		
Partula affinis	1995	Baie Pierere, Te Pari (Tahiti Iti)
Partula hyalina	1987 + 1995	Tahaute Valley, Mix
Partula nodosa	1984	Papehue Valley
Moorea		
Partula mooreana	1985	Atimaha ridge, Maatea Valley
Partula mirabilis	1984 + 1985	Mix
Partula suturalis vexillum	1982, 85, 86	Atimaha ridge, FareaitoValley, Haapiti Valley
Partula suturalis strigosa	1980 + 1985	Hotutea Valley, Maatea Valley
Partula taeniata nucleola	1981 +1982	Faatoai Valley
Partula taeniata simulans	1982 + 1986	Haapiti Valley, Hotutea Valley
Partula tohiveana	1982	Fareaito Valley
Huahine		
Partula rosea	1987	Mahuti Valley
Partula varia	1991 + 1994	Mix, Fare Valley
Raiatea		
Partula navigatoria (prev. dentifera)	1991	Hamoa Valley
Partula hebe bella	1991	Hotopuu Valley
Partula garretii (prev. tristis)	1991	Tevaitoa Valley

The captive maintenance of partulid snails has evolved over time into the main recommended procedure as follows. Although some experimentation has taken place, the snails do seem sensitive to change, and therefore caution is advised with any deviation from this procedure (which should only be considered with agreement from the species co-ordinator). The current protocols have worked very well for the majority of species so should be adhered to, unless agreed experimentation with snails deemed as excess stock.

Note these intensive 'artificial' rearing methods have proved very successful over the long term (several decades without supplementation from the wild) for most taxa. The snails have never thrived in naturalistic enclosures e.g. with plants and soil substrate, and there has been very limited success with releases into biome-type forest displays, but no long-term survival.

2.1 Enclosure

Although populations of snails are maintained in a number of individual containers, a key enclosure consideration is the wider room environment in which they are held. Ideally this should be a dedicated animal room capable of providing the required temperature and humidity range (20-24˚C and 60 – 80% respectively). See 2.1.4 for environment details.

It is recommended the room where the snails are held is self-contained for maintenance and improved quarantine, with shelving racks, worktops, dedicated equipment and spare container storage and a sink/drainer plus room for a dishwasher. The room should have a threshold to allow suitable quarantine, with isolation from other species (particularly molluscs) and use of lab coats. If possible, this facility should be on display, where the public can see the snails and staff maintaining them, with interpretation to explain the story. A variety of suitable graphic information is available to share, see EAZA Committee for assistance.

Room design should also consider details such as having easily cleaned surfaces for sterilising. The floor should be of a 'soft' material such as linoleum, rather than stone or concrete where a dropped snail is more likely to be damaged on impact.

Individual enclosure design has evolved greatly from the initial lab-based keeping system of small plastic sandwich boxes (11cm wide by 17cm long by 5cm deep). Although these units are still used in a small number of cases (where very small populations are being maintained) comparative keeping trials have resulted in the current standard aquarium glass tanks (3mm glass with simple silicon sealant joins). The two main sizes in use are 40cm long by 25cm wide by 30cm high and the larger 50cm long by 25cm wide by 30 cm high. The former size is designed to allow a good fit of standard hygiene roll tissue to make maintenance easier.

Standard glass tanks with clingfilm top and two feed plates at Bristol and London

All containers of snails should be clearly marked to identify the populations they hold. This should include the enclosure number, ZIMS group number, species of *Partula*, and any further useful information. This should of course be linked to the ZIMS records.

It should be added, there have been times when populations of some species have been high enough to allow experimentation with releases into biome-type areas. This has not proved very successful long term therefore this option is not discussed in detail here.

However, this has allowed some observations of snails in semi-natural conditions (particularly see Pearce-Kelly et al 1995).

2.1.1 Boundary

In addition to being able climbers, new-born snails are only a few mm long and therefore the tanks need to be escape-proof. A clingfilm cover over the top of the tank prevents escape and helps to provide the micro-humidity conditions. The same procedure applies for the Perspex boxes with the exception that the Clingfilm is placed on the open front of the upright box. The clingfilm must be of a type identified by the food safe symbol, and semi-permeable 'breathable' type, not just plastic wrap. Perforated catering clingfilm on a roll is easiest to use (see product list). The clingfilm needs to be sealed all around the outer edge of the tank, but care should be taken not to over-tighten which can lead to splits at the glass edges or tears in the perforation join. The clingfilm can be re-used and part rolled back when servicing, but normally needs replacing to remove build-up of slime/faeces after a few weeks dependant on stocking levels.

Care needs to be taken when servicing the container or counting the snails (especially if placed out on a worktop surface) as they can be surprisingly speedy and zip off while your back is turned!

2.1.2 Substrate

The bottom of the standard glass tank has four layers of 2-ply tissue roll (often called hygiene roll, R302 or equivalent). The small plastic box containers are provided with a similar tissue base folded to size to create the same 4 layers, or single lab box type tissue (which typically measure 20 x 20cm) repeatedly folded so as to cover the base of the up-turned box (see photo).

Box type containers with tissue base

Caution: As the snails will ingest some of the tissue substrate, care should be taken to avoid use of perfumed or printed pattern varieties.

The tissue substrate needs to cover the entire base of the tank (or box) and is dampened with the same filtered tap water which is used for the diet make up (see 2.2.4) to a saturation level that is damp to the touch but not sodden. If the tank substrate tissue appears too dry it can be given a spray in between feed change times, but if too wet the base tissue should be changed.

2.1.3 Furnishings and Maintenance

Other than the two Perspex food plates (see 2.2.3 and above photo) the only other furnishing is a small shallow-sided Petri dish (usually 5cm diameter dishes) to keep the cuttlefish bone off the damp substrate, this can also be given holes cut out of the base, to facilitate drainage from accidental spraying (see 2.2.2). The food plates do also provide additional climbing and retreat areas. As the snails are arboreal in nature they will tend to favour the sides and top of the containers so what might at first appear to be a sparse and artificial set up does in fact allow the animals to express their natural climbing behaviour (in nature this would most often on the undersides of smooth leafed plants) and provides the required smooth surface for the snail to adhere to when resting.

Often the snails exhibit thigmotaxis, where they prefer to be in contact with another surface, so will congregate at tank corners, under food plates or touching each other. This needs to be born in mind when servicing and they can also sometimes rest within the tissue, so it is best to press down the edges of the tissue base to retrieve small snails hiding in this way and avoid them potentially being accidentally thrown away.

Maintenance guidance
Collections do vary with exact details of how the snails are serviced, so general details are given here. It does appear the snails are sensitive but do get used to a routine way of being maintained. Therefore, as long as a particular method works at an institution, this is fine. The programme co-ordinators would prefer that any major changes being considered are discussed before implementation.

It is recommended the enclosures are fed and given a basic clean at least twice a week, with smaller plastic boxes best serviced three times weekly. The cycle is basically cleaning the enclosures, damping new tissue and replacing the food, then letting the containers naturally dry out before servicing again – this mimics periods of rain and dry the snails would experience in the wild. Some collections only change the base tissue once a week, just replace the foodplates once or twice in between, to minimise disturbance. See Appendix 4 for a detailed example of a daily servicing regime.

It is important hands are thoroughly cleaned and rinsed before maintenance (see 2.6.2). The snails are very delicate and newborns small, therefore using bare hands rather than gloves is preferred.

Snail servicing at Edinburgh Zoo (Ross Poulter/RZSS)

The sides of the containers do not need to be thoroughly cleaned each time. They can be given a basic wipe with damp tissue, for example properly cleaning about half the panels. This prevents too much disturbance of the snails and they are sometimes seen to eat faeces, which may allow re-assimilation of gut flora. Note the build-up of transparent slime on the glass needs to be removed, not just the faeces. When snails are moved for cleaning, they can be placed on a piece of tissue or, to prevent over-handling of young, put in a petri dish which can then easily be placed back in the tank to allow them to move off themselves (this can be particularly useful when counting).

Partula snail re-ingesting faecal material in captivity (Dave Clarke/ZSL)

Containers should be given a thorough clean approximately once a month, to remove all build-up of faeces and slime. This can usefully be carried out at the end of the month when a thorough count is made. Chemical detergents can be used but must be thoroughly rinsed afterwards, or very hot water can be adequate. Steam cleaners (which if suitable could remove the need for any detergent use) are also being investigated.

Basic servicing for a small number of containers takes a relatively short period of time given the overall number of animals, each tank requiring only about 5 minutes. However, when you consider preparation, especially diet, and regular thorough counting of the snails and record keeping, this can mount up. When keeping a large number of containers, it becomes quite a commitment, as care of *Partula* populations can take several hours, 2-3 times per week, and this needs to be apportioned for.

Once again natural substrates are not normally recommended in captivity, unless the snails are part of an agreed trial, for example in a biome exhibit.

2.1.4 Environment

Environmental parameters are extremely important for all molluscs. Partulids typically come from montane rainforest areas, where temperatures are cool tropical. Experience has shown that in captivity, keeping the holding room environmental conditions within a temperature range of 20 – 24˚C and a humidity range of 60 - 80%, produces the best background conditions for the dampened tissue containers to generate the snails' micro-environmental conditions. Short-term fluctuations either side of these background ranges can be tolerated without undue stress but are to be avoided over sustained periods. It is recommended the temperatures stay as stable as possible. Absolute range beyond which snails are likely to suffer are below 18 degrees or at and above 28 degrees centigrade. Note that just after servicing the internal humidity in the containers can go to 99%, but this naturally drops over time due to the semi-permeable clingfilm.

It is also acceptable and indeed recommended for the substrate to become relatively dry to the touch by next service period, usually after a few days.

Temperature in the breeding room can be controlled by domestic air conditioning systems with suitable thermostat control, ideally in tandem with humidification, as air conditioning tends to dry the atmosphere. An active form of chilling is important where background temperatures may become high e.g. in the summer months, but where this is less of an issue, just basic heating can be provided (as at Edinburgh Zoo).

Temperatures should be monitored daily by checking a min/max thermo-hygrometer so that any issues can be corrected swiftly. Ideally an electronic datalogger should be used in the room to give a graph reading (data from which can also be uploaded to ZIMS). The snails are precious enough to consider remote environmental monitoring systems to alert any system failures.

Some ventilation for the room is required, although should not overly compromise environmental control.

Lighting has not proved a known issue, and the snails are normally nocturnal. It is generally recommended for invertebrates that full spectrum lighting is provided, with a 12:12 photoperiod for tropical species, controlled by timer. *Partula* do well under relatively subdued lighting, fluorescents being adequate and low temperature. T5 is recommended to avoid any flicker affect. For example, at ZSL London Zoo a room 3m x 3m has three Arcadia full spectrum 6% UV luminaire units of 39 watts across the ceiling. However, Edinburgh Zoo use no full spectrum and only lighting, and the snails do very well. There may be species-specific variables yet to be determined. An additional room light may be considered beneficial for servicing.

