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Technical Report 194

Development of tree snail protection enclosures: From design to implementation

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This technical report series began in 1973 with the formation of the Cooperative National Park Resources Studies Unit at the University of Hawai'i at Mānoa. In 2000, it continued under the Pacific Cooperative Studies Unit (PCSU). The series currently is supported by the PCSU.

The Pacific Cooperative Studies Unit at the University of Hawai'i at Mānoa works to protect cultural and natural biodiversity in the Pacific while encouraging a sustainable economy. PCSU works cooperatively with private, state and federal land management organizations, allowing them to pool and coordinate their efforts to address problems across the landscape.

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ABSTRACT

Hawaiian land snails in the endangered, endemic genus *Achatinella* have experienced major declines in population and distribution over the last 100 years. Threats to *Achatinella* today include invasive, non-native predators (*Euglandina rosea, Rattus rattus* and *Trioceros jacksonii*), habitat degradation due to human disturbance and possibly climate change, and historically, collection by humans. The O‘ahu Army Natural Resources Program (OANRP) is required to stabilize select remaining populations of *A. mustelina*. Stabilization goals are to maintain 300 mature snails at eight managed sites and control threats within sites. This report describes OANRP efforts to combat invasive predators by means of predator-free and -proof snail enclosures. A couple of prior attempts at excluding predatory snails were marginally successful but the identification of additional predators required substantial additional barriers. The design and construction of the enclosure at Pu‘u Hapapa is used as a case study. This report includes detailed information on the physical development of predator-proof barriers, construction and costs. Additional needs for monitoring and maintenance, predator removal, *Achatinella* reintroduction, *Achatinella* population monitoring, and habitat improvement were also developed.

INTRODUCTION

In 1998, the U.S. Army (Army) initiated formal consultation under section 7 of the Endangered Species Act (16 U.S.C. 1531 et seq.) with the U.S. Fish and Wildlife Service (USFWS) to determine if routine military training at Makua Military Reservation would jeopardize the continued existence of 41 endangered species. The Army is responsible for maintaining the stability of each of these taxa, and applying additional management specified to those taxa below stable population levels. To stabilize the target taxa each taxon must be maintained with sufficient numbers of populations to ensure their long-term viability. Additionally, threats to the managed and reproducing individuals in each population must be controlled, and each taxon must be adequately represented in *ex situ* collections. Stabilization is only the first step toward eventual recovery of these endangered species.

For the Hawaiian tree snails on O‘ahu only the endangered, endemic genus *Achatinella* is currently identified for management (Fig. 1). The Army is obliged to:

- manage snail populations at eight field locations to encompass the extant range of the species and to include all six genetically defined evolutionarily significant units (ESU).
- achieve at least 300 snails per population.
- maintain captive populations for each of the six recognized ESUs.
- control all threats at each managed field location.

These snails reproduce at very slow rates and appear to be defenseless against the non-indigenous predators. Their population levels have been diminishing at an alarming rate. Controlling the principal threats in the field is difficult, if not impossible. Maintaining populations in the laboratory has been disappointingly unrealistic. To meet the above obligations, the only possible approach was to create small protected areas in the wild that prevented the predators from gaining access. This paper describes our attempt to create safe area enclosures.

Figure 1. Mature *A. mustelina*, Wai‘anae range, O‘ahu

The O‘ahu Tree Snails

Hawaiian tree snails of the subfamily Achatinellinae are unique to the Hawaiian Islands and highly endangered in the wild. The entire genus *Achatinella* has been listed on the Endangered Species List since 1981 (Kobayashi and Hadfield 1996). *Achatinella* species are arboreal, pulmonate gastropods and are found only on the island of O‘ahu in both the Ko‘olau and Wai‘anae mountain ranges (Fig. 2). These rare snails have long been of interest to students of animal evolution because of the radiation of attractive shell color, banding patterns and shapes (Gulick 1905).

The O‘ahu Army Natural Resources Program (OANRP) has worked with numerous species of *Achatinella* including *A. bulimoides, A. byronii/decipiens, A. lila, A. livida, A. mustelina* and *A. sowerbyana.* These tree snails have slow growth, late maturity, low motility and a low rate of fecundity – between one and four live births per year (Hadfield and Kobayashi 1996). During years of drought, chances of reproduction are further diminished. If predation is constant and sufficiently intense, the prey population will eventually die off. The *Achatinella* species probably had no predators in pre-human times; therefore, they were able to form dense populations. Like many other plants and animals of oceanic islands, they lost defenses against introduced predators and competitors. Unfortunately, the destruction of habitat and rat, carnivorous snail and more recently Jackson chameleon predation are pushing these endemic species toward imminent extinction. This report documents efforts to provide a safe habitat for the tree snails.

Achatinella Biology and OANRP Management

Achatinella snails are born live and relatively large (4-5 mm), grow slowly (approximately 2 mm/year), become reproductively mature at a relatively late age (4-5 years old), and have low fecundity compared with most other terrestrial snails like *Achatina fulica* (Kekauoha 1966), *Partula* spp. (Murray and Clarke 1966), *Liguus fasciatus* (Voss 1976), *Caracolus* and *Polydontes* spp. (Heatwole and Heatwole 1978). The average lifespan is estimated to be at least 11 years (USFWS 1992).

Pilsbury and Cooke (1914) reported that tree snails could no longer be found below 305 m elevation. Previously in the mid 1800s tree snails had been found in abundance at lower elevations. Today, when surveying for tree snails, OANRP uses the 610 m elevation contour as a guide for identifying potential habitat because there are very few populations known below this elevation. Old locations below 610 m that used to have rich snail concentrations are only extensive shell graveyards today.

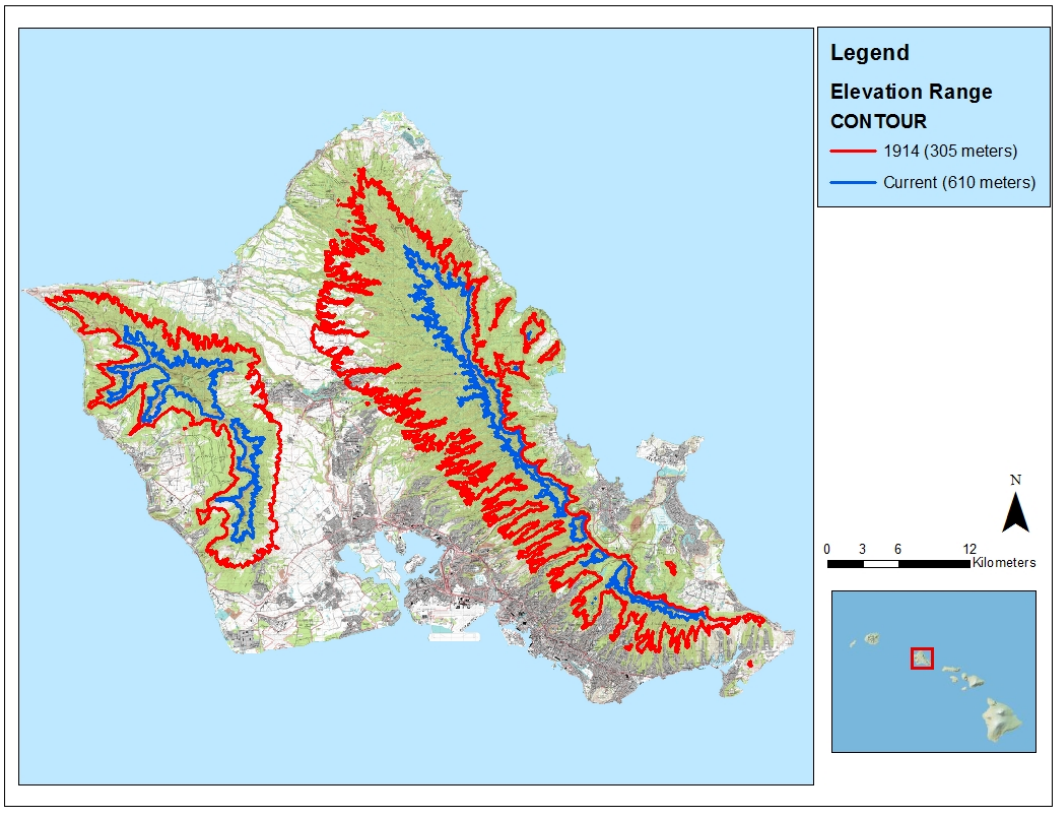

Figure 2. Approximate 1914 *Achatinella* range contrasted with current range

OANRP MANAGEMENT STRATEGY

Achatinella species occur within four Army training areas on O‘ahu:

- In the Wai‘anae range, Makua Military Reservation and Schofield Barracks Military Reservation have populations of *Achatinella mustelina* (Fig. 2).
- In the Ko‘olau range at the Kawailoa Training Area and Schofield Barracks East Range, there are several species of extant *Achatinella*.

Initially we tried to manage species in both the Wai‘anae and Ko‘olau ranges principally by maintaining captive populations in the laboratory. However, in the last five years we have focused on *A. mustelina* only. Management of this species was designed after a genetic analysis which indicated that there were six Evolutionary Significant Units (ESU) across the Wai‘anae range (Holland & Hadfield 2002). The Makua Implementation Plan (MIP) directed OANRP efforts to these six units (Makua Implementation Plan 2003). Two of the six ESUs are geographically extensive (Fig. 3). For these ESUs, two locations were designated for management representing the geographical spread. Thus, *A. mustelina* is currently managed in a total of eight sites. The stabilization goal is to have 300 snails under some form of threat control at each of these sites (Makua Implementation Plan 2003). The level of control varies from total exclusion of all three predators to intensive management of rats only. The overarching determinant of which approach is taken is the availability of suitable sites to create predator enclosures, the preferred management alternative.

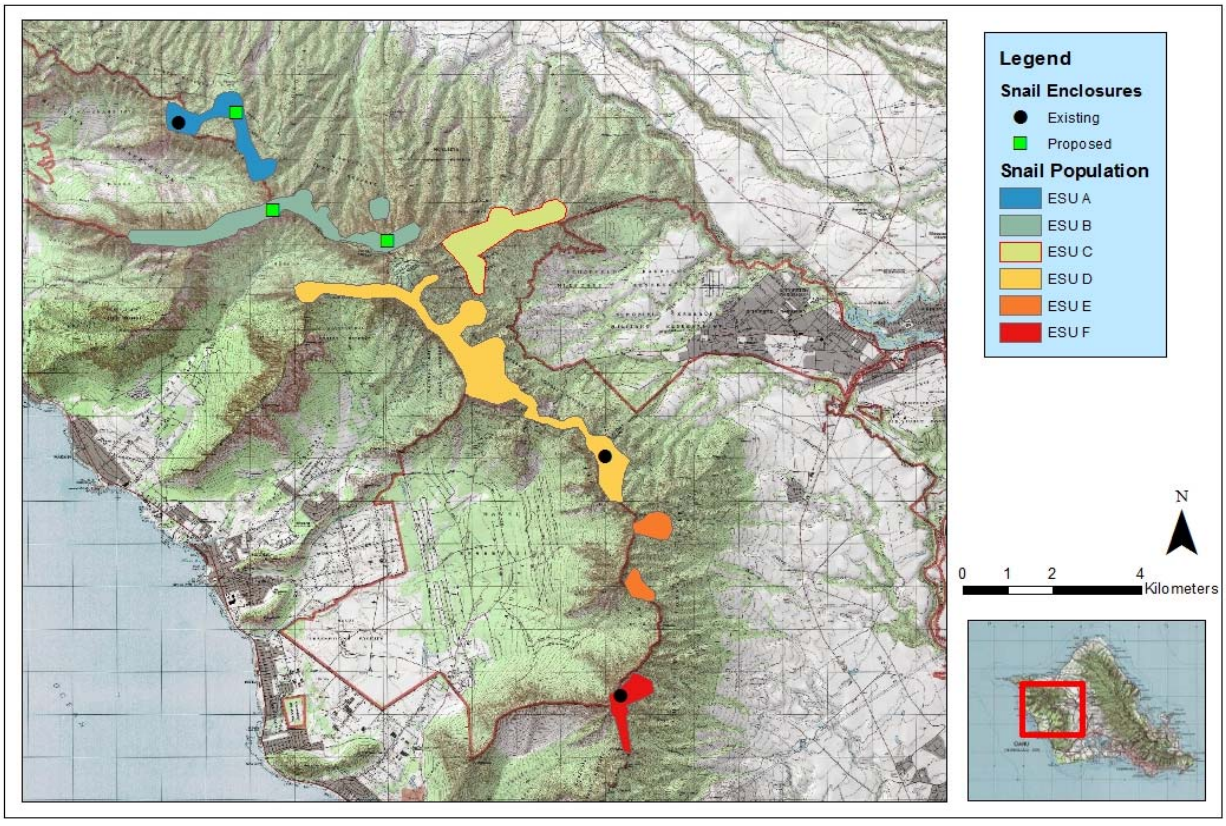

Figure 3. *A. mustelina* Evolutionary Significant Units (ESU) in Wai‘anae mountains O‘ahu

Non-native Predators of *Achatinella*

Predators of *Achatinella* include Black rats (*Rattus rattus*), rosy wolfsnails (*Euglandina rosea**)***, and Jackson's chameleons (*Chamaeleo jacksonii* subsp. X*antholophus.* Each presents its own unique challenges as discussed below. This triple threat of predators confounded by environmental factors has resulted in massive extinctions.

Rats. There are three species of rat present in Hawai'i; *Rattus rattus* the Black or Roof Rat*, R. norvegicus* the sewer or wharf rat and *R. exulans* the Polynesian rat. Of these three *R. rattus,* in particular, is common in the forested areas of tree snail habitat and typically arboreal. *R. rattus* is known to predate on tree snails with devastating results (Hadfield & Mountain, 1980). Hadfield studied tree snails at Pu'u Kanehoa and Pahole in the Wai'anae range in the 1970s and 1980s where he observed rat-eaten shells at both of these sites (Fig. 4). Shell caches are often observed where rats discard shells of eaten snails. When black rats hone in on tree snails as a food source they can rapidly destroy a population as they are very adept at moving through the canopy finding and destroying tree snails. OANRP staff has seen rats climbing in the canopy during night surveys in tree snail population areas. Rats can be controlled locally by kill traps. However, in most control areas there is insufficient access or resources to install and maintain an array of traps that would completely exclude rats. In tree snail populations where OANRP conduct rat control, rats are continuously removed but it is likely that a low level of tree snail predation continues despite trapping efforts. The benefit of constructing a rat barrier is complete rat removal without continually maintaining a kill-trap grid.

Figure 4. Rat predated *A. mustelina* shells

Rosy wolfsnail. *Euglandina rosea* is known to have a catastrophic impact on tree snail populations (Hadfield & Mountain, 1980; Meyer & Cowie, 2010). Since its introduction in 1955 *E. rosea* has spread to all tree snail habitats on O‘ahu concurrent with a significant decline or extirpation of the snails. *Euglandina rosea* is cryptic, highly mobile and fecund (Fig. 4). On O‘ahu they are found from sea level to at least 1100 m on the northern slopes of Ka’ala. The highest known site for *Achatinella* is 1140 m, thus their habitat is almost sympatric. Many naturalists report that once *E. rosea* move into an area, the tree snails decline. Hadfield (pers. comm.) stated that at Pu‘u Kanehoa it was the appearance of *E. rosea* that most clearly coincided with the disappearance of all tree snails from the study site. In many areas of the Wai‘anae range, locations of once healthy populations of snails are only evident in the shells that remain scattered on the ground. In these areas, there are often concentrations of *E. rosea* shells as well. The common assumption is that *E. rosea* numbers peak when native snails are common but as the tree snail population declined so did the *E. rosea*. There are no baits or traps available to control *E. rosea*. Currently, the only actions possible are searches and hand removal. This is a difficult task, see discussion below for some evidence on the effort OANRP exerted to remove them from within the enclosure at Pu‘u Hapapa.

Figure 5. *Euglandina rosea* attacking an *A. mustelina*

Jackson's chameleons. Jackson's chameleons (*Chamaeleo jacksonii* subsp. x*antholophus*) are a newly documented threat to tree snails (Holland *et. al.* 2010). Since this discovery, OANRP personnel have been removing chameleons from tree snail areas. One large female chameleon was collected on Pu'u Hapapa in 2012 that had five *Achatinella* shells in its stomach at various stages of decomposition. Further dissections conducted by the University of Hawai'i Tree Snail Conservation Laboratory (HTSCL) have recorded predation on *Achatinella* by chameleons on at least four other occasions. This includes one from within an enclosure and three in close proximity to the Pu'u Hapapa enclosure. These observations likely underestimate the impact of Jackson's chameleons as the snail shells pass through the gut within 3-7 days. The only currently available control option is hand removal, a work intensive operation because the chameleons are very difficult to detect in the canopy.

These three predators present multiple challenges for managers intent on conserving the tree snails. None of the current control techniques is long-lasting. They are all labor intensive and thereby expensive. The only feasible option was to permanently isolate the tree snails from the predators using protective enclosures.

SNAIL ENCLOSURES

Early Enclosures

Initial efforts were started with funding from the State of Hawai'i Natural Area Reserves (NAR) Program. Two prototype enclosures, one each at Pahole Gulch in Pahole NAR and Kahanahaiki in Makua Military Reservation, were constructed with the goal of excluding both rats and *E. rosea*. These barriers were based on examples from Moorea, Tahiti that were designed to protect rare tree snails (Coote *et al*. 2004). Both enclosures were constructed of a solid four foot wall (plywood for one enclosure and corrugated material for the other). At the top of the vertical wall an overhang to the outside of the wall was constructed to deter rats. Under the overhang two barriers were installed to exclude *E. rosea*, one electrical and one chemical. The electrical barrier consisted of two wires. One was grounded and the second wire connected to a Red Snapper™ electric fence energizer. This system would deliver a potentially lethal jolt to any snail that touched the energized wire while contacting the ground wire. This prototype system was not ideal as it was very difficult to maintain the energized wire in close proximity to the ground wire. Often it would short and send a spark across to the ground wire. In addition, this barrier could be potentially lethal to native snails. Beneath the wires a small vinyl trough was installed that was designed to hold a salt-saturated piece of carpet in hopes that this would deter *E. rosea*. This barrier proved difficult to maintain as the salt rapidly dissolves in high humidity environments. The humidity in Kahanahaiki and Pahole often reaches 100% at night. Rainfall is about 1400 mm per year (Online Rainfall Atlas of Hawai'i).

OANRP never formally tested these barrier systems in a controlled setting against any of the predators but incursions were noted. Over time, OANRP found that baits and traps were

needed to control rats as the barrier was not effective. The salt was corrosive and damaged the vegetation as it washed into the soil. *Achatinella* continued to decline.

Development of New Barriers

In March 2008, two OANRP staff members went to New Zealand to evaluate the vertebrate predator control fence technologies and explore their applicability to Hawai‘i. They discovered that the rodent exclusion system was robust enough to act as the backbone for additional barriers to keep out *E. rosea*. The basic design of the fence includes a buried section to prevent animals from digging underneath, a tight mesh vertical fence and a seamless solid metal hood overhanging the exterior of the fence (Fig. 6). A variety of materials have been used very effectively indicating considerable adaptability to suit various needs.

Figure 6. Small mammal pest proof fencing in New Zealand showing the basic structure and its use to enclose very large areas.

After the trip OANRP focused on methods to exclude *E. rosea*. Various electrical and structural configurations were studied, three of which proved effective at excluding *E. rosea*. These were:

- a downward oriented reflexed metal sheet (angle barrier),
- a non-lethal four wire electric barrier, and,
- a barrier of closely placed erect wires much like a brush (cut-mesh barrier).

Trials to test each of the three barriers against a control (no barrier) were conducted at the HTSCL. Boxes with each of the three types of barriers were covered with a larger screen to prevent any *E. rosea* escaping. A similar open plywood box served as the control. In each trial ten *E. rosea* were placed in each box and left for 24 hours. Higher numbers of snails were used for the control group. Instead of 10 snails, 30 were placed in the plywood box with no barriers. The proportion of snails escaping from the barrier box was compared to the control to determine success. Results and final design specifications for each barrier type are given below.

Angle Barrier. Commercial snail propagation operations use tubs with a steep angle overhangs to prevent snails escaping from their pens (Holland, *pers. comm*.). This method of deterring snails is also advised on organic gardening websites (Pests in Gardens and Landscapes, Controlling Snails in Your Yard The Organic Way). Based on this design, OANRP constructed a small trial box that had a Plexiglas™ overhang (Fig. 7). A 15˚ angle was set as the standard between the vertical wall and the Plexiglas. The precise angle is not critical; however for *E. rosea* it should be close to 15˚. The outside edge of the angle is at least seven cm from the vertical wall of the fence which prevents the snails from arching out to reach the edge of the

angle barrier. Snails climbing the vertical wall of the fence proceed under the outside edge of the angle without touching the barrier. They proceed to a point where their shell contacts the overhang at which point their movement up the wall is stopped. While it is possible for a snail to roll one-hundred eighty degrees and transfer itself to the over-hang, this rarely happens. Most frequently the snails just stop or back out (Fig. 7). Over a 48 hour period, only one of 40 *E. rosea* (2%) escaped the angle barrier.

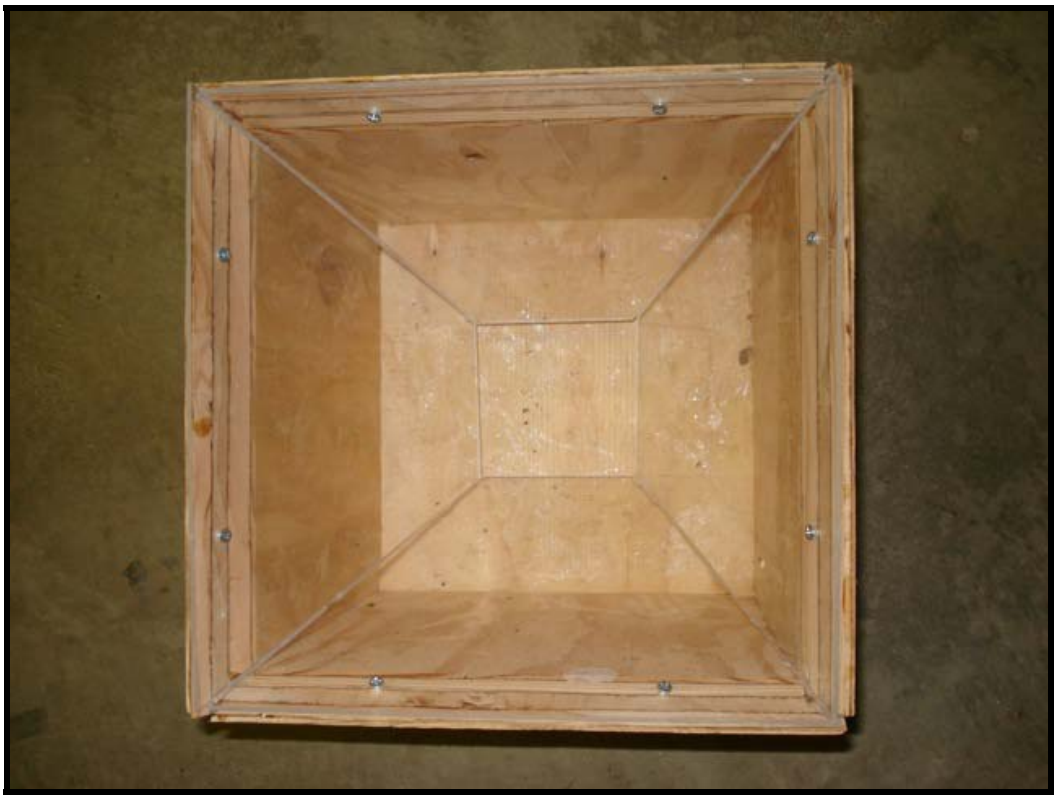

Figure 7. Angle barrier trial box

While not 100% effective, the angle barrier is the cheapest of the three barriers developed (Table 1). It does not rely on batteries to function, and it is simple to install but the 15˚ flashing has to be ordered specially. The angle is made from 26 gauge pre-finished roof flashing. The barrier should be at least 10 cm off the ground (Fig. 8, 9) because it allows room to inspect, maintain and remove snails trapped in the barrier. Currently OANRP uses a mirror mounted on a golf club to see under the angle for trapped *E. rosea.* (Fig. 10). In addition, it needs to be high enough to avoid the possibility of vegetation settling against it creating a bridge that *E. rosea* could cross. Vegetation must be regularly removed from the vicinity of the barrier. The angle barrier should be mounted such that it is strong enough to withstand environmental conditions. In areas of high winds, the angle could be exposed to strain. We use a piece of synthetic lumber trim to strengthen points of connection.