It is important to prevent any contact with chemical cleaners as the snails are very sensitive to contamination. Any use of cleaning chemicals must be minimised & thoroughly rinsed afterwards.

2.1.5 Dimensions

The two main glass tank sizes in use are 40cm long by 25cm by 30cm high, and 50cm long by 25cm wide by 30 cm high. The standard small plastic boxes size is approx. 110mm wide by 170mm long by 50mm deep, with an intermediate box size of 160cm wide by 280mm long by 90cm deep (boxes supplied by Stewarts Plastics, see Appendix 4).

Stocking density can be up to 50 adults and associated young in the tanks, dependant on size of species. Therefore, for a small partulid species there can be up to ~150 snails per tank, however this can mean more frequent cleaning is required. In contrast the smallest box may only be suitable for up to 5 adults and their young.

2.2 Feeding

2.2.1 Basic Diet

The recommended EAZA region diet is as follows:

- Grass pellets 300g (*Drygrass* Ltd - 25% oil, 16% protein, 25% fibre, 9% ash).
- Oats 300g
- Trout pellet 150g (*Vextra* Trout Intermediate 3mm -18% oil, 45% protein, 2% fibre, 8.5% ash)
- Cuttlebone 150g (only the clean inner part is used).
- *Stress* multi vitamins 25g (see supplements notes).

The diet is reduced (one ingredient at the time) in a coffee bean blender to a fine powder and then combined. Providing the diet is kept dry in a sealed container the prepared diet can be stored for up to six months. Use by dates should be monitored for all individual ingredients are replaced as necessary.

The required amount of dry diet is mixed with filtered tap water to create a runny paste, usually using a spatula. This is then left for five minutes to absorb liquid after which more water is added to reform a runny paste. For the plastic box keeping system a small amount of diet is smeared directly onto one of the upright sides.

There are problems obtaining some of these diets in the AZA region and the following alternative diet has been used for many years:

- Trout Chow 1.5 tsp
- Bone meal 3 tsp
- Dried nettle 3 tsp (nettle herb/Michael's herb)
- Quaker rolled oats 3 tsp
- Stress supplement 25mgs
- Vitamin E (Harlann Labs) 20mgs

As mentioned above, snails will often also ingest tissue from the food plates or enclosure base. This is basically cellulose so is not a concern, although it is important to ensure uncontaminated tissue is used and be aware that any excess eating of tissue in preference to diet may indicate an issue.

Vegetables (e.g. lettuce, carrot) are known to have been taken by the snails but are not generally recommended. The diet above is designed as a complete food, and fresh vegetables can introduce the risk of chemical contamination.

Extra cuttlefish bone is also provided, to enable the snails to obtain additional calcium if needed. Generally, this is readily taken by the snails. It is important to use good quality cuttlefish bone with the outer surface removed to reduce bacterial growth. The cuttlefish bone should be changed weekly, particularly if well chewed. A cut piece approximately 20x10x10mm can be provided per tank, and it is recommended that cuttlebone is placed on a shallow plastic dish rather than directly on the wet tissue, as it absorbs water.

2.2.2 Method of Feeding

Depending on the population of the container, the required amount of paste is spread onto the food plates or box side using a spatula. Care should be taken not to spread the paste too thickly as this can result in fungal growth. Feeding is recommended to be every second/third day but not normally any longer to prevent spoiling.

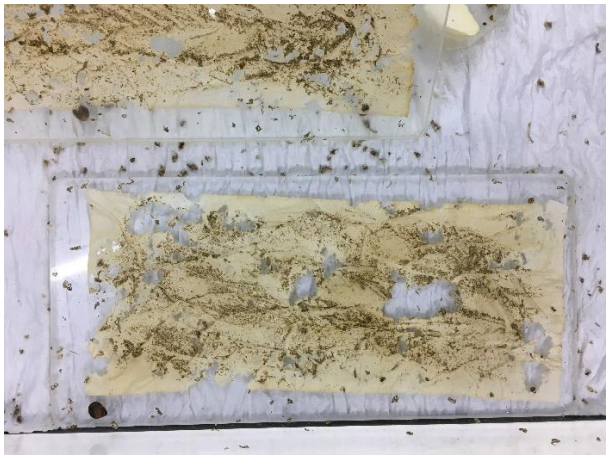
Two feeding methods are currently used in the tank systems, both using acrylic food plates of 5-6mm thickness, usually 280mm by 130mm (although larger 320 x 150mm have also been used):

a) Paste is spread onto a single piece of dampened tissue (same type as the substrate tissue) that is placed on the feed plate and positioned on one side of the substrate. The second feed plate is placed on the base plate or on the tissue close to it (to avoid slipping on the base plate) and lent against the tank wall. The tissue is cut to ensure a gap of at least one centimetre between the edge of the tissue and the edge of the acrylic. This reduces the risk of food transfer to the substrate.

Newly prepared food plates at Edinburgh Zoo (Ross Poulter/RZSS)

b) As above only without using tissue substrate under the food. When no feed plate tissue substrate is used the diet needs to be a little thicker to prevent run off.

There are differing opinions on whether it is necessary to use tissue under the food on the plates, although it probably helps keep the food damper for longer, it also adds time to overall food preparation. Often snails consume quite a lot of the tissue when feeding, which appears to do them no harm, as it is primarily cellulose anyway.



Heavily chewed tissue by *Partula mirabilis* (Dave Clarke/ZSL)

If a large amount of diet is prepared, it is best to remix from time to time, to keep it blended (some elements can start to drop to the bottom of the dish).

Food plates can be communally soaked in standard Milton solution for sterilisation, then washed in a dishwasher with no detergent at 70° intensive wash. As the plates are re-used in different enclosures each time, this helps minimise potential spread of disease.

The only other furnishing is the small Petri dish, used to keep the cuttlefish bone off the damp substrate. The shallow lid or deeper base can be used, dependant on the size of snails/container. This can also have a few holes cut into the base to provide drainage.

2.2.3 Water

To avoid variable direct mains water quality, it is recommended to use filtered water using a standard domestic water filter unit e.g. 'Brita' water filter (a large capacity version e.g. the 8.2 litre 'flow' version is recommended) or a dechlorinising carbon filter attached to mains supply. It is best to use freshly filtered water rather than left in any containers for several days in a warm room, where bacteria may multiply. Water should be allowed to reach the room temperature before use in tanks or diet.

2.3 Social structure

2.3.1 Basic Social Structure

See section 1 for comments on wild structure. In captivity, partulid snails will live in groups ranging from a few animals to over 100 individuals of mixed-age classes. No aggression has ever been observed although overcrowding stress cannot be discounted (especially in the context of higher infection potential).

The significance of density related factors such as increased slime trails might be an additional group benefit (for instance assisting new born snails to locate feed areas) which needs further investigation.

2.3.2 Changing Group Structure

In addition to birth, death and growth-related fluctuations in the resident populations, immigration and emigration can be regular features of the optimum population management protocol. Mixing populations can introduce fresh breeding opportunities but also risk introduction of disease. Transfers will normally need ratification by the species co-ordinator.

2.3.3 Sharing Enclosure with Other Species

At the room level it is very important not to have any other non-partulid mollusc species in the same area. At the individual tank and box level there should not be any other species. For research trials surplus partulids have been released into greenhouse condition environments (including exhibits) which can include other species but such populations are considered separate to the core-managed programme populations and should not be transferred back to core populations without prior discussion with programme coordinators.

2.4 Breeding

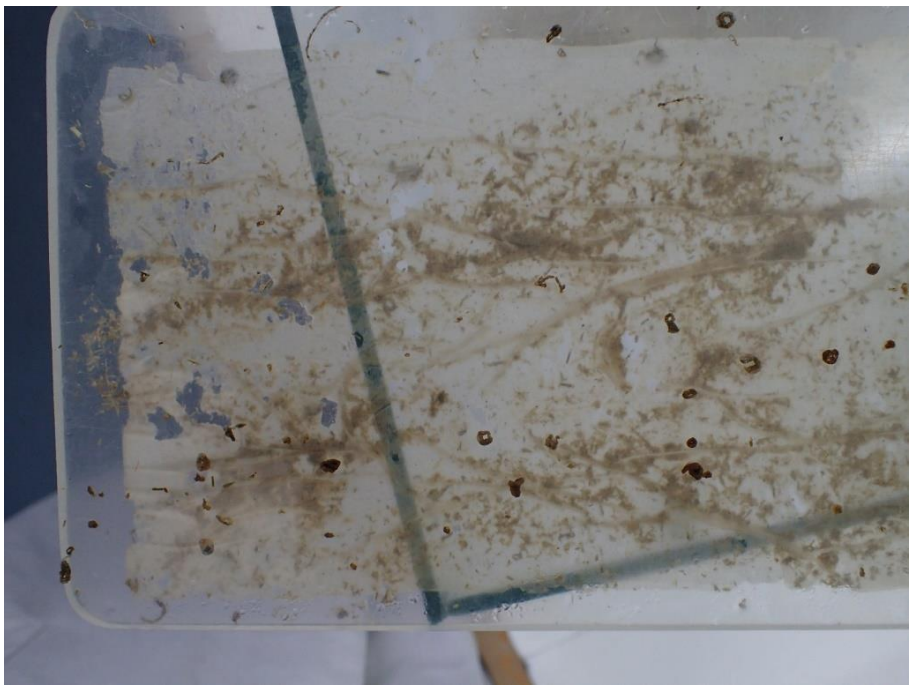
2.4.1 Mating

Few courtship observations have been recorded either in the field or in captivity, although direct mating is often seen in enclosures with good numbers of adults. Mating occurs more frequently at night (the main activity period) but can also be observed during the day – especially after the enclosure has been serviced with resultant elevated humidity levels. Self-fertilisation is also possible (see Biology Data section for further details).

Partula navigatoria mating in captivity (Dave Clarke/ZSL)

2.4.2 Pregnancy/birth

Young are born to the side of the adult and are immediately independent. They are usually left in with the container population, requiring no special attention other than extra care due to their small size that they are not accidentally disposed of when servicing. Newborn snails, often only a few millimetres in shell diameter, can be difficult to see amongst the faeces in an enclosure, even for the most experienced of staff, so must be checked for carefully.



Spot the newborn *Partula hebe*…

…see it now? (Dave Clarke/ZSL)

For this main reason in some cases young may be separated into rearing containers for more close scrutiny, using the smaller box method. It is however not impossible that some newborns disappear through cannibalisation.