Table 1. Angle Barrier Costs

Product	Cost	Notes
15 degree roof flashing	$27.15 per 10 ft.	This product needs to be custom ordered as it is not available as a standard product. Vendor: Kloeckner Metals Corporation.
Various stainless steel fasteners	$15 a pound	Stainless while the most expensive offers the durability needed
Labor for installation	On average 2 people can install 10 sheets an hour	The rate of installation depends on how many corners need to be negotiated

Figure 8. *Euglandina rosea* trapped in angle barrier

Figure 9. Angle barrier installed directly below cut mesh

Figure 10. Monitoring the angle barrier

Cut Mesh Barrier. The cut wire mesh barrier was designed by Mr. D. Tanji, the OANRP carpenter. The barrier works by presenting a surface that approximates a bed of nails (Fig. 11). When the board is inverted it provides very little surface area for the *E. rosea* to adhere to as it attempts to traverse the barrier. Thus, they fall off. This barrier proved to be very effective with zero successful crossings with the mesh facing downward whereas 46% of snails successfully crossed an upright barrier. Although durable materials of plastic lumber and copper screen are not cheap, this barrier will likely be the most long lasting and dependable of the three barriers. The overall cost of construction and installation is outlined in Table 2. The barrier requires no batteries to run and is overall a much more durable structure than the thin sheet metal creating angle barrier. Velcro, loom combs, brushes, and construction fastening plates where investigated as possible materials. None were found to be suitable. Many options were expensive, mostly iron and therefore prone to rusting, and the combs are generally thin gauge and somewhat easy to bend out of shape.

Figure 11. Finished cut mesh 2” x 6” plastic lumber boards ready to be transported to the field. The tan material in grooves is the construction adhesive.

Thirteen rows of 8 gauge copper screen are secured in groves cut in 2” x 6” (5 x 15 cm) synthetic or plastic lumber material using construction adhesive. These materials were chosen for their long term durability. The grooves are cut eight mm deep along the length of the plastic lumber. The grooves are 3-4 mm wide and 3-4 mm apart. The total width of the cut screen rows is about 8.5 cm. The copper screen is cut into 2 cm wide strips. Once secured the rows of screen stand about 1 cm above the plastic lumber. The screen is fastened into the groove with construction adhesive applied to the groove before the screen is inserted. The final product has 4 cm of plastic lumber border on one side and 1.5 cm on the other (Fig. 12, 13).

Table 2. Cut Mesh Barrier Costs

Product	Cost	Notes
2”x 6”x12’ plastic lumber	$59.72	100% plastic lumber. Freight cost is expensive.
1/8” copper screen roll, 3’ tall	$6.19 per sq. ft.	Was ordered from the U.S. mainland.
Construction adhesive	$2.47 per tube	Several types where used: thicker, stickier ones were best as less viscous types did not hold copper in place
Various stainless steel fasteners	$15 per pound	Stainless steel more durable, expensive. Approx. 10 meters can be fastened with ½ lb of fasteners.
Labor to construct	Approximately two person hours per 12’ board	It is most efficient to work in a team of three with one person cutting the copper and two working to secure it with the adhesive.
Labor to install on fence	On average 2 people can install 3 boards per hour	The rate of installation depends on how many angle changes the fence takes in both the vertical and horizontal. Straight flat runs are easiest.

Figure 12. Raw materials for cut mesh barrier

Figure 13. Partially completed cut mesh board, ten grooves filled with copper mesh with three not yet finished

With this design, the cut edges of the screen are spaced close enough to force *E. rosea* of all sizes to pass over the wire without being able to reach the plastic lumber. In addition, the eyes of the copper mesh are small enough to exclude snails from going through it. Even new born *E. rosea* are too large to pass through the screen. The 13 rows of wire (8.5 cm) are wide enough to prevent large *E. rosea* from reaching over it. Small snails less than 1.5 cm (Fig. 14) are able to move along the length of the screens but cannot cross at right angles to the rows. Large snails cannot remain on surface of cut mesh when moving parallel due to their weight.

Figure 14. Small *E. rosea* attempting to cross the cut mesh barrier

The cut mesh barrier is a heavy structure which has been fashioned in a variety of ways. The weight of a fabricated twelve foot board is 80 lb. On enclosures of thin sheet metal (such as the Xcluder type) there must be a u-channel or angle iron secured on the backside of the fence to provide additional bracing. On plywood structures the barrier can be mounted using deck screws from the inside of the enclosure. At corners or where slope changes occur, compound miter cuts are required (Fig 15). Care must also be taken to ensure that the barrier is mounted tightly to the vertical wall as well as at the joins. Small gaps could allow small *E. rosea* access. The plastic lumber is somewhat forgiving as it has more flex than standard lumber. For example, you can push it into corners and pull joints together with fasteners. This barrier must

be mounted no closer than one meter from the fence hood because the 2” x 6” (5 cm x 15 cm) plastic lumber platform presents a potential risk as a jumping platform for rodents. Studies in New Zealand have shown that rats are able to jump almost one meter vertically (Xcluder staff, *pers. com*.).

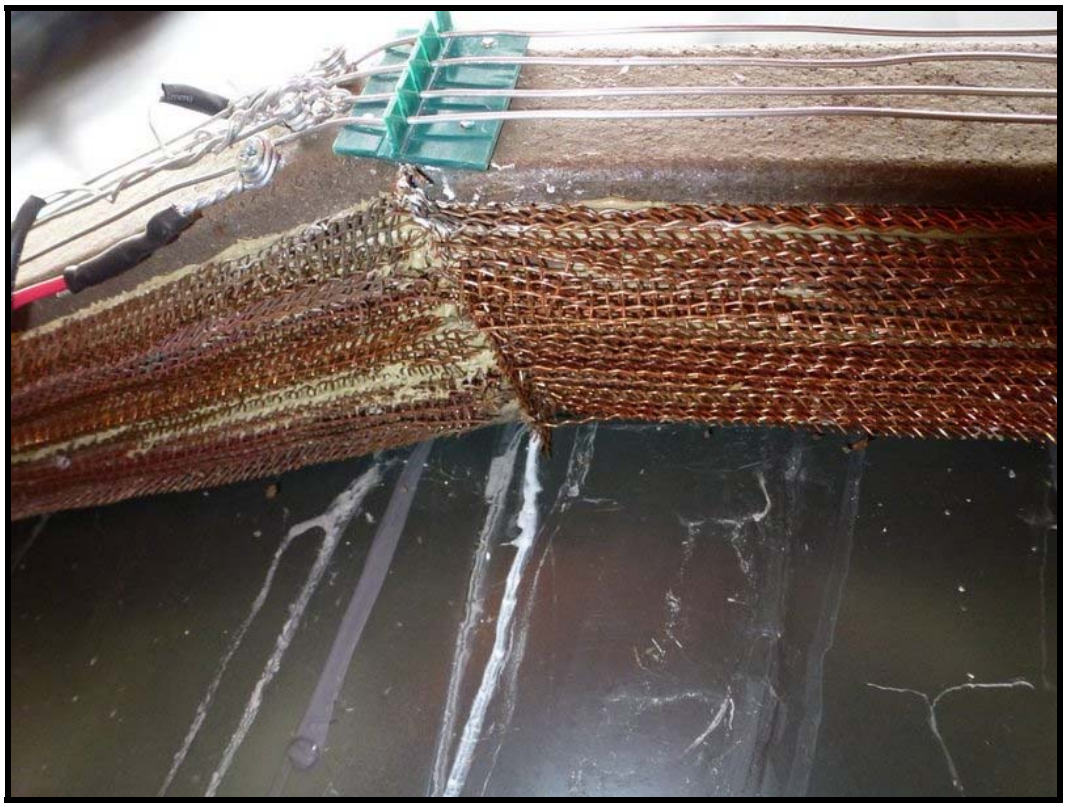

Figure 15. Cut mesh barrier corner joint

Electric Barrier. The electric barrier was a difficult system to develop and went through many iterations before a suitable design was determined. Early in development, OANRP moved away from industry standard electric fence energizers because these systems had previously proved problematic on the Kahanahaiki and Pahole prototype fences. There were two main problems:

- First, unlike livestock, *E. rosea* are not standing on the ground when contacting the electrics. Instead, *E. rosea* are on an insulated surface when they touch the energized wire. Because of this, an additional grounding wire had to be added to the prototype system to ensure shock delivery. The installation of a ground wire with tolerances close enough to ensure small snails would be in contact with both wires at the same time was difficult: at 7,000 – 8,000 volts, the electrified wire often shorted to the ground wire even under dry climatic conditions and drained the battery quickly.
- The second drawback of conventional electric fence systems is that the shock delivered is potentially fatal to any snail that receives it. In areas with native snails, there is no way to prevent contact with *Achatinella* moving out of the enclosure.

With these concerns in mind, the first electrical barrier evaluated was the industry standard 16-wire livestock tape fastened in parallel (Fig. 16). This barrier was extremely effective on the

small trial box scale but lost effectiveness when translated up to the larger scale. This loss was primarily due to the moisture causing shorts in the system.

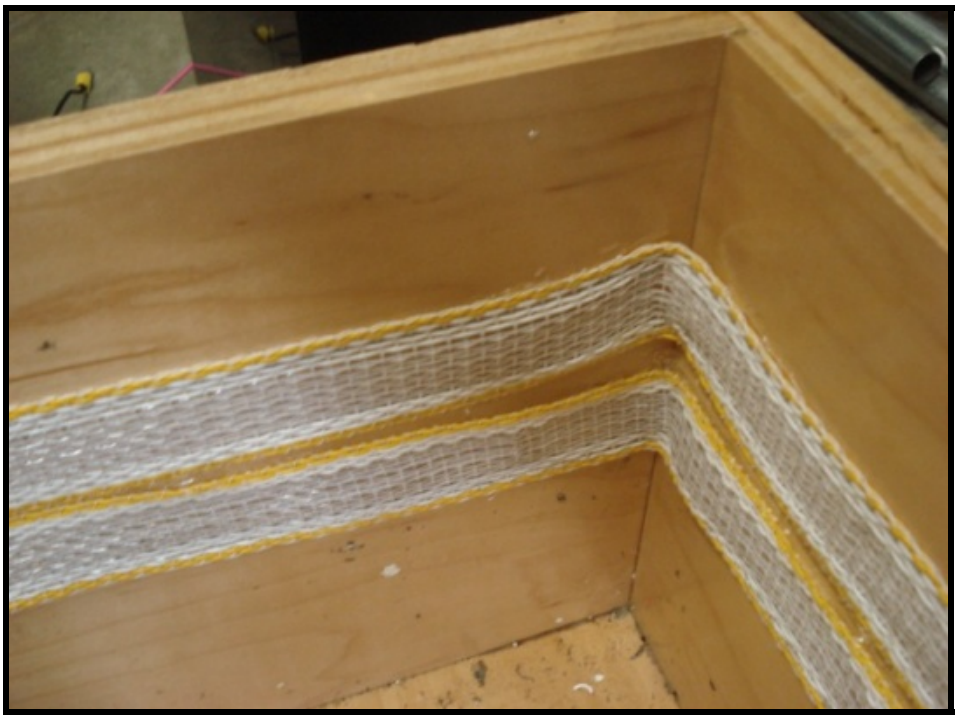

Figure 16. Electrical Barrier trial box

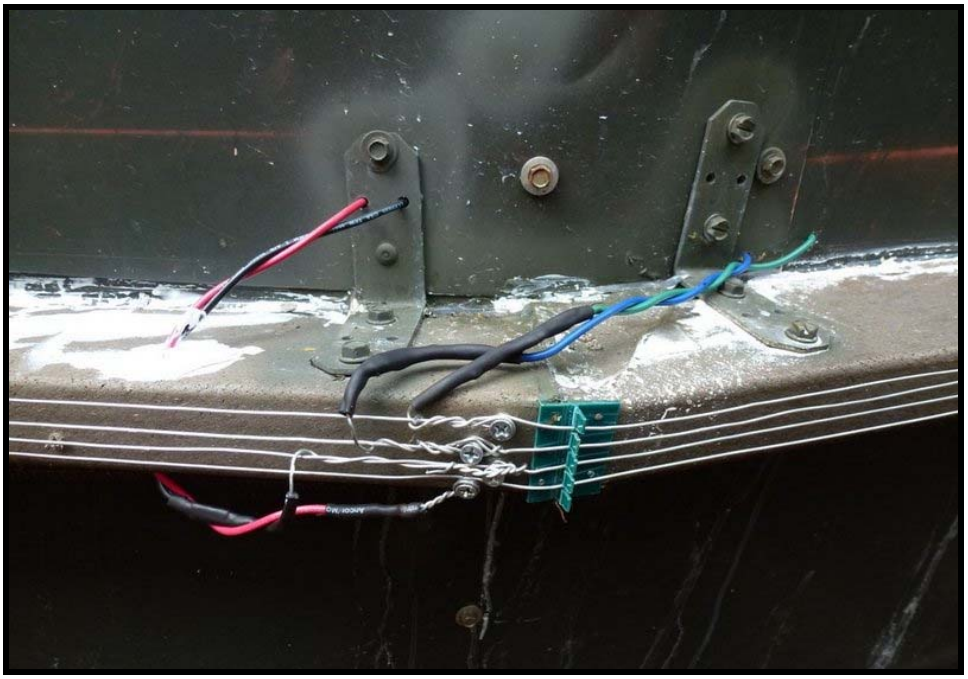

Figure 17. Connection point of electrical barrier wires. By convention the Blue and Green wires are referred to as the Blue wire in the monitoring read out and the Red and Black as Green

In tests at the HTSCL none of the 40 snails crossed the electric barrier within 24 hours. In field trials, it was determined that a 12 volt system that forced *E. rosea* to cross two wires, one charged positive (+) and the other grounded (–), was the best way to deliver a deterrent shock. OANRP tested several kinds of wires, and found that 16-gauge copper gave the best combination of conductivity and durability. In the present system (Fig. 17), four wires run in parallel around the entire fence. The four wires are divided into two independent systems to increase reliability. Spacing between the wires is maintained with custom made vinyl mounts that hold the wires approximately 0.5 cm apart and about 0.3 cm off of the surface of the enclosure. Stainless steel staples are used to secure the wires to the 2" x 6" cut mesh barrier and maintain individual wire spacing between the vinyl mounts. Each row of copper wire is installed in sections to facilitate troubleshooting and repair. For the system to work, extreme care must be taken to ensure that the + and – wires are fully insulated from each other. Costs for this barrier are presented in Table 3.

Table 3. Electrical Barrier Costs

Product	Cost	Notes
Energizer components	$50.00	For a single system.
Battery (1)	$45.00	For a single system.
Solar panel (1)	$75.00	For a single system.
Charge controller (1)	$35.00	For a single system.
16 gauge copper wire	$0.10 per foot	OANRP ordered in 15lb rolls
Brackets and mounting hardware	$35-75	High cost due to custom manufacturing
Person hours electronics	~10 hours	Building and installing energizer and associated electronics. This time varies by experience and size of enclosure.
Person hours installation	~40 hours	Installing mounting brackets and wire around enclosure. This time varies by experience and size of enclosure.

Consistently energizing exposed wires in a humid environment proved challenging. Initial attempts connecting batteries directly to the wires led to shorts in the system that quickly drained batteries and left the wires "cold". After some trial and error, OANRP designed custom energizers based on 555 timing chips and transistors that deliver a consistent, deterring, non-lethal shock (Fig. 18). From a technical standpoint, the energizers are simple, inexpensive (< $50 in parts and 1-2 hrs to construct), time-tested (a-stable 555 chip diagrams date back to the 1970s) and robust (several units have been running continuously for a couple of years now as of October 2013 with only one failure). The energizers pulse a ~600 mA (12V) current at 0.83 Hz (50% duty cycle, ~600 ms on, ~600 ms off) down the wires. The voltage, current, and length of the cycle strike a compromise between power conservation and shock capability. In field tests, *E. rosea* react to the current, even on mildly corroded wires, often falling off the enclosure in response to the shock.

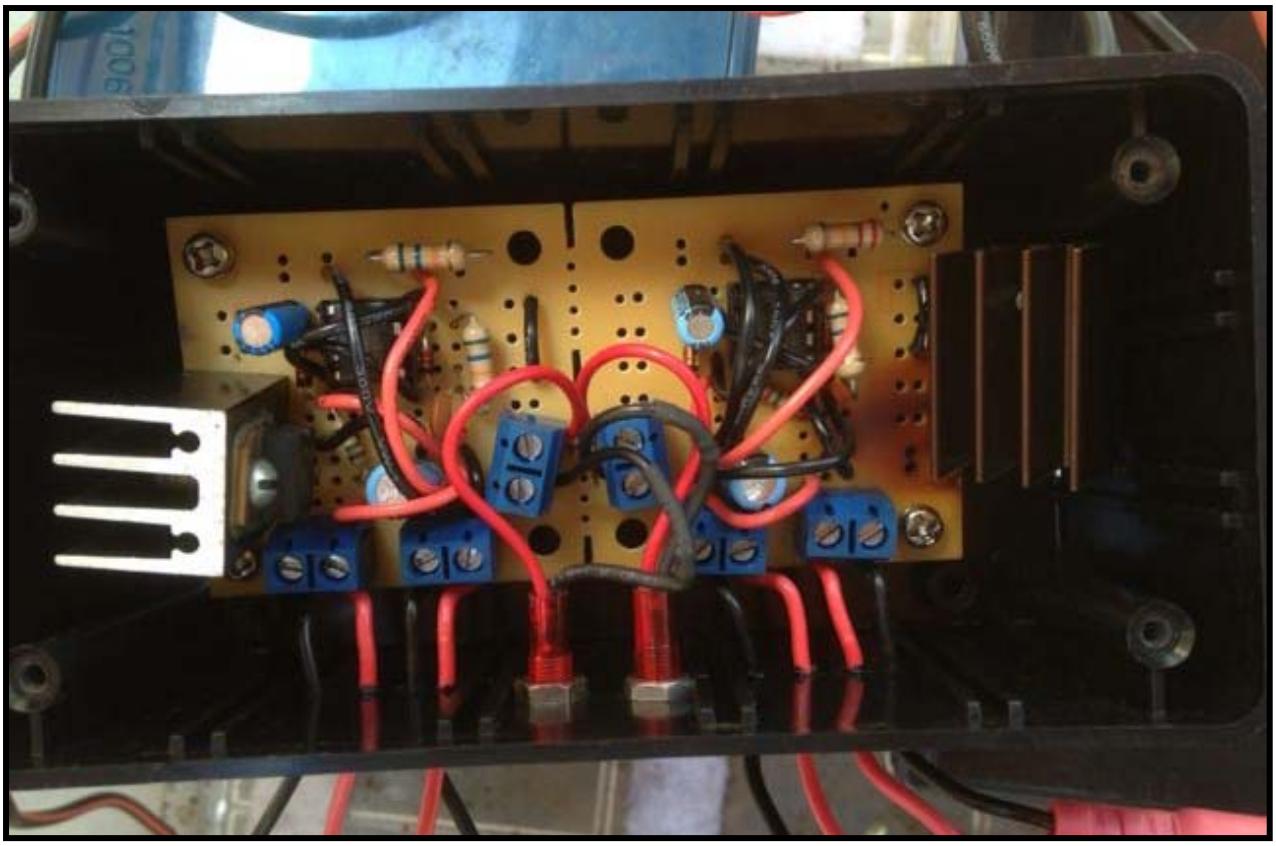

Figure 18. Chip (555) used in electric barrier

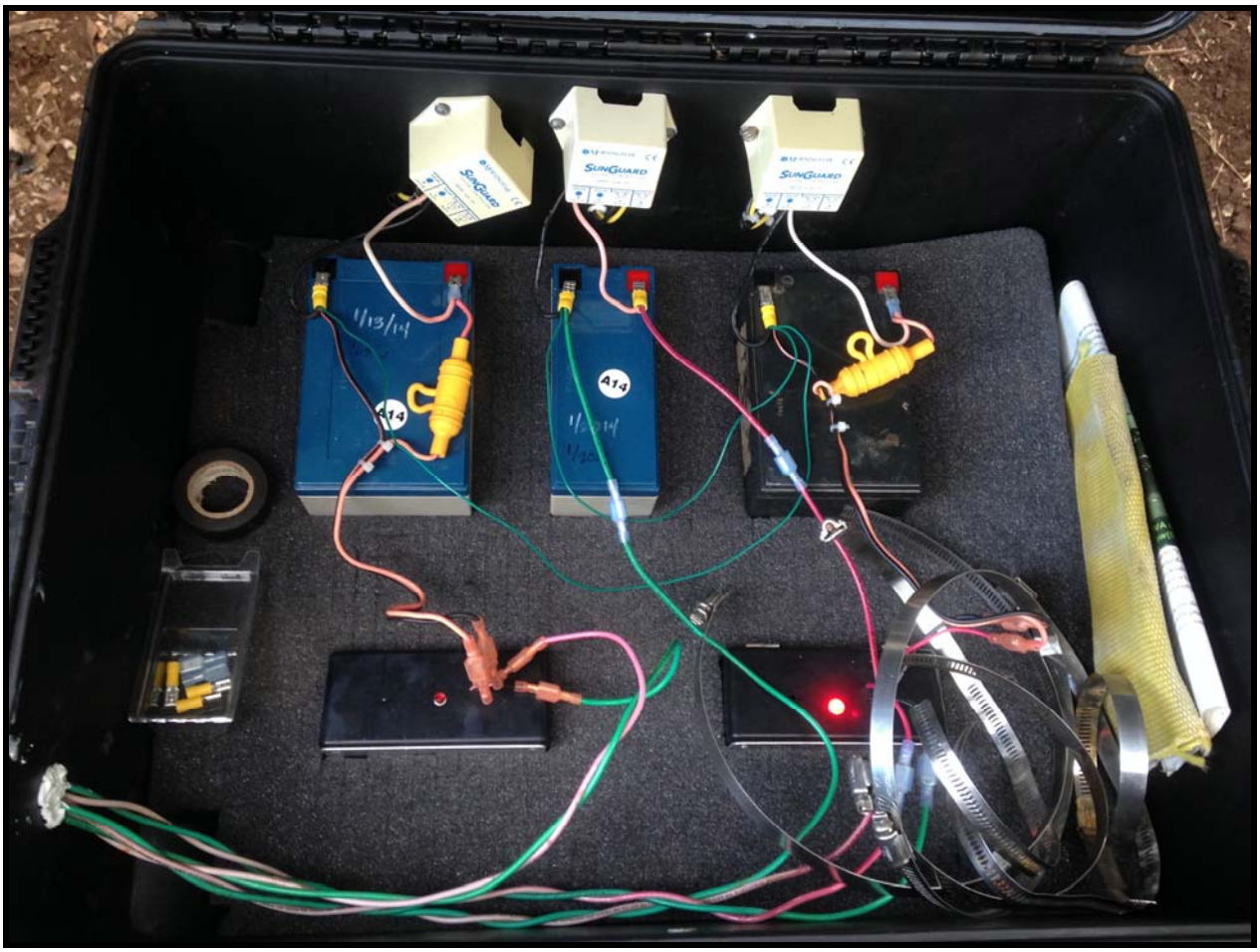

Figure 19. Weather proof housing used for electric components and batteries

The energizers are powered by a 12 Ah, 12 V deep-cycle battery that is connected to a 20 watt solar panel. OANRP estimates that the system can run at maximum capacity (~300 mA, which can only occur if there is a short between the + and – wires) for ~40 hours without sun. At minimum capacity (nothing connecting the + and –), the system draws ~15 mA and can run for ~800 hours without sun. Theoretically, the system can run at full power indefinitely with 5 hours of sun daily. The entire system is stored in a weather proof case and stored in a small structure on site (Figure 19).

Complete Fence

The Xcluder 10-pest fence was used as the backbone for OANRP enclosures. This fence is a 1.3 m fence with a rolled hood and a buried mesh skirt (Fig. 20). Costs for materials and installation are included in Table 4 and based on a competitive bidding process for this specific project. In New Zealand this fence had been proven to exclude small vertebrate pests up to the size of cats (Xcluder pers. comm.). As the only vertebrate pests OANRP needed to exclude were rats, mice, and Jackson's chameleons, this 1.3 m fence was adequate. This fence is too short to exclude cats which require a 2 m fence due to their jumping abilities. The fence excludes these small

vertebrates in three ways. First, it is too tall to jump over. Second, the rolled hood at the top cannot be negotiated by small vertebrates. Third, the buried mesh prevents animals from digging under the fence. This design was developed and extensively tested in New Zealand by Xcluder. One additional modification was made to the “10-pest.” Instead of having a mesh section on the lower half of the fence, the lower half was constructed of solid sheeting to exclude small *E. rosea*.

Figure 20. Xcluder 10-pest fence

Table 4. Pest Proof Fence Materials and Installation Costs

Product	Cost	Notes
“10-pest” Xcluder Inc. fencing or comparable	$200 per meter	Includes materials and installation. Prices vary with site accessibility, terrain, size of the job and different companies. Cost figure based on competitive bid.

The three barriers to exclude *E. rosea* were positioned 300 mm above the soil level. This location was chosen as it was just far enough off the ground to allow the angle barrier to work effectively and provide space for staff to monitor and remove *E. rosea* trapped in the angle. It was also 1000 mm from the top of the fence. One meter is beyond a rat's jumping capability (Xcluder staff, pers. comm.). The entire fence (Fig. 21) must also be constructed with great attention to detail to ensure that all parts are fastened tightly together leaving no gaps or edges that could provide an opportunity for rats or Jackson's chameleons to climb the barrier.