As in 1.7.1, eggs are occasionally produced in captivity but are usually non-viable. They can be disposed of and only noted if desired, although a large number of eggs being produced may indicate an issue.

Eggs produced by *Partula navigatoria* (Dave Clarke/ZSL)

2.4.3 Development and Care of Young

In pure and spanning generation populations the young need to be removed from their parents before they reach the post sub-adult stage (i.e. when the 'lip' is clearly defined around the operculum of the shell). In mixed generation populations young are allowed to develop through to adult and combine with the parent population. No direct post-natal parental care has been observed to date, although it is possible young benefit from staying with adults by sharing gut flora.

Note mortality is usually highest in the newborn stage, as it is the most delicate and most prone to desiccation. Normally dead newborns can easily be identified when the soft body dries up, by holding the shell up to light (candling). It can be difficult to confirm straight away if an older animal is dead, as they can retract into the shell. However it is obviously important to try and remove any dead animals as soon as possible. See appendix for mortality guidance document.

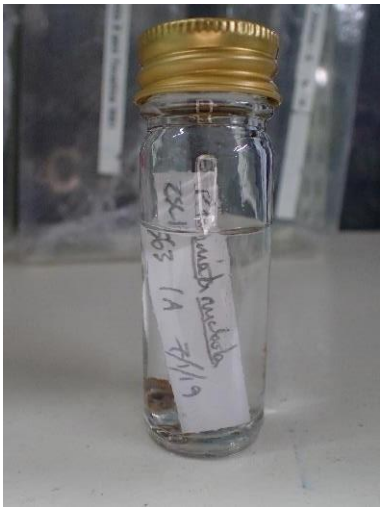
2.4.4 Population management

The current agreed target population level is 240 adults and associated younger life stages for each taxa, as identified at the 1994 meeting (Pearce-Kelly et al, 1994). The International Breeding Programme was established in 1986. The SSP Partulid Programme was established in 1989 and the formal EEP Partulid Programme in 1995. The annual International Partulid Studbook is the principle breeding programme dataset.

The culling of some over-represented *Partula* populations can be an essential management tool for ensuring earliest viable generation representation and the avoidance of health-related problems that can arise from overcrowding. In all cases, it is important to liaise with the respective regional programme coordinators before any culls are undertaken.

Euthanasia of individuals might be necessary for a number of reasons: culling of surplus stock as a population management tool, disease monitoring or individual welfare. It is important to avoid unnecessary suffering during euthanasia, and humane methods need to be employed which achieve rapid unconsciousness and death with minimal pain and distress (BIAZA Euthanasia Guidelines 2018). The most humane method for young snails can be physical crushing, however if disease or other investigations are required, this method is too destructive and renders the carcass unsuitable for post mortem examination. In these cases, a chemical means of euthanasia needs to be applied. When euthanasia is necessary for disease investigations the method employed needs to be appropriate to the investigation being performed, especially for histopathology requirements to help investigate health issues. It is also important to consider keeping intact shells and genetic material for species identification purposes (see Appendix for post mortem protocols).

Institutions participating in the programme are asked to carry out post mortem examinations on recently deceased or dying specimens suspected of illness, so the disease status of the populations can be monitored (see section 2.6 and appendix 1). The current method of euthanasia prior to PM examination is exposure to an overdose of isofluorane. A cotton wool ball soaked in the anaesthetic agent is placed in the snails' container (ideally a small container in which the snails are submitted) and left for a minimum of one hour; or longer if there are any signs of life. If post mortem examination cannot be carried out, the bodies should be stored for future reference in ethanol (ideally 96% Analar A) at room temperature, at a volume ratio of at least one-part snail to ten parts ethanol, which will preserve the tissue, shell and DNA, but not for bacteriological, parasitological or histopathological testing. Care should be taken to ensure the correct records are associated with the specimen, including taxon, development stage, container reference and date of death. The data is best written in pencil on a piece of paper inserted into the alcohol. Usually it is mainly adults that are stored in this way. The method employed for excess stock euthanasia if necessary, is immersion into as near 100% ethanol as is possible. Immersing the snails in > 90% ethanol kills them quickly and so appears to be humane.



Fresh dead specimen stored and labelled in alcohol

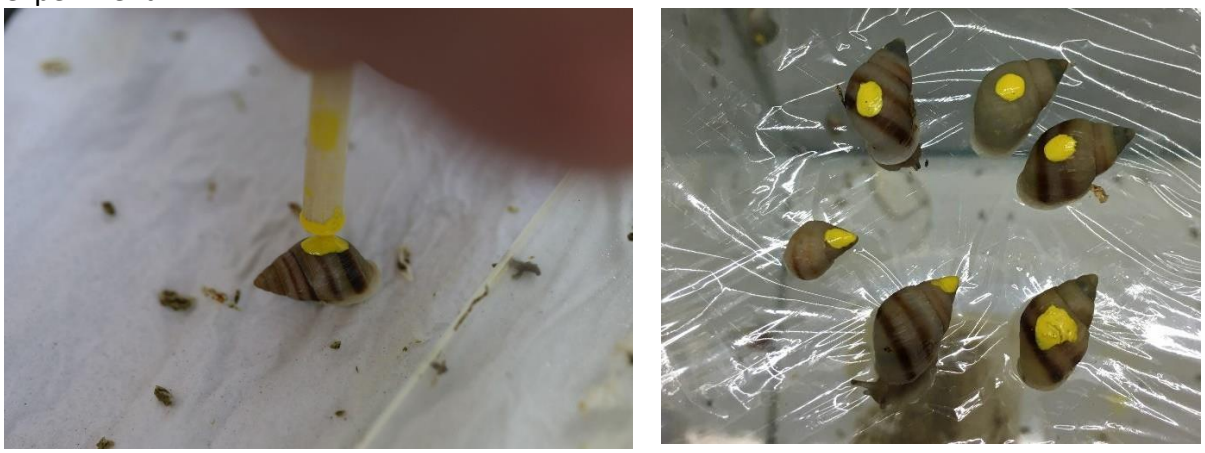
2.5 Behavioural enrichment

No direct behavioural enrichment needs are documented for partulids, or indeed any terrestrial molluscs. However, providing suitable enclosure space whilst ensuring survivorship and keeping the snails in groups allowing social interaction, hopefully satisfies their needs. This is another area for potential research.

2.6 Handling

2.6.1 Individual Identification

Group-based management needs necessitate tracking at the life stage level rather than at the individual level. However, if required several marking options are possible. Individual marking methods used to date have included indelible ink marks on the shell, and most effectively for wild releases in recent years, water-based enamel paint (by Plastikote). Different coloured paints have been used for each year of releases, adults marked with a spot on the whorl, and young stages on the tip of the spire, Other non-individual marking methods have included the attachment of beta lights for night tracking, as used in the Kew experiment.



Marking snails with yellow enamel paint pre-release 2016 (Dave Clarke/ZSL)

A technical review of marking options has been produced, with a priority on the snails welfare considerations (Gerlach, unpublished report 2017).

From an early stage in the breeding programme identification of development stages has been important, with snails being allocated to four developmental stages through their life, as detailed in section 1.7.2.)

2.6.2 General Handling and restraint

Care must be taken when handling the snails to prevent shell damage, especially the young stages where the shell is not fully formed, especially at the growing edge. Most adult *Partula* are remarkably tough once the shell is fully developed, and the thickened lip formed at the shell opening. Care must always be taken to avoid rupturing the delicate foot and body when removing any snail from any surface it is adhered to. Snails should be gently slid off surfaces

rather than pulled directly, using extra moisture if required. They tend to retract into the shell when disturbed but can become active very quickly afterwards.

Although gloves can be used, they can inhibit safe manipulation of small snails. Therefore, bare hands should be washed with an anti-bacterial soap (Hibiscrub is commonly used) and thoroughly rinsed afterwards before commencing working with the snails. This exercise should also be repeated after dealing with any dead snails and at the end of the working session. A desirable additional hygiene measure is to wear dedicated laboratory coats when working in the *Partula* room, particularly if keepers are working with other similar species.

If any escapees are found outside of the enclosures, it is vital that they only get returned if it is completely clear which container they came from (e.g. if a split in the clingfilm is identified. This is particularly to prevent mixing of species, confusing count records, and also as a protection against cross-contamination risks.

Snails have a very delicate soft body, therefore it is important not to put the snails on surfaces which may have chemicals present. For this reason, it is recommended that if snails are temporarily removed from tanks they are placed on a piece of tissue or box rather than directly on a worktop.

2.6.3 Transportation

Other than very brief journeys (e.g. within a zoo grounds) partulids are best transported in a temporary aestivation-like state. This is achieved by wrapping small numbers of snails in several layers of tissue to both protect the snails and encourage them to rest, then putting them in a rigid container with minimal ventilation, and outer insulated carry box.

If being shipped by air or courier it is necessary to ensure all relevant legislation is adhered to e.g. the 2000, IATA 'Live Animal Regulations; 27th Ed.'. Full detailed guidance on international shipping has been developed following a commissioned review of transport options for the reintroduction programme. The detailed and approved protocol is published here in Appendix

Dry wrapped snails in tissue and cardboard tube, being placed in an insulated travel crate

For new arrivals it is considered essential that they undergo a quarantine isolation period of at least 30 days with some disease/faecal screening, ideally in a separate room, before being mixed with resident stock.

2.6.4 Safety

Outside of the wildest of possible human/snail interactive scenarios the only practical safety consideration to highlight is the need for due care in the area of ensuring basic hygiene (washing of hands) before and after working with the animals. There have been no reported incidence of zoonoses or allergic reactions arising from working with partulids but on the principle that almost any micro-organism are potentially capable of being transferred to humans (as with any animal). It is worth stressing that this basic hygiene protocol is equally important for the continued wellbeing of the snails. Regular health screening of snail faecal samples and whole post mortems are always desirable for maintaining relatively up to date micro-organism profiles (again of equal value to snails and human health considerations).