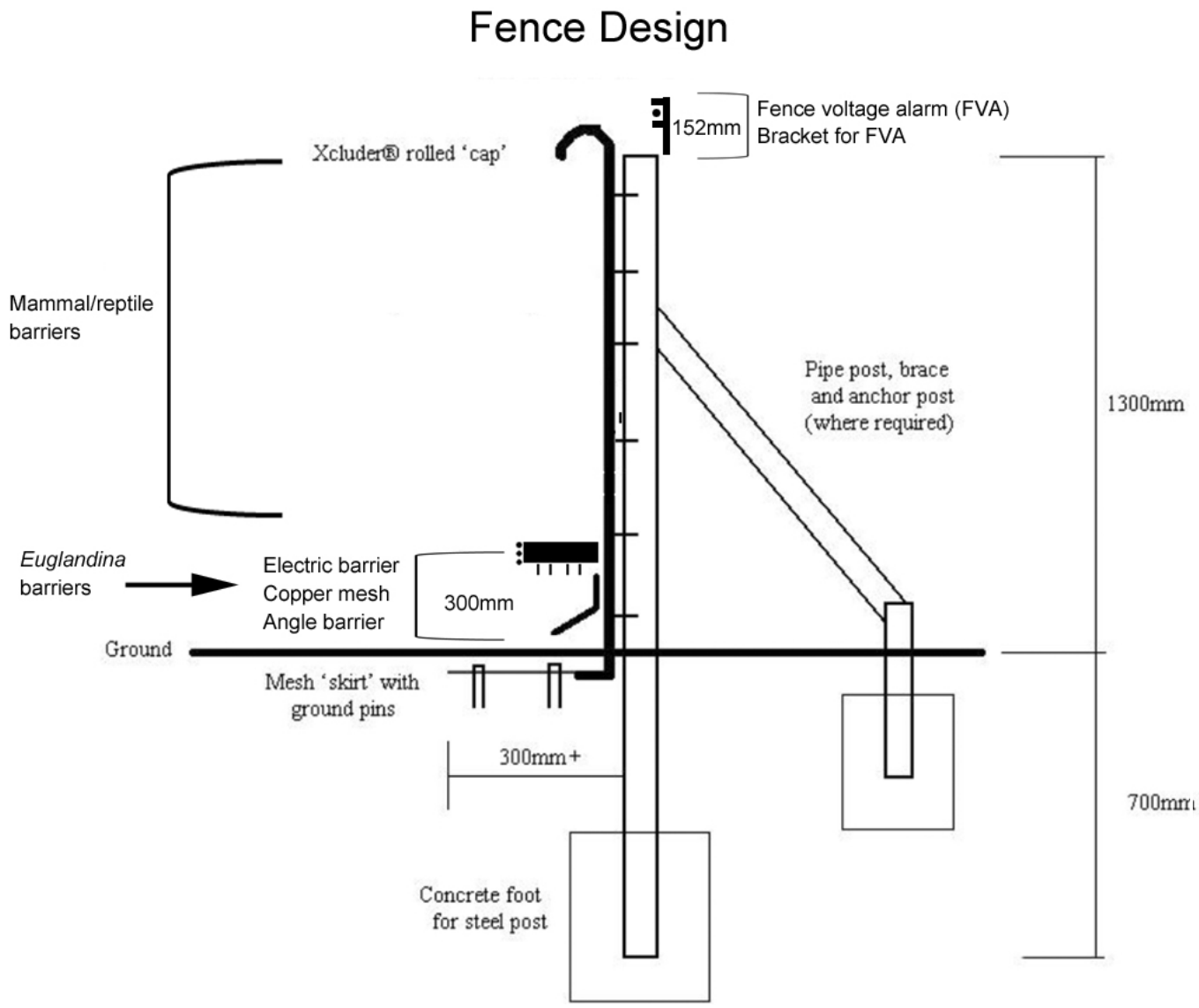

Figure 21: Xcluder "10-pest" fence with *E. rosea* exclusion barriers

Jackson's Chameleons Exclusion. The "10-pest" fence had never been tested against Jackson's Chameleons. However, after studying the morphology of the chameleons, Xcluder was confident that their fence would work. We tested the ability of Jackson's chameleons to climb

the fence to verify Xcluder's opinion. We spent approximately 20 hours observing chameleons tethered to or placed on the barrier. On all attempts, the chameleons did not make any progress up the barrier. Jackson's chameleons have a two-part grasping foot with small claws on the toes. This anatomy serves them well in trees and vegetation. However, on the flat smooth surface of the fence there is nothing to grasp. The only place they could get a grasp was at the fasteners and it was impossible for them to climb the fence on the fasteners. The hood also proved impossible for the lizards to negotiate. If placed on the outside edge, they could barely hold on and it was impossible to move beyond the hood as the upper surface is entirely smooth. Based on these observations and Xcluder's opinion, OANRP staff determined the fence was an effective barrier against Jackson's chameleons.

Fence Integrity Monitoring. It requires hundreds of man hours to remove the *E. rosea* and Jackson's chameleon from within the enclosures. If there was damage to the fence and predators were able to gain access it would again require a massive effort to remove them. As a result, OANRP determined that the best management practice would be to install a remote monitoring system (Fig. 22) with the goal of continuously monitoring the integrity of the enclosure and alerting OANRP when problems occurred. OANRP worked with Intelesense Technologies™ to develop a system to achieve these goals. Intelecell computers are used to collect important diagnostics and transmit this data hourly via fixed Yagi antennae to base stations that then send data to the Internet and make it available to authorized personnel via password-controlled access. In addition, if there are any problems with the fence integrity and barriers, an email is sent to alert OANRP staff. The units are powered by 12-volt batteries and charged by small photovoltaic systems.

Figure 22. Remote Monitoring System Components

Debris alarm monitoring. The most devastating event that could occur at an enclosure is a significant disruption to the structural integrity of the barriers, including the 10-pest excluder fence and the three *E. rosea* exclusion barriers. A tree fall would be the most likely cause of such a disruption. To guard against such an occurrence, the enclosure is equipped with a fence voltage alarm (FVA). This is a standard electric fence monitoring device that is used commercially to ensure the integrity of electric livestock fences. This system is made up of three components: a standard fence energizer unit (OANRP uses a Gallagher S50™); a FVA monitoring unit (also available from Gallagher); and a standard electric fence wire and insulators. A standard electric fence wire is mounted on 10 cm insulators along the top of the barrier. The wire is energized by the Gallgher S50 and monitored by the FVA. If the fence current is disrupted for more than five seconds then the FVA sends an alarm to the Intelecell. OANRP staff are alerted and work to quickly respond and clear the disruption and ensure no predators breach the enclosure. Figures 23 and 24 are screen captures from the Intelesense website. Figure 23 shows the FVA over four days which reads 1.0 as there are no alarms. In the event of an alarm the graph drops to 0 and an email is sent to alert staff.

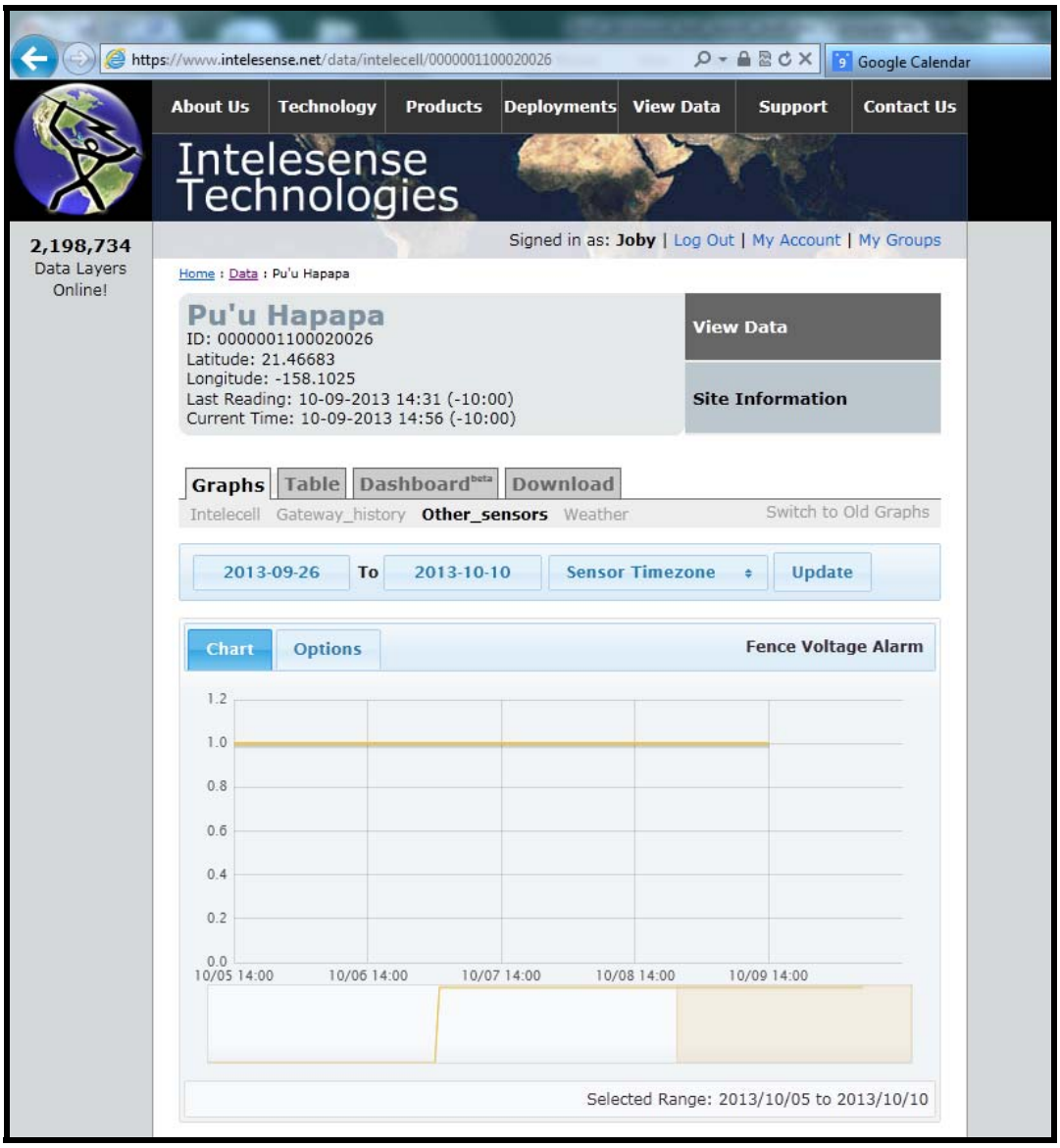

Figure 23. FVA monitoring read out over a four day period showing normal condition.

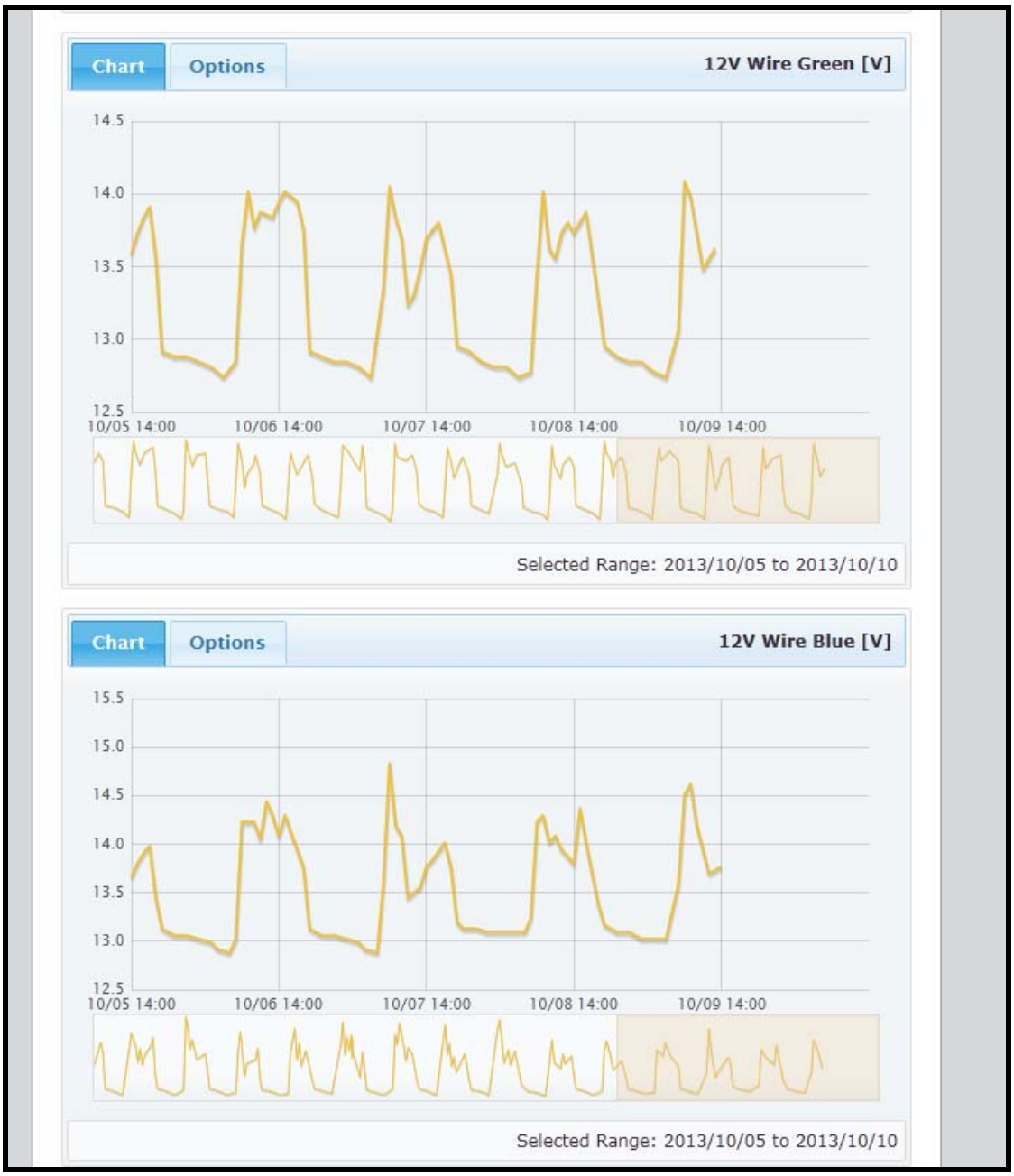

Figure 24. Electric Barrier status

Electric fence monitoring. The second parameter monitored by the remote sensing system is the voltage in the electric snail barrier. As described above there are two independent low voltage systems protecting the enclosure from *E. rosea* (labeled Blue and Green). When the system is up and running correctly it operates around 12-14 volts. If at any time the system drops below 10.5 volts an alarm is triggered and the Intelecell sends an alert. Figure 24 shows the two systems (green and blue wire) with the typical daily pattern. During the day, the voltage is between 13.5 and 14.0 and at night, when the system runs just on battery, it is between 12.5 and 13.0. The cost to build the fence monitoring system are shown in Table 5.