2.7 Veterinary: Considerations for health and welfare

Problems have been encountered in establishing viable populations of some species after initial collection from the wild, but those that have successfully established do not appear to have suffered from any identifiable diseases. Population declines are thought to be linked to environmental factors as no infectious disease agents have been implicated. Flagellated and ciliated protozoa, nematode larvae and a variety of bacteria are routinely found in faecal samples, both from wild-caught and captive *Partula* (Cunningham et al 1996) and are thought to be normal intestinal flora. They may be noted within tissues in histological sections of dead snails, but without cellular reaction - indicating *post mortem* invasion. Microsporidia may also be noted histologically and were linked to the deaths of the last surviving *Partula turgida* (Cunningham and Daszak 1998). However, they are commonly seen in dead and culled captive snails without any associated cellular changes, and have also been found in wild-caught, museum-held specimens (M.Stidworthy, personal communication), indicating that they are most likely commensal organisms. The same is probably true of *Cryptosporidium*-like organisms that have been detected as 4-5 micron diameter, acid-fast staining cysts in faeces and apex impression smears of captive *Partula* for many years. With a change from fixation of snails *post mortem* in 70% ethanol to 10% buffered formalin, these have also been detected in many dead and culled snails without associated cellular lesions. Undoubtedly some infectious causes of ill-health and death will be discovered in the future, so it is vital that any suspicious deaths be investigated thoroughly, and the agreed *Partula* EEP protocol for pre-screening of snails for reintroduction to the wild (Appendix 2) be undertaken.

The snails do seem particularly sensitive to change. Therefore, any significant adjustments to the keeping method need to be carefully considered and discussed with the EAZA Committee before any changes are made.

2.8 Specific problems

Some snails can have algae growing on the shells in captivity. This is not completely unknown but rarely seen in wild snails. This in itself may not be too harmful but can contribute to poor shell condition from affecting the growing edge of the shell (Dave Clarke pers comm).

Green algae growing on *Partula taeniata nucleola* shells in captivity (Dave Clarke/ZSL)

Phorid or 'scuttle' flies (Diptera, Phoridae) can sometimes be an issue in *Partula* containers. Usually they are linked to build up in soil from other species held in other parts of the collection, which make their way into the room where the snails are kept, as otherwise frequent substrate changes remove their ability to breed. They can enter tanks through the smallest of holes, for example the perforations on clingfilm. They can be a problem as they are attracted to the diet and to ill/dead snails, where maggots can develop within only a few days. They can be controlled by minimising ability to enter the room and containers, and by using yellow sticky traps or UV flytraps (although they do not seem particularly attracted to these).

2.9 Recommended research

Investigations are currently being carried out into:

- Respective keeping systems success factors
- Diet improvement including calcium type/levels
- Size variation between successive generations
- Genetic relatedness within and between sub-populations
- Reproductive behaviour
- Post-reintroduction behaviours and survival
- Invasive species threat assessments and management
- ZIMS studbook group-based management

Other potential areas for research include:

- Behavioural enrichment

Section 3

References & bibliography

Bloxham Q M C, Tonge S J. and Horton R (1984) A preliminary account of the history of the management of populations of endangered tree snail *Partula* sp at Jersey Wildlife Preservation Trust. Dodo (Journal of the Jersey Wildlife Preservation Trust) 20:73-
Bick, C. S., Pearce-Kelly, P., Coote, T. and Foighil, O'D (2018). Survival among critically endangered partulid tree snails is correlated with higher clutch sizes in the wild and higher reproductive rates in captivity. Biological Journal of the Linnean Society 125(3):508-52
Burch J B (2004). Endangered Tahitian snails found in Michigan freezer. Nature 428: 687
Clarke B C, Johnson M S and Murray J J (1996). Clines in the genetic distance between two species of island land snails: how molecular leakage can mislead us about speciation. Phil. Trans. R. Soc. Lond. B 351: 773-784.
Clarke B C, Johnson M S and Murray J J (1998). How 'molecular leakage' can mislead us about island speciation. In: Evolution on Islands (ed. P R Grant), OUP.
Coote, T + 27 others (2019). Fourth year of *Partula* species reintroductions into natural habitat on Tahiti and Moorea Tentacle 27: 35-38
Coote, T + 27 others (2018) Evolving strategy for release of *Partula* species back onto their islands of origin Tentacle 26: 16-19
Coote, T (2012). Tahiti becomes the focus of Partula conservation. Tentacle 20: 28-30.
Coote, T (2007). Partulids on Tahiti : differential persistence of a minority of endemic taxa among relict populations. Amer. Malac. Bull. 22: 83-87.
Coote, T, Dave Clarke, Carole S Hickman, James Murray, and Paul Pearce-Kelly (2004). Experimental release of endemic Partula species, extinct in the wild, into a protected area of natural habitat on Moorea. Pacific Science 58(3): 429-434
Coote T (1998). Problems with the barrier method: Moorean snail reserve update. Vet. Invert. Soc. Newsletter 14: 9-11.
Coote T and Loeve E (2002). From 61 species to 5: endemic tree snails of the Society Islands, French Polynesia fall prey to an ill-judged biological control programme. Oryx 37(1): 91-96
Coote T, Loeve E, Meyer, J-Y & Clarke D (1999). Extant populations of endemic partulids on Tahiti. Oryx 33(3): 215-222.
Cowie R H (1992). Evolution and extinction of Partulidae, endemic Pacific island land snails. Phil. Trans. R. Soc. Lond. B 335: 167-191.
Cowie R H. (1996). Pacific Island Land Snails: Relationship, origins and determinants of diversity. In: The Origin and Evolution of Pacific Island biotas, New Guinea to Eastern Polynesia: Patterns and Processes.Ed. A.Keast & S.E.Miller, Academic Publishing, Amsterdam: 347-372.
Cowie R H. (1998). Catalog of the Nonmarine Snails and Slugs of the Samoan Islands. Bernice P.Bishop Mus. Bull.in Zool. 3:73-74.
Crampton H E (1916). Studies on the variation, distribution and evolution of the genus *Partula*. The species inhabiting Tahiti. Carnegie Inst. Wash. Pub. 228: 1-311.
Crampton H E (1932). Studies on the variation, distribution and evolution of the genus *Partula*. The species inhabiting Moorea. Carnegie Inst. Wash. Pub. 420: 1-335.
Cunningham A A and Daszak P (1998). Extinction of a species of land snail due to infection with a Microsporidian parasite. Conservation Biology 12(5):1139-1141.

Cunningham A A, Daszak P, Macgregor S K, Foster I, Clarke D and Pearce-Kelly P (1996). Mortality of endangered snails of the genus Partula:Preliminary results of pathologic investigations. J. Zoo and Wildlife Medicine 27(1): 19-27.
Gerlach, Justin(2016) *Partula compressa*: not extinct after all. Tentacle 24: 49
Gerlach J (2016). Icons of Evolution: Pacific Island tree snails: family Partulidae. Phelsuma Press, Cambridge, UK.
Goodacre S L (2001). Genetic variation in a Pacific Island land snail: Population history versus current drift and selection. Proceedings of the Royal Society of London - Series B: Biological Sciences. 268(1463): 121-126.
Goodacre S L (2002). Population structure, history and gene flow in a group of closely related land snails: Genetic variation in Partula from the Society Islands of the Pacific. Molecular Ecology 11(1): 55–58.
Goodacre S L and Wade C M (2001). Molecular evolutionary relationships between partulid land snails of the Pacific. Proceedings of the Royal Society of London - Series B: Biological Sciences. 268(1462): 1-7.
Goodacre, S. L Wade, C. M. (2001). Patterns of genetic variation in Pacific island land snails: the distribution of cytochrome b lineages samong Society Island Partula Biological Journal-Linnean Society, vol. 73, no. 1, pp. 131-138
Gouveia A R, Paul Pearce-Kelly, Donald L. J. Quicke & Simon R. Leather (2011) Effects of Different Calcium Concentrations Supplemented on the Diet of *Partula gibba* on their Morphometric Growth Parameters, Weight and Reproduction Success. Malacologia, 54(1–2):139-146. 2011.
Hickman C S (1999). Origin, evolutionary radiation, and extinction in the modern world: the story of partulid tree snails. In: Evolution: Investigating the Evidence (J Scotchmoor and D A Springer, eds.) Paleontological Society Special Edition 9.
Johnson M S, Murray J. and Clarke B C (2000). Parallel evolution in Marquesan partulid land snails. Biological Journal of the Linnean Society 69: 577-598
Johnson M S, J Murray, and B C Clarke. (1993). The ecological genetics and adaptive radiation of *Partula* on Moorea. Oxford Studies In Evolutionary Biology: 167-238.
Kondo Y (1968). Partulidae: Preview of Anatomical revision. Nautilus 81 (3): 73-77.
Lee, T., Burch, J., **Coote, T**., Fontaine, B., Gargominy, O., Pearce-Kelly, P., and Ó Foighil, D. (2007). Prehistoric Inter-Archipelago Trading of Polynesian Tree Snails Leaves a Conservation Legacy. Proc. R. Soc. B. **274**: 2907.2914
Lee, T., Burch, J.B., Jung, Y., **Coote, T**., Pearce-Kelly, P., and Ó Foighil, D. (2007). Tahitian tree snail mitochondrial clades survived recent mass-extirpation. Current Biology **17**: No 13, R502-R503.
Lipton C S and Murray J J (1979). Courtship of land snails of the genus *Partula*. Malacologia 19: 129-146. Lipton & Murray 1979?
Mace G. Pearce-Kelly P. Clarke D. (1998) An integrated conservation programme for the tree snail of Polynesia (Partulidae): A review of captive and wild elements. J. Conch. Sp. Pub (2): 89 – 96.
Murray J and Clarke B C (1984). Movement and gene flow in *Partula taeniata*. Malacologia 25(2): 343-348.
Murray J and Clarke B C (1976). Supergenes in polymorphic land snails I. *Partula taeniata*. Heredity 37(2): 253-269.
Murray J, Johnson M S and Clarke B C (1982). Microhabitat differences among genetically similar species of *Partula*. Evolution 36(2): 316-325.

Nichols R A and G M Hewitt (1994). The genetic consequences of long-distance dispersal during colonization. Heredity 72(3): 312-317.
Owczarzak R L (2001). Fitzroy Dearborn, US Snail, Partula. In: Encyclopedia of the World's Zoos, R-Z (ed. C E Bell)
Parrish R, Sherley G and Aviss M (1995). Giant Land Snail Recovery Plan, *Placostylus* spp., *Paryphanta* sp. Threatened Species Recovery Plan Series No 13, Department of Conservation (*Te Papa Atawhai*), New Zealand.
Pearce-Kelly P, Clarke D, Walker C. and Atkin P. (1997). A conservation programme for the partulid tree snails of the Pacific region. Memoirs of the Museum of Victoria 56(2): 431-433.
Pearce-Kelly P, Mace G and Clarke D (1995). The release of captive bred snails (*Partula taeniata*) into a semi-natural environment. Biodiv. Cons. 4:645-663.
Tonge S. and Bloxham Q. (1991) A review of the captive breeding programme for Polynesian tree snails (*Partula* spp). International Zoo Yearbook. 30: 51 – 59.
Pearce-Kelly et al (2019). Fourth year of Partula species reintroductions into natural habitat on Tahiti and Moorea. Tentacle No. 27

Unpublished reports

Clarke D, Coote T, Murray J, Spencer W, Cracknell J. (1995). PILSG 1995 Expedition to French Polynesia - Report. ZSL, Regent's Park, London, NW1.
Coote T. (2004). Stratégie 2003-2005 pour la conservation et la gestion des escargots terrestres endémiques de Polynésie française et de leur habitat : rapport des prospections à Tahiti 2004. Direction de l'Environnement de la Polynésie Française et la Societé Zoologique de Londres (en français et anglais).
Coote T. (2005). Stratégie 2003-2005 pour la conservation et la gestion des escargots terrestres endémiques de Polynésie française et de leur habitat : rapport des prospections 2005. Direction de l'Environnement de la Polynésie Française et la Societé Zoologique de Londres (en français et anglais).
Coote T. (2005) Conservation Action Plan for the long term protection of French Polynesia's last surviving populations of endemic tree snails of the genera Partula, Samoana and Trochomorpha.
Coote T (1999). The genetics and conservation of Polynesian tree snails (family Partulidae) PhD thesis, University of London.
Coote T (2001). An urgent briefing report for the French Polynesian Government, associated agencies and the IUCN on the conservation status of the endemic tree snails (Partulidae) of French Polynesia. ZSL, Regents Park, London, NW1
Coote T (2001). Revised Red List categories for Partulidae (submitted to IUCN).
Coote T (2002). Partula: a lesson still being learnt. Antenna, newsletter of the London Zoo volunteers.
Coote T Clarke D., Hickman C.S., Murray J.J., Pearce-Kelly P. (1999). Experimental Release - Afareaito Valley, Moorea draft: 1-9 (Final Report)
Davis, Elizabeth C (2005). Predation by *Euglandina rosea* on local versus non-local gastropods: no difference in mucus trail following? Powerpoint presentation, 71st Annual Meeting, American Malacological Society, Asilomar Conference Centre.
Fontaine B and Gargominy O (2002). Rapport pour le Délégation à la Recherche du Gouvernement de Polynésie française, le Muséum national d'Histoire naturelle, et le Comité

français pour l'UICN. Inventaire des mollusques continentaux de l'archipel des Australes (Raivavae, Rapa).
Fontaine B and Gargominy O (2004). Rapport pour le Délégation à la Recherche du Gouvernement de Polynésie française, le Muséum national d'Histoire naturelle, et le Comité français pour l'UICN. Inventaire des mollusques continentaux de l'archipel des Australes (Rimatara)
Gargominy O and Fontaîne B (2003). Rapport pour le Délégation à la Recherche du Gouvernement de Polynésie française, le Muséum national d'Histoire naturelle, et le Comité français pour l'UICN. Inventaire des mollusques continentaux de l'archipel des Australes (Rurutu, Tubuai).
Gerlach, J (2014) – Diet of the Partula species of Moorea
Gerlach, J – (2017) Review of snail marking methods for ZSL
Goodacre S L (1999). Studies on the evolution of Partula. PhD Thesis, University of Nottingham,
Gouveia A R (2011) Investigation of the factors affecting the population dynamics of captive Partula snails. PhD thesis. Imperial College, London.
Hickman C.S., Hadfield M.G. (1998). Conservation of endemic arboreal gastropods on Moorea and Tahiti, French Polynesia. Report of Field Excursion 25-31 Oct 1998: 1-6
Hulbert, A., Flach, E., Jayson, S. and Masters, N. Pre-release Health Screening of Partula species: June-September 2017 (ZSL)
Loève E. (1983). Contribution à l'étude des Gastropodes Terrestres de Polynésie Française. Ecologie et Biogéographie. Mémoire de DEA, USTL, Montpellier
Meyer, Jean-Yves, Jean-Claude Thibault, Jean-François Butaud, Jacques Florence, Trevor Coote, Ronald Englund (2005). Sites de conservation importants et prioritaires en Polynésie française. Contribution à la Biodiversité de Polynésie française N°13 Sites Naturels d'Intérêt Ecologique V.
Meyer, J.-Y. & Florence, J. (1998). Exploration du Mont Mauru (Tahiti, Hitia'a). Contribution à la Biodiversité de Polynésie française N°6 : Sites Naturels d'Intérêt Ecologique I. *Délégation à la Recherche*, Papeete, 20 pages.
Meyer, J.-Y. & Jay, H. (2000). Prospection des haut-plateaux de Faufiru ou Fare Aro (Tahiti, Mahina). Contribution à la Biodiversité de Polynésie française N°7 : Sites Naturels d'Intérêt Ecologique II. *Délégation à la Recherche*, Papeete, 26 pages.
Murray J. (1995). Conservation of Pacific land snails: Monitoring and rescue of endangered species. A preliminary report to Flora and Fauna International. Cambridge, U.K.
Pearce-Kelly, P., Clarke, D. and Mace, G. (1994). *Partula* '94: An action plan for the conservation of the family Partulidae, by the Pacific Island Land Snail Group. Zoological Society of London, Regent's Park, London NW1 4RY.
Pearce-Kelly, P., Clarke, D. and Mace G. (1995). Partula '95: Proceedings of the annual meeting of the Pacific Island Land Snail Group. Zoological Society of London, Regent's Park, London NW1 4RY.
Pearce-Kelly P., Clarke D., Mace G. (1997). *Partula* '97: Proceedings of the annual meeting of the Pacific Island Land Snail Group. Zoological Society of London, Regent's Park, London NW1 4RY.
Polytechnic of North London, Food Analysis Unit, Chemical analysis of 'snail food', 1991.
BIAZA - Invertebrate Euthanasia Guidelines 2018.

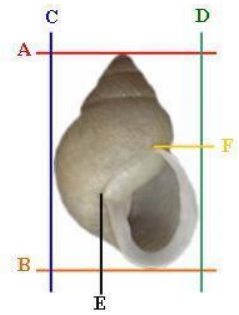
Appendices –

1. **Post mortem protocol for Polynesian Tree Snails**
2. **Pre-reintroduction health screening protocol for *Partula snail* species**
3. **Partula snail transportation guidelines**
4. **Typical detailed servicing regime (as at ZSL London Zoo)**
5. **Products mentioned in the text and currently in use at institutions**
6. **Chronology of significant partulid events**

Appendix 1 - Post mortem protocols

Post mortem protocols for Polynesian tree snails (*Partula sp. and Samoana* sp.)
Updated by Edmund Flach, January 2019

- Set up for necropsy
- Weigh the snail
- Take measurements of the shell as illustrated on the diagram below left – this is the standardised land snail measuring protocol (an important consideration when attempting to compare historic in situ and ex situ measurements). The use of electronic calipers is recommended.
    - Greatest length including the lip: line AB
    - Greatest width including the lip: line CD
    - Aperture length: point FB
    - Aperture width: point ED



Dextral chirality Sinistral chirality

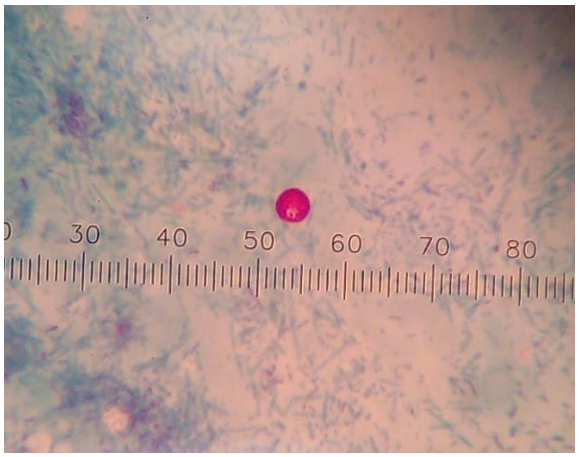
- Note whether the shell coils to the right or to the left (see above photo) and whether or not the aperture is lipped (evidence of sexual maturity)
- Remove the snail from the shell, either by gentle steady traction or by chipping open the shell using bone forceps.
- Identify grossly visible anatomical structures including the apex, foot, mantle, tentacles, digestive gland and lung if present.
- Examine the snail for granulomas, fungal infections or other obvious external abnormalities.
- In snails too small for gross recognition of most internal lesions avoid internal dissection, but take a bacteriological swab from the internal tissues.

- If fresh faeces are present, examine a direct faecal wet mount in physiological saline for parasites.

- Amputate the apex using a sterile scalpel blade.

- Make two or more touch impressions of the apex and apply modified Ziehl-Neelsen stain to one smear. An additional Gram's stain may also be used on a second smear, but is less diagnostic.

- Examine the cytology preparations, in particular searching for protozoan parasites such as Cryptosporidium-like cysts (4-5 microns in diameter) or Steinhausia species and other microsporidian spores (1.5-2 microns diameter). The cysts and spores appear dark purple with Gram's stain and bright red with modified Ziehl-Neelsen stain.

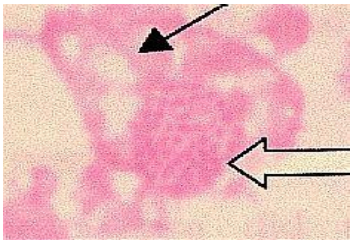


Modified ZN acid-fast cyst in apex impression smear, x100 magnification (ZSL)

- Freeze a portion of the foot for the DNA bank.

- Save the carcass including the amputated apex in 10% buffered formalin. 70% ethanol can be used, but the subsequent histopathological findings are likely to be less diagnostic.

- Submit formalin-fixed carcasses for routine histological processing and staining of the resulting sections with haematoxylin and eosin (H&E), Ziehl-Neelsen (ZN), Gram's and Luna-Peterson (L-P) stain. The stained sections should be examined by an histopathologist experienced in the pathology of molluscs.


Histology preparation, HE stain. Small arrow: vacuoles in degenerating digestive gland cell of partulid snail, large arrow: encysted *Steinhausia* sp. containing numerous spores. (Photomicrograph by L. D. Espinosa Aviles.)

- Further references for more detailed information on diseases of partulid snails:

    - Cunningham AA and Daszak P. 1998.Extinction of a Species of Land Snail Due to Infections with a Microsporidian Parasite, Cons. Biol. 12 (5): 1139 – 1141.

    - Cunningham AA *et al*. 1996.Mortality of Endangered Snails of the Genus *Partula*: Preliminary Results of Pathologic Investigations, J. Zoo. Wild. Med. 27 (1): 19-27.

Appendix 2 – Pre-reintroduction health screening protocol

Pre-reintroduction health screening protocol for *Partula snail* species

Edmund Flach, Zoo and Wildlife Pathologist, Zoological Society of London, Regent's Park, London NW14RY.