Table 5. Fence Monitoring System Costs

Product	Cost	Notes
Fence Voltage Alarm	$479.99	Ordered from the U.S. mainland
Fence Energizer (Gallagher S50)	$510.99	Ordered from the U.S. mainland
Fence insulators	$20 for a bag of 30	Sometimes available locally. One fastener per meter.
16 gauge electric fence wire	$8.75 for 50 m	Available locally
Intelecell	$2,500	Only available from Intelesense Technologies
Solar panel, 20 watt	$76	Available locally
Weather station	$5,300	Prices vary depending on manufacturer and model. OANRP had the best success using systems without moving parts
Battery	$45 each	12V 12Ah sealed lead acid battery.

Fence Management

Introduction and Enclosure Background. In the following section, our experience with the construction of the enclosure at Puʻu Hapapa is used to describe the process that OANRP followed to construct the enclosure, remove the predators, reintroduce and monitor *A. mustelina*, and conduct site restoration.

Puʻu Hapapa was chosen as the first site to construct the new enclosures beyond early prototypes in Kahanahaiki and Pahole. The site, near the summit of Puʻu Hapapa at 2,600 feet, is a mix of native and introduced vegetation protected from ungulates by fencing (Fig. 25). The vegetation is predominantly native consisting of several large *Pisonia umbellifera* (Pāpala kēpau) trees with *Pipturus albidus* (Mamaki) shrub understory and the scandent *Freycinetia arborea* (ʻieʻie) and with scattered weeds, principally *Schinus terebinthifolius* (Christmasberry), the dominant tree in the surrounding areas. The humidity at Puʻu Hapapa often reaches 100% at night. Rainfall averages 1182 mm per year (Online Rainfall Atlas of Hawaiʻi). The area is intensively managed, with ongoing weed control, outplanting and monitoring of endangered species.

Figure 25. Aerial view of Pu‘u Hapapa enclosure looking northeast. Note the highly eroded landscape offering very few opportunities for construction of such enclosures.

The area where the Waieli and Kalua‘a Gulches come together below Pu‘u Hapapa in the Wai‘anae Mountains on O‘ahu has been a known hot spot for snails dating back to the 1800s. In 2000 the Army contracted botanists Steve Perlman and Ken Wood to perform rare plant surveys in the area. When Ken returned from a long day in the field he spoke not about the plants he had found that day but about the snails. He described how he saw a branch of ‘ie‘ie that must have had 30 snails on it – not just the endangered *Achatinella mustelina* but rarer snails including *Laminella sanguinea*, *Amastra micans*, and *Cookeconcha* as well as common native snails such as *Auriculella*, Tornatellinids, *Philonesia*, and succineids. In each ‘ie‘ie patch there must have been 30 branches with 30 snails on each branch (900 snails) and that there must have been ten ‘ie‘ie patches (9,000 snails) and probably another 1000 snails in between the ten ‘ie‘ie patches. He said there must be at least 10,000 snails up there. His calculations were impressive and before we had even gone there to survey specifically for snails we had already started calling it the Land of 10,000 Snails.

In January 2009, OANRP observed *E. rosea* densities increasing beyond levels seen in previous years. During an overnight camping trip staff collected a total of 50 *E. rosea*. Such a high number was unprecedented. A total of 221 were collected that year. At first these numbers

were staggering but this was just the beginning (Fig. 26). In 2010 a lot of time was spent clearing the fenceline to construct the enclosure so less time was given to collecting *E. rosea* and only 195 were found. By 2011 when the fenceline was complete more time was devoted to *E. rosea* control and 645 were collected. In 2012 *A. mustelina* were released from the HTSCL and a lot of time was spent searching inside the enclosure for the last few *E. rosea*. It took many hours to find a small number of snails. A total of 122 were found during the entire year. By 2013 *E. rosea* was absent inside the enclosure and more time again was devoted to collecting *E. rosea* from outside the enclosure. A total of 526 were found that year. Similarly, a total of 594 were found in 2014.

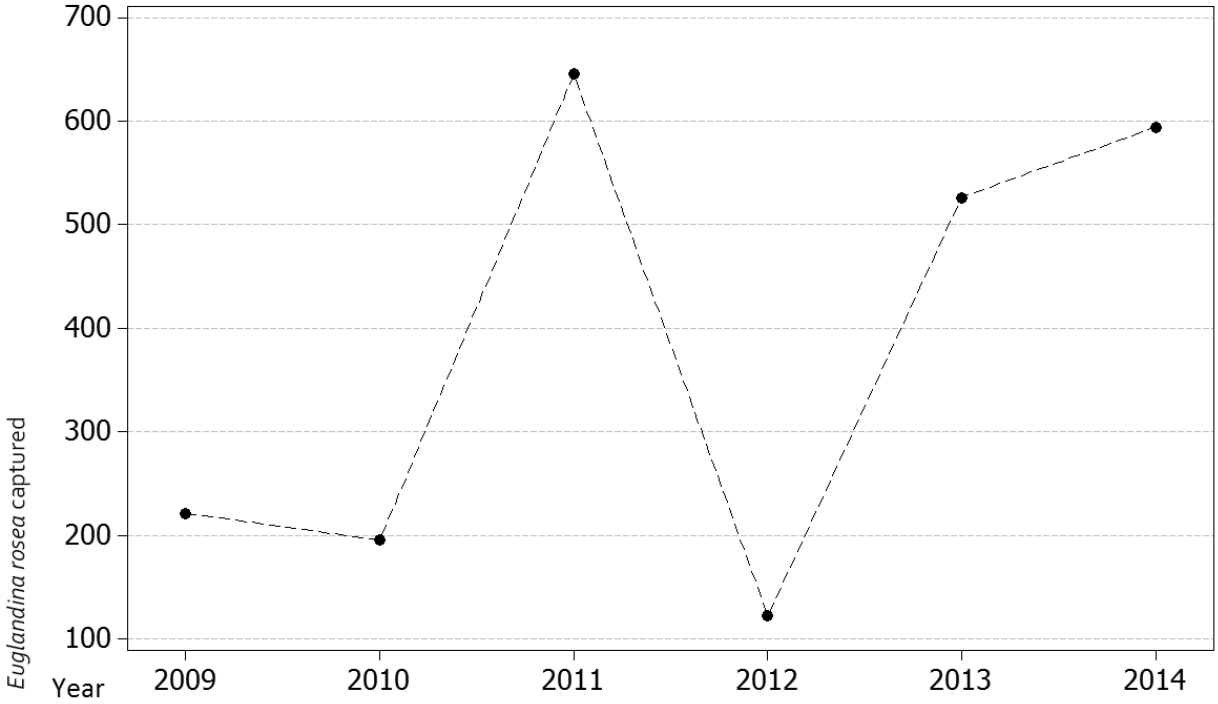

Figure 26. *Euglandina rosea* captured over time at Pu‘u Hapapa

Prior to 2009 the numbers of *A. mustelina* were declining steadily (Fig. 27). But as a result of the sharp decline by December 2009, OANRP began removing *E. rosea* from the area and moving *A. mustelina* to the HTSCL until an enclosure could be constructed.

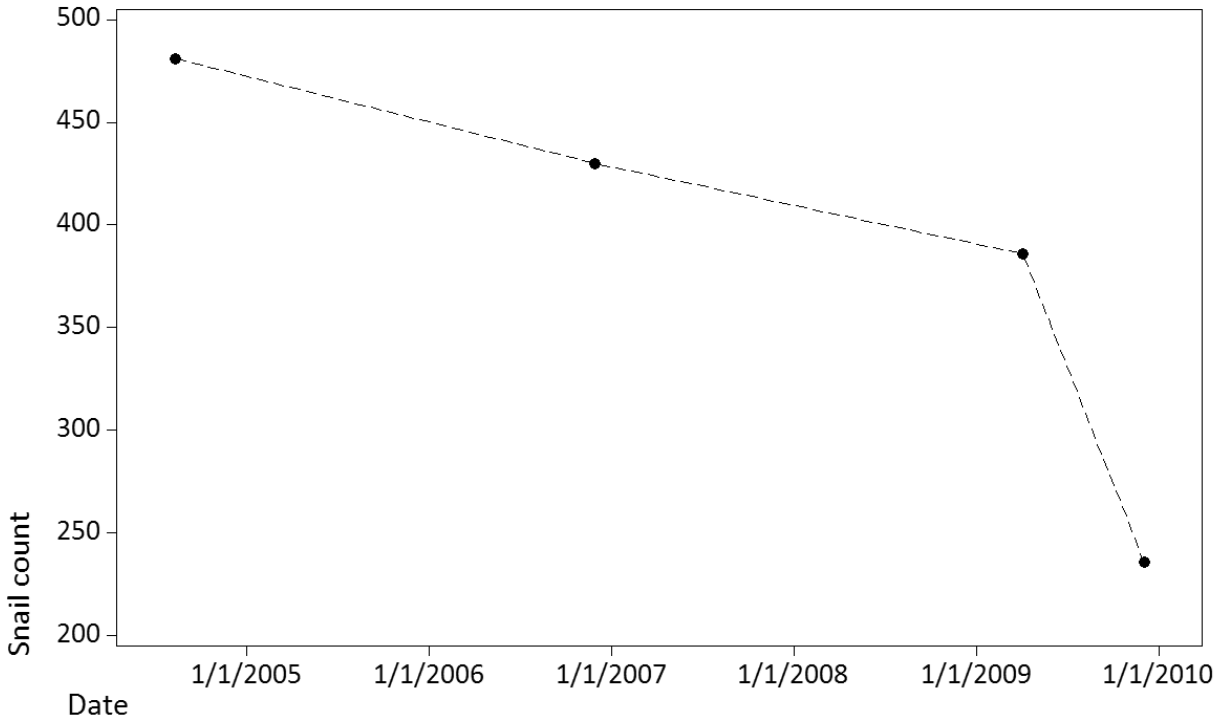

Figure 27. *Achatinella mustelina* counts over time at Pu‘u Hapapa

The relatively flat, non-rocky terrain at Pu‘u Hapapa made it a good spot to attempt the first enclosure (Fig. 25). This terrain made for much easier construction. It would be extremely difficult to build this type of enclosure in steep areas. Difficulties include the large amount and type of material (long sheets of metal) required to be maneuvered into place, the concrete footings that are required, requirements for very tight construction and the biggest issue, the long term maintenance of the interface between the ground and the fence. Erosion is an essential consideration for fence construction because the bottom edge of the fence must remain below the surface. Installation of the fence also changes the patterns of runoff and soil movement in the area both within and outside the enclosure.

At Pu‘u Hapapa OANRP spent over one thousand hours clearing the proposed 160 m fence line. It was densely overgrown with *Schinus terebinthifolius.* The Nature Conservancy (TNC) had previously planted important host plants for the tree snails under the *Schinus*. Protecting them whilst removing the *Schinus* took an enormous amount of time. The construction of the enclosure was contracted for approximately $100,000 to Xcluder Pest Proof Fencing Company from New Zealand. A crew of three to four Xcluder staff from New Zealand worked over four separate trips to construct the enclosure starting in the summer of 2011. OANRP completed the final details on the barriers in December of 2011. The electric fence was the final barrier to be installed requiring multiple prototypes. The enclosure protects approximately 1,564 m^2.