Christophe Giraud, Vétérinaire Officiel, Le Département de la Qualité Alimentaire et de l'Action Vétérinaire (DQAAV), BP 100 - 98713 Papeete, Tahiti.

Miri Tatarata, Department Chief, Service Direction de l'Environnement, BP 4562 Papeete, Tahiti.

Paul Pearce-Kelly, EEP and International *Partula* snail Programme Coordinator

Reviewed and endorsed by Partula EEP Committee February 2019

Introduction

This health screening protocol is intended to help ensure that all *ex situ Partula* snail populations have the greatest chance of inclusion in the reintroduction phase of the *Partula* conservation programme and ensure compliance with the *Partula* reintroduction disease risk analysis (Dalziel *et al*, 2013) to minimise the risk of infectious and non-infectious diseases compromising reintroduction success. The protocol draws upon and synthesises best health-screening practice employed by the *Partula* programme and other invertebrate programmes.

Background

Intensive health-screening carried out at ZSL (Goodey & Flach, 2015 and 2016) in anticipation of the pre-release export involved monitoring and testing for the presence of potentially significant pathogens and pathogenic lesions. The screening consisted of examination of fresh faeces, individuals found dead, and individuals euthanized, for the presence of parasites and bacteria, plus histopathological examination of snail sections for evidence of micro-organisms and pathological processes. Although a variety of parasites and bacteria were observed and cultured, none were considered significant. There was evidence of a suspect microsporidian infection observed commonly in the past and characterised by acid-fast cysts with a diameter of between 4 and 5 microns in the intestines (observed in impression smears of the apex) and *faeces*. Definite microsporidian bodies were identified histologically and were suspected to be *Steinhausia* species, but there was no cell damage, reaction, or inflammation associated with these bodies. Similar bodies were detected in museum-derived, wild-caught individuals, again without evidence of pathogenicity. It was concluded that the health of the snails was good, and that they were free of any pathogens likely to cause disease in them, or to native French Polynesian snails, and other species, after release.

Pre-release Screening

The current screening protocol is based closely on the 2015 and 2016 recommendations, but with three main changes: a) closer monitoring of population numbers with early warning to the veterinarian(s) in charge of any deaths, b) reduced bacteriological culturing from faecal samples; just one faecal bacteriological examination is proposed, and c) snails to be fixed in 10% buffered formalin because of the successful identification of *Cryptosporidium*-like bodies in *Partula* snails that had been so fixed (Stidworthy & Lopez, pers. comm.).

Tanks of snails from the resident collection may be monitored and sampled *in situ*, but any populations imported from other collections for pre-export screening should be introduced into a separate, bio-secure room and serviced by staff that do not work with the resident snails. If this is not possible, staff dealing with the resident collection should service this first, and then put on laboratory coats and change shoes before working on the quarantined populations. It is acceptable, and makes final certification of health easier, if tanks from the resident collection intended for export are also moved into this room so that all populations for export are managed and screened together and at the same time.

During the period of quarantine (suggested to be a minimum of one month) the following surveillance should be done:

1. Monitoring of mortality
Each tank population should be checked each morning and all dead snails submitted immediately to the veterinary department. In addition, there should be counts, at least weekly, of numbers of newborn, juvenile, sub-adult and adult snails. Abnormally high numbers of deaths in a tank should be reported to, and discussed with, the veterinarian in charge and the population withheld from the export, unless the deaths can be proved to be due to an environmental factor (e.g. temperature fluctuation, abnormal humidity, exposure to toxic chemicals on food plates).

2. Faecal screening: A minimum of three faecal samples taken from each tank for parasitological testing, one of which should also undergo bacteriological testing. Faeces should be collected into sterile plastic bijou containers (or similar), labelled with the tank number, species and date, and submitted to the diagnostic laboratory.

3. *Post mortem* examinations: Dead snails should be examined in three circumstances: a) individuals found dead, reasonably fresh and on a day when they could be submitted for immediate examination, b) individuals found dead, but not fresh, or on a day when immediate examination is not possible; these to be fixed in 70% ethanol and then submitted, and c) healthy individuals randomly selected for euthanasia (carried out by the collection's veterinary department and using exposure to an overdose of the volatile anaesthetic drug isoflurane for a minimum of one hour) and submitted for immediate, fresh examination. The aim is to examine approximately 5-10% of the population of adults in each tank, with at least one individual examined fresh after euthanasia from tanks with large enough populations.
As is the case with many other invertebrate species reintroduction programmes, the need to include euthanised specimens in the pre-release health screening process is due to a combination of: a) high prevalence of severe autolysis of dead snails, and b) the need to screen sufficient individuals in order to have confidence of the results of the tests. Unfortunately, despite many years of examining snails there are still no definitive prevalence rates for clinical disease in *Partula* snails due to microbial pathogens. Most are present in low numbers and are detected intermittently, so even when found in populations undergoing unusually high mortality their contribution to the mortality (compared to environmental factors) is unknown.

Snails presented already fixed in ethanol are weighed and measured, but not examined grossly. A proportion may be submitted for histopathology, but the diagnostic value of these individuals is usually very low due to autolytic changes and fixation in ethanol. All others are

weighed and measured as previously described (Pearce-Kelly et al, 2007). If the snail does not easily slide from the shell when traction is applied it is recommended that heavy-duty scissors are used to open the shell to ensure minimal crush and stretching artefacts to the carcass. The extracted body is weighed, and examined externally. The body opened with a sterile scalpel blade, an internal ("coelom") swab is taken for bacteriological culture, and the internal tissues are examined grossly. The tip of the apex is then removed with the scalpel blade and impression smears of the cut surface made on a microscope slide for parasitological examination (see below). A small piece of foot tissue is removed and frozen as a source of DNA, and finally the remaining carcass, plus the amputated apex, are fixed in 10% buffered formalin, placed in histological cassettes and forwarded to the diagnostic laboratory performing histopathological services. Here at ZSL snails are sent to the Royal Veterinary College (RVC) for processing (including the following stains: haematoxylin and eosin (H&E), Ziehl-Neelsen (ZN), Gram's, Periodic acid-Schiff (PAS) and Luna-Peterson (L-P) specifically for microsporidia (Peterson *et al*, 2011)) and thence to the International Zoo Veterinary Group (IZVG) for histopathological assessment and reporting.

4. Diagnostic testing

4.1 Parasitology

Faeces are examined by direct microscopy of a wet preparation and microscopy of a dry smear stained with modified Ziehl-Neelsen stain (MZN). Apex smears are also examined microscopically after staining with MZN.

1. Wet preparations are prepared by mixing a small amount of faeces (a bacteriological Nichrome wire loop-full) in two drops of sterile physiological saline solution and examined microscopically for the presence of any parasites or their ova, with particular emphasis on: helminths, flagellated protozoa and ciliated protozoa.

2. MZN-stained smears are prepared and examined microscopically under oil immersion for the presence of pink-staining (acid-fast) bacilli (possible *Mycobacterium* species) and cysts (4-5microns diameter; suspect *Cryptosporidium*-like, 1-2microns; suspect microsporidian). Cyst diameter should be measured with an eye-piece graticule that has been calibrated from a micrometer stage slide, or by electronic means.

4.2 Bacteriology

Faecal samples and "coelom" swabs are normally cultured on 5% horse blood agar plates and incubated at 25°c for 48 hours. Plates are examined and, if the culture is mixed, single bacteria colonies of predominant types are sub-cultured onto further plates. The resulting purecultures are then identified by standard methods: colony morphology, appearance and Gram'sstaining characteristics, and biochemical reactions (using commercial analytical profile index (API) biochemical test kits). The diagnostic laboratory used for testing should be consulted prior to submission of samples.

4.3 Histopathology

Histopathology reports (external or in-house) should include details of all of the tissues seen in the sections, any cellular changes and the presence of any micro-organisms and interpretation of their significance.

5. Interpretation of results

Results should be recorded in appropriate spreadsheets and reviewed regularly. Examples are given in Appendices 1-4.

It is likely that a large number of micro-organisms will be detected during the screening, but in our experience the majority are of no, or doubtful, significance to the snails' health. Flagellated protozoa are commonly found in the faeces of *Partula* snails both in captivity and the wild (Cunningham *et al*, 1996) and are likely to act symbiotically in breaking down cellulose and other plant fibres in the snails' diet. Rhabditid nematodes were also found in the faeces of wild-caught snails, so the presence of nematodes is generally accepted as a normal finding, although they have the capacity to invade snail tissues after death and therefore are potential opportunistic pathogens.

A wide range of bacteria were isolated from the faeces of wild and captive snails by Cunningham *et al* (1996). This list included *Flavobacterium breve* which is now *Empedobacter brevis* and was identified in this study, albeit undifferentiated from *Weeksella virosa*. *Myroides* species are also closely related to *Flavobacterium* and one of them, *M. odoratimimus* has been reclassified as *F. odoratum*. These, plus *Aeromonas hydrophila*, *Brevundimonas vesicularis* (formerly *Pseudomonas vesicularis*), *Bacillus* species and *Pantoea* species are all common environmental bacteria and therefore likely to form part of the normal flora of plant-eating snails. *Flavobacterium*-related bacterial species were much less commonly predominant in cultures from dead and euthanized snails, as also noted by Cunningham *et al* (1996), but *Aeromonas hydrophila/caviae* was predominant on several occasions.

As previously stated, cysts that stain positively in MZN (and ZN) stains are commonly found in faecal and apex smears; most often with a diameter of 4-5microns and assumed to be the *Cryptosporidium*-like protozoa that have recently been identified histologically and by electron microscopy. These have been observed repeatedly in the past, but have often been referred to under different names, including protozoal cysts, protozoal oocysts, protists and microsporidian cysts. They can be found commonly, but intermittently, in captivity (ZSL veterinary records and reports) generally without association with disease or increased mortality. However, increased mortality in populations of *P. gibba* and *P. tohiveana* at ZSL in 2006-8 was linked to an apparent increase in the prevalence of cysts in faeces and apex smears, and also the presence of microsporidian bodies in histological sections of snails testing positive, or from tanks containing positives (Flach *et al*, 2008). These bodies were similar to the microsporidia described by Cunningham and Daszak (1998), but the *Steinhausia* cysts described in the paper (2 micron diameter) were not identified, and there was no evidence of a cellular, pathological reaction to their presence in the cases seen in 2006-8. In addition, the use of ethanol fixation at the time (and up until this year) was not ideal for sectioning of *Partula* tissues and the *Cryptosporidium*-like bodies were never identified. In the 2015 health screening it was observed that cysts appeared in the first faecal samples from several tanks, the second faecal sample from a lower number, and were then absent in third and subsequent samples. This suggests that they may be excreted during times of stress, such as transportation, and acclimatisation to a new environment (the quarantine room), and raises the possibility that other stresses causing increased mortality might at the same time lead to increased shedding and therefore detection of the cysts. Gouveia (2011) investigated many

factors that affected the well-being of captive *Partula* species and identified different species sensitivities to a number of environmental factors, especially temperature, humidity and diet.

Microsporidian bodies were frequently seen in histological sections, mainly in digestiveglands, of snails during the 2015 and 2016 health screening, but with no associated cellular lesions. Interestingly, and tellingly, similar microsporidian bodies were found in wild caught, museum-archived individuals of two of the three species screened for export in that year (*P.hyalina* and P.*nodosa*); again with no evidence of any pathological reaction associated with them. Microsporidia have been recorded in the literature to infect all vertebrates and most invertebrates (Keeling and Fast, 2002), and Weiser (1976) even estimates there to be microsporidia in every living invertebrate. Therefore, the vast majority of infections are sub-clinical, but because of the occurrence of microsporidia belonging to the genus *Steinhausia* in some of the last individuals of *Partula turgida* that died at London Zoo in 1996 (Cunningham and Daszak, 1998) they have always been considered pathogenic in *Partula* and potential causes of death. The histopathological evaluation of fresh snails, and lack of cellular response to any microsporidian bodies seen, is the best indicator that the source population is unaffected by them.

6. Certification

The veterinarian in charge will have to be confident that he/she can sign the health certificate issued by the exporting country's official animal-health service which, in turn, will be based on the requirements of the importing country (French Polynesia). Discussions should be heldat an early stage to ensure that extra testing may be added, if requested, and additional requirements met (e.g. use of sterile substrate prior to and during transportation).

Follow-up

It would be helpful if the results of health screening be shared with all collections holding *Partula* species, so that we can increase our knowledge of normal and potentially significant micro-organisms. Also, details of mortality during transportation, during post import quarantine and acclimatisation, and in the post release monitoring period, should be recorded and shared. Whenever possible, any freshly dead snails found during this period should be fixed in 10% buffered formalin and examined histopathologically (postage back to Europe for examination by the same histopathologist(s) that did the pre-export screening is preferable).

References

Coote, T. Clarke, D. Hickman, C.S. Murray, J. & Pearce-Kelly, P., 2004. Experimental Release of Endemic *Partula* Species Extinct in the Wild, into a Protected Area of Natural Habitat on Moorea. *Pacific Science.* 58(3) pp 429-434.

Cunningham, A.A. Daszak, P. Macgregor, S.K. Foster, I. Clarke, D. & Pearce-Kelly, P. 1996. Mortality of Endangered Snails of the Genus *Partula:* Preliminary Results of Pathological Investigations. *Journal of Zoo and Wildlife Medicine.* 27(1) pp 19-27.

Cunningham, A.A. & Daszak, P., 1998. Extinction of a Species of Land Snail Due to Infection with a Microsporidian Parasite. *Conservation Biology. 12(5) pp 1139-1141.*

Dalziel, A., Flach, E., Pearce-Kelly, P., McFarlane, D. & Sainsbury, A., 2013. Disease risk analysis for the translocation of captive bred Partulid Polynesian tree snails. Report for the Partulid Global Species Management Group, La Service Direction de l'Environnement de la Polynésie française and the IUCN Mollusc and Reintroduction Specialist Groups. ZSL.

Flach, E.J., Pizzi, R., Macgregor, S. & Clark, B., 2008. Protozoal infections in *Partula* snails at the Zoological Society of London; 2006-8. Proceedings of the British Veterinary Zoological Society Spring meeting, 17-18th May 2008, Woburn Safari Park. Page 25.

Goodey, G. & Flach, E.J., 2015. Pre-release health screening of Partula species: 2015. ZSL internal report, shared with all collections holding *Partula* species.

Goodey, G. & Flach, E.J., 2016. Pre-release health screening of Partula species at ZSL London zoo: 2016. ZSL internal report, shared with all collections holding *Partula* species.

Gouveia, A. 2011. Investigation of the Factors Affecting the Population Dynamics of Captive *Partula* Snails. PhD Dissertation Imperial College London, University of London.

Keeling, P.J. & Fast, N.M., 2002. Microsporidia: Biology and Evolution of Highly Reduced Intracellular Parasites. *The Annual Review of Microbiology.* 56 pp 93-116.

Pearce-Kelly, P., Blake, E., Goellner, R. & Snider, A., 2007. Management guidelines for the welfare of zoo animals: Polynesian tree snails, family *Partulidae.* BIAZA, p 24-25.

Peterson, T.S., Spitsbergen, J.M., Feist, S.W. & Kent, M.L., 2011. The Luna stain, and improved selective stain for detection of microsporidian spores in histologic sections. Diseases of Aquatic Organisms, 95, p 175.

Weiser, J., 1976. Microsporidia in Invertebrates: Host – Parasite Relations at the Organismal Level. *Comparative Pathobiology.* 1 pp 163-201.

Appendix 1. Monitoring tank populations.

Source Zoo	Species	Tank pop'n	Date	Census				Deaths/losses since last census				No.euthanised (Adult unless stated)	No.submitted for screening
				N	**J**	**S**	**A**	**N**	**J**	**S**	**A**		

Appendix 2. **Faecal sample parasitology results.**

Source Zoo	Species	Tank pop'n	Sample no.	Date	Lab ref.	Wet prep.findings	MZN findings	Bacteriology*
			1					
			2					xxxxxxxxxxxxxxxxx
			3					xxxxxxxxxxxxxxxxx
			4**					xxxxxxxxxxxxxxxxx

* Only one faecal sample needs to be sent for bacteriological culture

* * Extra faecal checks if acid-fast cysts found; to ensure three clear faecal samples before export

Appendix 3. *Post mortem* examination results.

Source Zoo	Species	Tank pop'n	Dead/euth	PM number	Date	Apex MZN	Bacteriology	Histopathology

Appendix 4. Screening summary sheet

Source Zoo	Species	Tank pop'n	Initial pop's number	Mortality (No.)	Mortality (%)	Faecal parasites	Faecal bacti	PM exams (dead)	PM exams (euth)	Apex MZN	Coelom bacti	Histo-path findings	Clear to export?

Appendix 3 – Partula snail transportation guidance

Guidance for Partula snail transportation containers and environmental conditions

(Updated March 2019)

The following guidance is intended to help facilitate live animal cargo shipments of *Partula* snails between participating collections and for reintroductions to French Polynesia.

Wrapping the snails

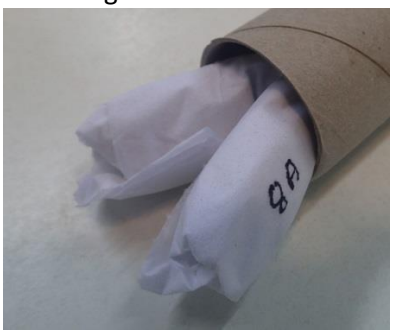
Other than very brief journeys, *Partula* snails are best transported in an inactive, semi-aestivation like state. This is achieved by wrapping the snails in several layers of tissue and placing them inside a rigid cardboard tube (see photos below) to both protect the snails and ensure that the environment is conducive to producing and maintaining the desired aestivation like state.

Adult snails ready for rolling in double layer tissue

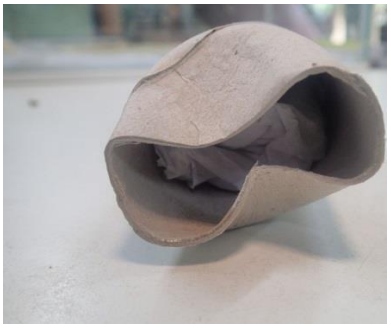
Rolling tissue to mid-point prior to folding

Folding in half

Folded tissue ready for tube



Folded tissue being placed in tube with written development stage numbers



Pinched ends of tube holding up to 30 adults

Inner container

The tubes are then placed inside either a cardboard box or a plastic container (if the latter be sure to include ventilation on the lid). A standard 25cm long cardboard tube can accommodate up to 30 adults or at least twice as many younger life stage snails (i.e. new-born and/or juveniles) but if shipment room allows it's desirable to have fewer adult snails in each tube.

Cardboard tubes placed in inner container with logger

Shipment crate

A wooden IATA compliant crate of the kind and size below (1cm thick, 52cm wide by 40cm deep by 42cm high) with an inner polystyrene box (2cm thick) is sufficient for transporting 500 adult Partula with enough remaining space for the inner snail container to be surrounded by the water bags (see below). Larger numbers (if all adults) would need a commensurately larger shipment crate. Ventilation is provided on both the wooden and polystyrene boxes and covered with fine mesh (NB. The polystyrene box lid can also be ventilated with lots of small holes - removing the need to mesh).

1cm thick plywood crate with 2cm thick inner polystyrene box with ventilated lid

Inner container (inc temp and RH logger) surrounded by water bags

Water bags surrounding inner Partula container with egg carton on top to facilitate air flow to inner container

Water bag placed atop of egg carton topped inner container

A 'space-bag' insulated liner has also been used successfully in more recent shipments, replacing the need for polystyrene.

'Space bag' liner used in 2017

Temperature stabilising water bags.

Experience has shown that the inclusion of water bags is a great aid in ensuring that good temperatures are maintained inside the container even if the crate is subjected to unexpectedly high and/or low temperatures at any stage of the journey.

Half fill the water bags and expel all the air (a half filled 10L bag will weigh about 4.5kg) and ideally surround the inner *Partula* container with these bags on all sides (i.e. 6 bags – one on each side).

If these water bags have been filled and left in the *Partula* facility overnight prior to packing they should be at the ideal temperature and tests have shown that they do a good job of maintaining a stable temperature.

The 10L Folding Drinking Water Container Storage Bag (40cm x 28cm). Bag in photo is blue but the clear option may be better for visibility of contents. These can be obtained from Amazon for around €4.5/£4 each. 5L bags (33cm x 30cm) are also obtainable and a combination of these can be useful.

Don't forget the logger!
It's very valuable to place a temperature logger (or better still a temperature and humidity logger) inside the inner *Partula* container to ensure that a full temperature record is obtained throughout the transportation period. Also having a logger attached to the outside of the crate will enable comparative temperatures to be made (helping to determine the effectiveness of the insulated internal crate environment.

Securing the crate lid
Screws are the best option as this enables the crate to be opened for inspection etc. In addition, a cord or cable tie can be used in place of a padlock.

Labels are as for any other live animal shipment (see photo) and as a precaution a full set of the movement paperwork can be included (either on or inside the crate).