Threat Control

Major threats to *A. mustelina* at Pu‘u Hapapa include rodents, specifically the black rat (*Rattus rattus*), rosy wolfsnail (*E. rosea*) and Jackson’s chameleons (*Chamaeleo jacksonii* subsp. x*antholophus*). All three predators have been successfully eradicated from the enclosure since September of 2012. Rodents were quickly removed using traps and baits soon after the completion of the enclosure in March 2011. Jackson’s chameleons and *E. rosea* were much more difficult to remove requiring 786 staff hours of searching both during the day and at night since December 2011. We discuss aspects of managing these predators below.

Rodent eradication efforts at the Pu‘u Hapapa enclosure. Rodent eradication from within the enclosure required the least amount of effort compared with other threats. OANRP used Victor™ snap traps and diphacinone rodent bait in tamper proof dispensers to remove rats. Monitoring efforts to ensure that rats were removed included tracking tunnels baited with peanut butter and wax chew tabs. Rodent control has been continuous across the area for many years and no rats were detected within the enclosure once complete. As mentioned, the Xcluder 10-Pest fence is a proven barrier to rodents. Four maintenance considerations must be continuously followed to ensure that rodents do not breach the enclosure.

- First and foremost, the vegetation along the outside edge must be continually trimmed back to prevent rats jumping from adjacent trees and shrubs. OANRP uses the following guidelines to trim vegetation. Nothing must be within 2 m of the fence at the hood level. In areas with taller canopy OANRP ensure that there is no vegetation within 3-4 m of the fence, increasing the distance cleared with the height of the vegetation.
- Second, the buried mesh must always be securely fastened to the bottom edge of the fence and any damage must be quickly repaired to ensure there is no possibility for rodents to burrow under.
- Third, the hood structure and vertical wall must be inspected regularly to ensure that they are secure. Of particular concern are the unions where sheets overlap.
- Finally, rodent monitoring must be conducted consistently within the enclosure. OANRP conducts monitoring with snap traps and tracking tunnels quarterly.

Jackson’s chameleon eradication efforts at the Pu‘u Hapapa enclosure. Jackson’s chameleons are reproductively active at least twice a year in Hawai‘i, in December and February (Goldberg & Kraus, 2011). Gestation lasts from 6-9 months allowing a potential maximum of two clutches per year. One clutch per year is more common (Jacksons Chameleon Reproduction, Rearick *et. al*). Litter sizes are, on average, 12 (range 7-21) in Hawai‘i, with larger females able to produce more young. The growth rates are shown in Table 6. For males, the minimum size at first reproduction was smaller in Hawai‘i than in their native habitat (Kenya). Here, males measuring 70 mm snout to vent length (SVL) had mature gonads while in Kenya they needed to reach 90 mm. For females, it was the reverse, with the smallest gravid female found in Hawai‘i measuring 94 SVL compared to only 80 SVL in Kenya (Lin & Nelson 1980).

Table 6. Chameleon growth rates derived from Goldberg & Kraus (2011).

Age class	Description	Size	Age
Newborn	Sexes indistinguishable	<29 mm	<1 month
Juvenile	Premature horns present on males, tails thickened	30-69 mm	1-4 months
Adult male	sexually mature	>70 SVL	5-9 months
Adult female	sexually mature	>94 SVL	9-12 months

Since December 2011, when the enclosure was completed and sealed from predator incursion, OANRP staff have removed 30 chameleons. Dissection revealed one of the 30 had consumed a single *Achatinella* within the previous week. Although no further *Achatinella* were found in gut contents, others may have been consumed but not detected. The last chameleon found inside the enclosure was an adult male (84 mm SVL) removed on 20 August 2012 (Fig. 28). No chameleons have been detected since this date, despite repeated searching (Table 7).

Table 7. Chameleon capture in Pu‘u Hapapa enclosure since closure.

Year	Month	Search effort (person hours)	Chameleons found
2011	Dec.	40	6
2012	Jan.	108	5
2012	Feb.	2	1
2012	Mar.	7	0
2012	Apr.	10	2
2012	May	20	2
2012	Jun.	46	8
2012	July	42	5
2012	Aug.	35	1
2012	Sep.	35	0
2013	Feb.	6	0
2013	Mar.	4	0
2013	Apr.	5	0
2013	May	3	0
2013	Jun.	2	0
2013	Aug.	7	0
2013	Sept.	1	0
Total		373	30

Figure 28. One of the last chameleons removed from the Pu‘u Hapapa enclosure

The last mature female in the enclosure was removed on 30 Jan. 2012 (101.6 mm SVL). It was one of only two mature females found (the first was removed 28 December 2011). Dissection confirmed neither was pregnant at the time of capture. Juveniles found following their capture, based on size (Table 6) appear to be cohorts from one mother (Fig. 29). No new chameleons have been captured since August 2012.

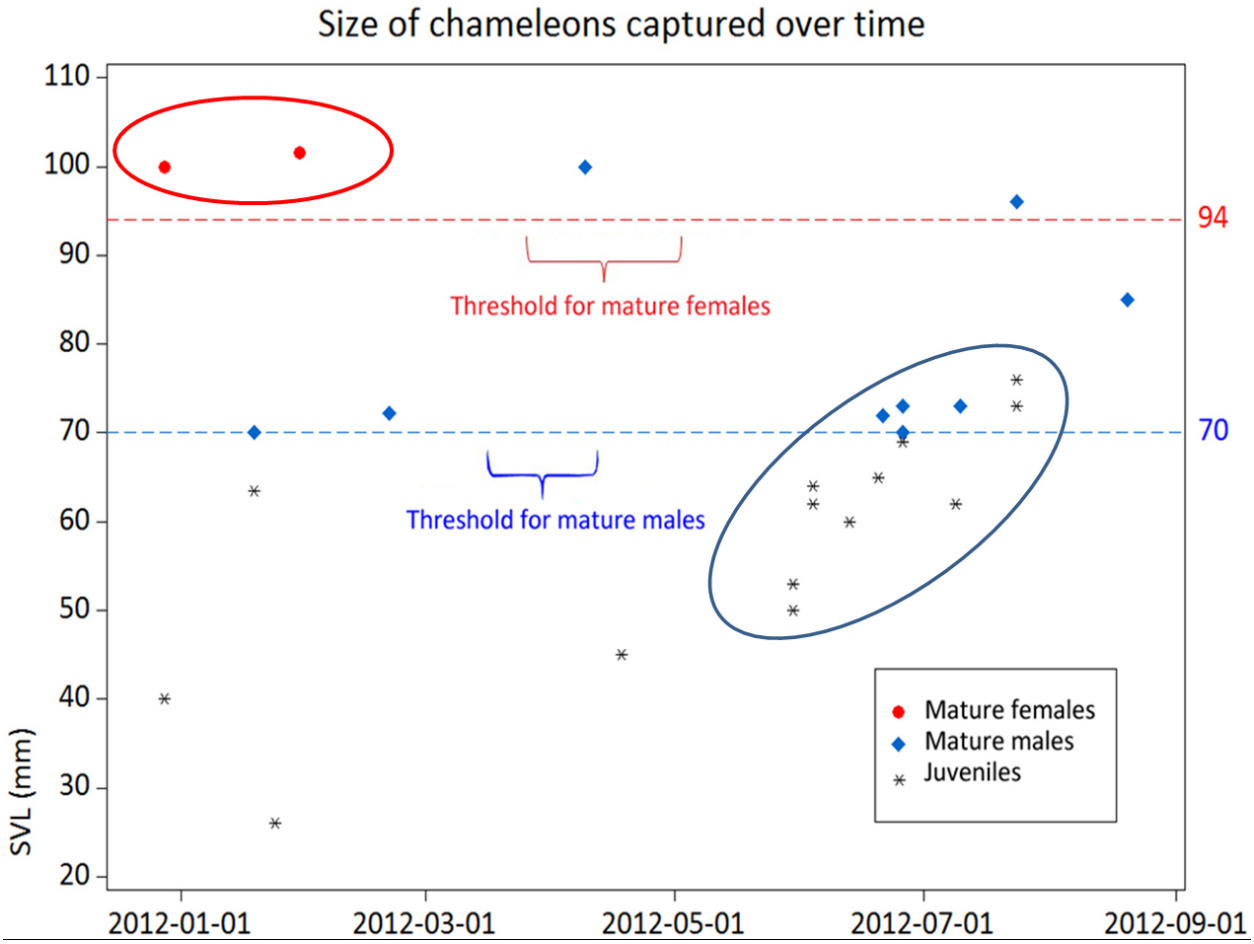

Figure 29. The similar size of the juveniles (circle lower right) captured after the last mature female was removed, suggests these may be siblings born from one of the females (circle upper left).

Challenges. Chameleons are cryptic (Fig. 30). Despite all the work that took place, we only became aware that they were established in the area in 2011. Given the size of the enclosure (1,564 m^2) and the number of animals removed, they were present at a possible density of 1 per 50 m^2 but were probably much higher. Some of the trees are 12-15 m tall and were difficult to search. Tree climbing techniques were used to access high canopies both for day and night searching.

Figure 30. A newborn chameleon (on an AA battery for size reference) found in the enclosure.

***Euglandina rosea* eradication efforts**

***E. rosea* reproductive biology:**

The following data are all taken from Jerlach (1994). Under optimal laboratory conditions (between 25 and 30^{o} C and full feeding) *E. rosea* can reach sexual maturity in 263 days.

Table 8. *E. rosea* size classes observed to maturity and at a minimum size of 35.4 mm (Jerlach 1994).

Age class	Description	Size	Age
Hatchling	prior to shell thickening	<10 mm	0-41 days
Juvenile	thickened shell, immature	10-30mm	42-311 days
Subadult	sexually mature, not full grown	35-40mm	312-460 days
Adult	full grown	>40mm	460-550 days

Typically, they do not lay eggs until they were over 40 mm SVL and were 386 days old. Generally, nine eggs are produced per clutch and these hatch in 31 days. All eggs hatch at temperatures above 10^{o} C. Based on this growth rate data, the snails can be broken down into hatchling, juvenile, subadult and adult categories (Table 8). This data was used to inform us on whether the snails discovered were reproductive.

***Euglandina rosea* feeding**: Jerlach (1994) found adults preferentially consumed 100% of prey offered at the smallest size class (<15 mm including the shell). They consumed 80% of prey between 16-20 mm and 40% of prey between 21-30 mm. This preference was found to be flexible (Meyer & Cowie 2010). Prey that was formerly rejected when paired with smaller prey was later consumed when paired with even larger prey.

***Euglandina rosea* removal effort:** Below we describe four levels of *E. rosea* control; each was to be triggered under varied conditions outlined in a flow chart (Fig. 31). This effort was not achieved at all times, but served nonetheless as a guideline. Most searches for *E. rosea* took place during the day while a few occurred at night. The snails are easier to find during the day.

Highest removal effort = severe risk of *E. rosea* in enclosure: Three staff spend two days a week at 4 hours per day for 4 weeks. This would total to 24 hours of search time per week, 96 hours total for the month.

High removal effort = high risk of *E. rosea* in enclosure*:* Three staff dedicate one day a week for 4 hours per day for 4 weeks. This would total to 12 hours of search time per week, 48 hours total for the month.

Medium removal effort = some risk of *E. rosea* in enclosure: Three staff dedicate one day per month at 4 hours per day for 4 months. This would total 12 hours search time per month.

Lowest removal effort = low risk of *E. rosea* in enclosure: Between 2 and 3 staff dedicate a total of 10 staff hours one day every 3 months to sweep interior.