Feeding/watering/spraying. Because the above transportation conditions are designed to ensure that the snails go into and remain in an inactive semi-aestivating state throughout their journey the provision of food/water/spraying should be avoided as they will compromise the snails aestivating state.

Temperature conditions. The idea temperature is 20C but a couple of degrees either side of this (ie fluctuations between 18 – 22C) are acceptable.

Humidity. No humidity provision (e.g. spraying or wetting of tissue) is necessary as the tissue wrapped snails need to kept in their inactive state and the wrapping conditions will provide sufficient localised moisture conditions from the snails themselves.

Transportation duration. Providing temperature conditions inside the crate are within the acceptable 18-22C temperature range for the majority of their transportation, the snails should be fine in their semi-aestivated state for 48-72 hours.

Including a full copy of the transportation documentation (inc the signed veterinary certificate copy) inside the crate is a valuable action in case of lost transport documentation (this happened with one of the 2016 shipments and so having a full copy inside the crate at least provides the receiving destination with a full copy of what documentation was provided.

Appendix 4 – Typical detailed servicing regime

Typical detailed servicing regime (as at ZSL London Zoo)

Daily check:
Record min/max temperature and humidity in the morning and react to any issues ASAP. Data logger readings may be downloaded as required or monthly. General visual check of containers and snails for any issues/removal of dead.

Twice weekly (e.g. Monday/Friday) service tank containers –

- Prepare water, diet and bowl of Milton solution
- Make up some food plates ready for use (not too many, so they don't dry out)
- Work through tanks one at a time
- Throughout, carefully move snails only as necessary, taking particular care to look for newborns. They may be put onto a tissue or into a petri dish
- Peel back clingfilm for re-use or remove for replacement if necessary
- Remove old food plates, checking for snails both sides (old food can be sprayed to help cleaning)
- Wipe old food/tissue from the food plates into a bin, and place plates into Milton soak
- Remove soiled base tissue (very gently feel for any snails that may have climbed under/into the sheet).
- Aim to wipe ~1/3 to ½ of glass surfaces with a wet tissue to clean off both faeces and slime (the substrate tissue may be re-used for this). Usually at least the front panel is cleaned completely.
- Wipe dry cleaned surfaces and base of tank with a fresh tissue
- Add new tissue to cover base of tank, using extra folded tissue as required in larger units, aiming to maintain 4 layers of tissue throughout
- Damp tissue (fully damp but no free water present) and place two new food plates
- Replace snails into tank, and close clingfilm carefully
- When last tank is finished, tidy equipment and move all old food plates from soak into dishwasher
- Put dishwasher on, 70° intensive setting
- Clean room afterwards, including worktops and floor, and empty bin

Reminder that tanks should be allowed to dry out between cleans, but only just – re-damp in between if too dry.

Three times weekly (e.g. Monday/Wednesday/Friday) - service box containers

- Similar procedure to tanks, however food is smeared directly on one side of box
- Extra care needed when manipulating box as when set upright (as usual), may fall over and damage snails

Days when no direct servicing is carried out can be used to prepare materials, such as diet, cuttlebone chunks and piles of tissue for the tank bases.

Extra cleaning of tanks/box and food plates.

Each tank is given a thorough clean once a month. This involves soaking in hot tap water, washing with a scouring pad and suitable bactericidal detergent such as Safe4, or Sterex at a ratio of 1-part Sterex to 80 parts water in a standard hand sprayer. This is followed by thorough rinsing with hot water and finished with a warm rinse to warm the tank prior to snails being reintroduced if used immediately (it is worth having spare tanks to be able to rotate cleaning).

ZSL London Zoo, views of Partula room

Appendix 5 – Products in the text

Recommended products mentioned in the text and currently in use at institutions

- **Clingfilm** – Rolls of catering clingfilm, 35cm or 45cm perforated are ideal, depending upon availability. Note proper breathable clingfilm must be used, not PVC wrap. Table top dispensers should also be available from the same catering suppliers.
- **Tissue** ('Hygiene') roll – Simple towel paper on 25cm wide rolls, perforated is the norm, to fit tank dimensions, via catering suppliers.
- **Water filters** – Brita Maxtra filter and cartridges
- **Antimicrobial handwash** – HibiScrub (widely available)
- **Milton sterilising fluid** – Procter & Gamble Ltd
- **Safe4** – www.safe4disinfectant.com
- **Sterex** – by Unico www.unicodirect.com
- **Lighting** – full spectrum, Arcadia T5 6% luminaire, www.arcadiareptile.com
- **Dataloggers** – e.g. Homechip temperature/humidity units
- **Plastic containers**, Stewarts Plastics – www.stewart-solutions.co.uk
- **Stress food supplement** – Bob Martin limited (can be ordered via Amazon)
- **Food plates** – cut from standard acrylic sheet 6mm thick, dimensions 280x130mm.
- **Enamel paint** – Plastikote brand fast drying Project Enamel. Note although labelled as enamel, this is water based acrylic.

Appendix 6 – Chronology of events

Chronology of significant partulid events

1774	*Partula faba* from Raiatea brought back from Captain Cook's expedition (erroneously described as being collected from Tahiti)
1791	*Partula faba* becomes the first partulid species to be officially described, by Férussac
1884	Andrew Garrett. *The terrestrial mollusca inhabiting the Society Islands*. J. Acad. Nat. Sci. Philadelphia (2nd Series) 9 (Part 1)
1909-1910	Henry Pilsbury. *The Family Partulidae. Manual of Conchology*. Philad. (Section 2) 20
1916	Henry Edward Crampton. *Studies on the variation, distribution and evolution of the genus* Partula*. The species inhabiting Tahiti*. Carnegie Inst. Wash. Pub. 228
1925	Henry Edward Crampton. *Studies on the variation, distribution and evolution of the genus* Partula*. The species of the Marian Islands, Guam and Saipan*. Carnegie Inst. Wash. Pub. 228
1932	Henry Edward Crampton. *Studies on the variation, distribution and evolution of the genus* Partula*. The species inhabiting Moorea*. Carnegie Inst. Wash. Pub. 228
1962	Bryan C Clarke (University of Nottingham) and James J Murray (University of Virginia) begin thirty years of *Partula* research on Moorea (later joined by Michael S Johnson, University of Western Australia)
1967	*Achatina fulica* introduced onto Tahiti
1968	Yoshio Kondo. *Partulidae: Preview of Anatomical revision*. Nautilus 81 (3)
1969	B C Clarke and J Murray*. Ecological genetics and speciation in land snails of the genus* Partula. Biol. J. Linn. Soc. 1
1970	Extensive collection of partulids on Tahiti by J B Burch of the University of Michigan on behalf of Yoshio Kondo
1974	*Euglandina rosea* first introduced onto Tahiti

1977	*Euglandina rosea* introduced onto Moorea
	M S Johnson, B C Clarke and J Murray. *Genetic variation and reproductive isolation in* Partula. Evolution 31(1)
1980	J Murray and B C Clarke. *The genus* Partula *on Moorea: speciation in progress*. Proc. Roy. Soc. Lond. B 211
1981	First captive-breeding colony established in a zoo, at the Jersey Wildlife Preservation Trust (now the Durrell Wildlife Conservation Trust). Small collection also initiated at London Zoo (did not survive)
1986	M S Johnson, J Murray and B C Clarke. *Allozymic similarities among species of* Partula *on Moorea*. Heredity 56
	Proper ZSL captive collection initiated
1987	All species of *Partula* believed extinct on Moorea
	CBSG programme conception meeting
1988	J J Murray, E Murray, M S Johnson and B C Clarke. *The Extinction of* Partula *on Moorea*. Pac. Sci. 42(3-4)
1991	'Operation *Partula*' survey and crisis collection mission to Society Islands
	Creation of CERCI management database dedicated to recording and analysis of captive breeding data.
1993	Trial release and monitoring of *Partula* onto Polynesian plants at the Royal Botanic Gardens, Kew
	M S Johnson, J J Murray and B C Clarke. *The ecological genetics and adaptive radiation of Partula on Moorea*. Oxford Studies In Evolutionary Biology
1994	CBSG Action Plan meeting (Partula *'94: An Action Plan for the Conservation of the Family Partulidae*, by the Pacific Island Land Snail Group) and formation of Pacific Islands Land Snail Group (PISLG)
	'Operation *Partula* '94' expedition to Society Islands
	Construction of predator-proof reserve on Moorea
1995	PILSG 2 month expedition to Society and Marquesas Islands
1996	Restocking of Moorean reserve for restart of trial release experiment
	Last known *P. turgida* (now reclassified as *P. clarkei*) dies in captivity and species extinct

1997	Legislation passed by French Polynesian government for the protection of the Partulidae, and the declaration of *E. rosea* as a 'noxious species'.
1998	Termination of trial release experiment on Moorea due to continued issues with maintenance.
2001	*'An urgent briefing report for the French Polynesian Government, associated agencies and the IUCN on the conservation status of the endemic tree snails (Partulidae) of French Polynesia'* produced by Dr Trevor Coote on behalf of the Partulid Conservation Programme
	Confirmation of extinction of all Society Island partulid species outside of Tahiti and Moorea
	Promise of funding from French Polynesian Government for 2003
	New version of the Partulid Species Management System (PSMS)
2002	Construction of predator-proof reserve in Faaroa Valley, Tahiti
2003-2005	French Polynesian Government commissioned Dr Trevor Coote to undertake a three-year set of survey work for the development of a conservation strategy for French Polynesia's endemic tree snails and associated forest
	Partulid Programme member Consortium established to help cover Trevor Coote's salary (matching the Governments field related grant) to undertake the regional conservation strategy commission
	Remnant populations of partulids discovered or confirmed on Tahiti, Moorea, Huahine (Society Islands); Ua Pou, Ua Huka and Hiva Oa (Marquesas Islands); Rurutu*, Rimatara*, Rapa, Raivavae, Tubuai* (Austral Islands) **Partula hyalina,* originally from Tahiti
	Conservation Action Plan for the long term protection of French Polynesia's last surviving populations of endemic tree snails of the genera *Partula, Samoana* and *Trochomorpha* published (2005)
2006	Remnant populations of partulids discovered on Raiatea
2006-2008	Funding agreed by the French Polynesian Environment Ministry to implement the recommendations in the 'Long Term Action Plan for the Protection of the Endemic Tree Snails of Polynesia'
2012	Predator exclusion zone constructed in Te Faaiti Valley on Tahiti
2015	First release of Partula into Tahiti reserve – *P. hyalina, nodosa* and *affinis*, plus experimental release of *nodosa* into Mape trees
	Death of last captive specimen of *Partula faba* and species extinct

2016-2018	Continued release of snails direct into forest areas on Tahiti and Moorea, with experimental release on Raiatea (2016)