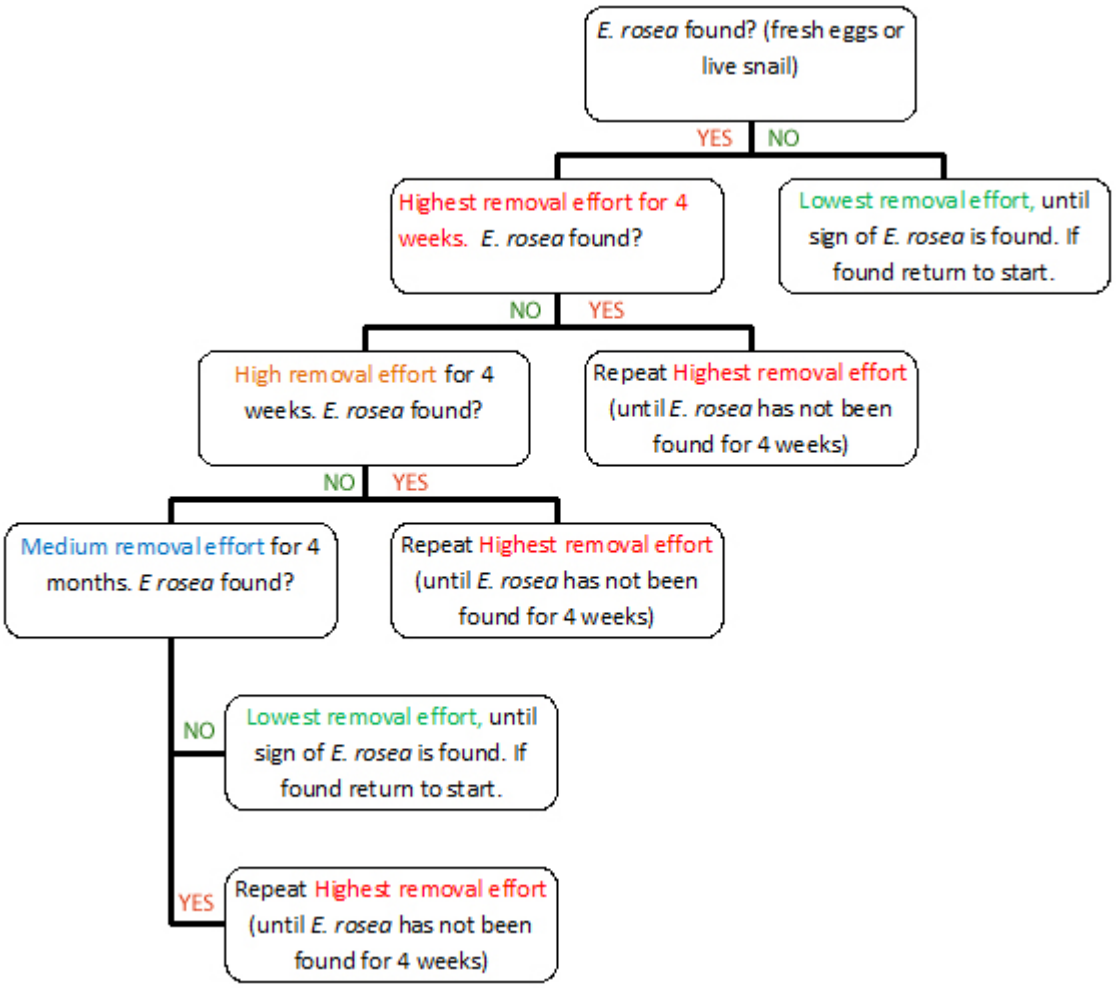

Figure 31. *Euglandina rosea* removal flowchart

Results. Over the past five years 1,439 *E. rosea* have been collected on the Pu'u Hapapa Bench. Forty six of these were collected inside the enclosure (after its construction in late 2011, Table 9). The usual location for these predatory snails is leaf litter. Sometimes they are found on the surface of the ground and sometimes in the vegetation but most often they are hidden under leaves. All of the leaves, sticks and rocks that were on the ground were raked into piles, collected into trash cans and dumped outside of the enclosure. After raking was complete only a few additional *E. rosea* were found. These were probably in the trees and descended to the ground as new leaf litter accumulated. Keeping the leaf litter intact would have contributed to maintaining more moisture inside the enclosure which would have been beneficial for the *Achatinella* but not clearing out the leaf litter would have made it impossible to eradicate *E. rosea*.

Reproductive snails were only found on two occasions (Table 9). In December 2011, three adults were found (not measured, simply recorded as "large") followed by another in July measuring 35 mm. Upon dissection, however, the latter snail did not prove to be reproductive (Holland, *pers. comm*. 2012). The last *E. rosea* removed from the enclosure was a juvenile

measuring 21 mm on 7 August 2012. Since finding that snail, staff have continued to search intensively (Table 9) with no new discoveries.

Table 9. Number of *E. rosea* captured in Pu‘u Hapapa enclosure since closure.

Year	Removal effort based on flowchart	Month	Search effort (person hours)	*E. rosea* found
2011	Lowest	Dec.	7	8*
2012	Highest	Jan.	153	29
2012	Medium	Feb.	15	3
2012	Medium	March	28	3
2012	High	April	47	2
2012	Medium	May	19	0
2012	Lowest	June	5	0
2012	Medium	July	23	3
2012	Medium	Aug.	25	1
2012	Medium	Sept.	31	0
2013	Lowest	Jan.	6	0
2013	Lowest	Feb.	6	0
2013	Lowest	March	1	0
2013	Medium	April	15	0
2013	Lowest	May	3	0
2013	Lowest	June	6	0
2013	Lowest	July	5	0
2013	Medium	Aug.	14	0
2013	Lowest	Sept.	4	0
		Total	413	46

*The collection contained snails large enough to be reproductive.

Euglandina rosea continued to encounter the enclosure over time (Fig. 32). Typically 0-2 snails but on four occasions up to eight were found under the angle barrier indicating that without the barriers the snails might have reached the protected area. As no *E. rosea* have been found inside the enclosure or above the angle barrier since late 2012 we have concluded that the search and eradication protocol is successful and the exclusion barriers are effective.

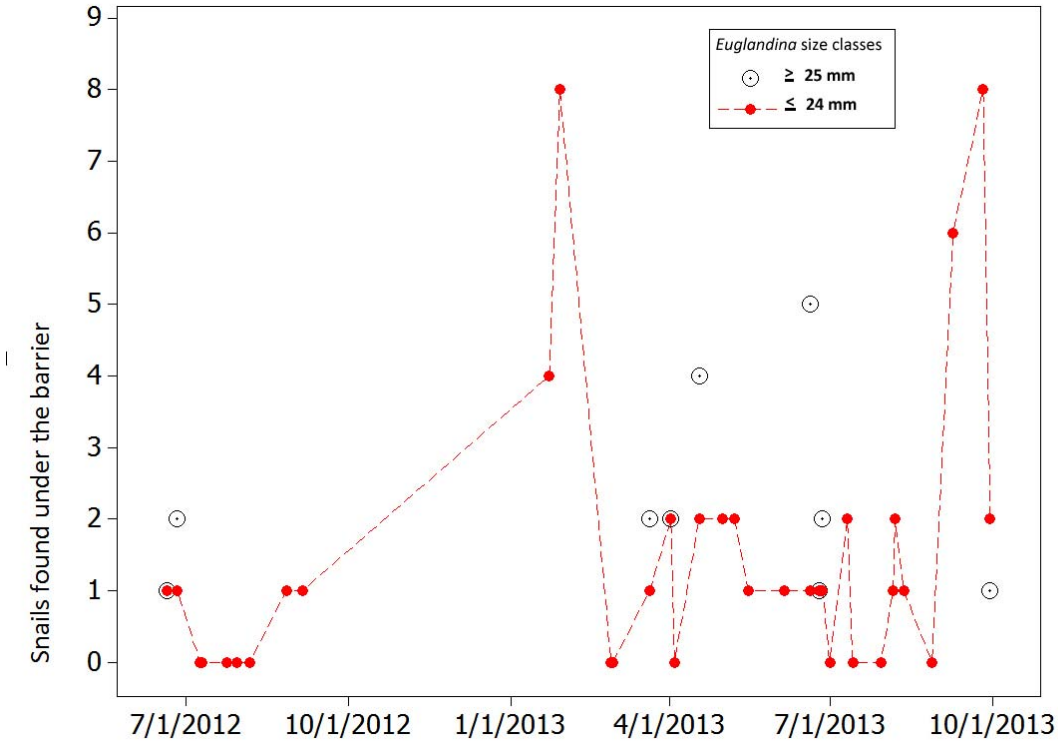

Figure 32. Graph showing the number and size of *E. rosea* found under the angle barrier over time.

The barrier is also effective against other mollusks e.g., slugs. Three introduced species are a serious threat to native plant regeneration. Though the angle barrier is ineffective alone, the bed of nails and the electrical barriers appear to be effective. The prevention of slug ingress is important to enabling native habitat restoration efforts within the enclosure.

HABITAT RESTORATION

Achatinella mustelina reintroduction within the Pu‘u Hapapa enclosure

A total of 202 *A. mustelina* were collected from the surrounding vegetation at Pu‘u Hapapa between February 18, 2010 and May 26, 2010, while the snail enclosure was being developed and taken to the HTSCL for safe keeping. Enclosure construction started one year and 3 months later and by then the snail population in the laboratory had increased to 340 snails; a 68% increase in number. However, many of the mature snails where lost over this period and the population consisted of mostly immature snails.

When the snails were transported from HTSCL great care was taken to ensure there was as little stress as possible. A specially made terrarium was used that provided adequate space and air flow (Figure 33). Snails were driven directly from the laboratory to a landing zone from which they were flown in a helicopter to the Pu‘u Hapapa release site.

Figure 33. OANRP Rare Snail Conservation Specialist with *A. mustelina* from HTSCL ready for release at Pu‘u Hapapa

Once at Pu‘u Hapapa the snails were carefully removed from the containers and placed in screen baskets in the trees (Fig. 34). The baskets were open at the top to allow the snails to exit into the trees. The screened baskets were kept moist by spraying water on them until all the snails had exited. The snails were monitored during the afternoon to ensure that they moved out of the baskets into the trees. The releases occurred in the mid-afternoon on cool winter days to reduce heat stress.

Figure 34. Photo of the screen basket used to release *Achatinella mustelina* into the enclosure at Pu‘u Hapapa.

Most of the snails from the HTSCL were reintroduced into the new enclosure in two efforts: The first on February 8, 2012 when 171 snails were released, and the final 169 on February 21. On two later dates small numbers from HTSCL originally collected from Pu‘u Hapapa were released as well. It is impossible to know precisely how many of the snails released from the lab were originally from Pu‘u Hapapa or were born in the laboratory because the snails collected were not marked in the laboratory. However, a large proportion of the snails returned were laboratory-born and immature.

Table 10. *Achatinella mustelina* introduced into Pu‘u Hapapa enclosure.

Date	From	Small	Medium	Large	Total
Jan 3, 2012	Extant within enclosure	4	6	14	24
Jan 4, 2012	Wild pop.	4	7	14	25
Jan 30, 2012	Wild pop.	2	13	11	26
Feb 8, 2012	HTSCL	109	9	53	171
Feb 8, 2012	Wild pop.	2	17	5	24
Feb 21, 2012	HTSCL	106	63	0	169
Mar 7, 2012	Wild pop.	1	3	5	9
Apr 11, 2012	Wild pop.	7	7	5	19
Nov 29, 2012	Wild pop.	0	1	3	4
Dec 31, 2012	HTSCL	0	3	4	7
Jan 23, 2013	Wild pop.	8	23	25	56
Feb 26, 2013	Wild pop.	9	36	27	72
Feb 27, 2013	Wild pop.	16	15	28	59
Mar 20, 2013	Wild pop.	2	4	8	14
Apr 2, 2013	Wild pop.	2	4	6	12
Apr 3, 2013	Wild pop.	0	1	2	3
Apr 4, 2013	Wild pop.	1	6	5	12
Apr 24, 2013	Wild pop.	3	15	11	29
May 1, 2013	Wild pop.	1	0	0	1
May 8, 2013	HTSCL	1	1	8	10
May 9, 2013	Wild pop.	26	135	122	283
May 10, 2013	Wild pop.	8	23	22	53
Jun 27, 2013	Wild pop.	0	1	0	1
Jul 30, 2013	Wild pop.	0	0	5	5
Aug 12, 2013	Wild pop.	3	5	3	11
Nov 5, 2013	KAL-A	3	10	11	24
Nov 26, 2013	KAL-A	13	19	20	52
Dec 11, 2013	KAL-A	53	99	143	295
Dec 18, 2013	KAL-A	4	12	7	23
Mar 5, 2014	ELI-A	2	10	22	34
Mar 6, 2014	KAL-B	1	44	37	82
Mar 7, 2014	KAL-A	0	2	0	2
Mar 8, 2014	KAL-F	11	11	47	69
Mar 9, 2014	KAL-A	1	2	8	11
Mar 10, 2014	KAL-D	3	19	22	44
Total		406	626	703	1735

On May 8, 2013, a further 283 snails were collected, most from north of Pu‘u Hapapa, and released into the enclosure. A total of 1735 snails have been introduced into the enclosure (Table 10).

Post-Release *A. mustelina* Monitoring. Ground shell plot and timed-count monitoring protocols were developed for the Pu‘u Hapapa ESU to track mortality following the initial reintroduction efforts and assess if intensive threat control was sufficient to reverse negative population trends. The following discussion outlines the monitoring methodologies and preliminary data analysis following reintroduction efforts, from February 2013 through July 2014.

Monitoring Protocols. *Ground Shell Plot Monitoring:* The U.S. Fish and Wildlife Service (USFWS) developed and initiated a ground shell plot monitoring protocol to detect mortality of the laboratory snails following reintroduction. Two ground shell plots were selected as the sampling units. They were installed directly below the core reintroduction zones (Fig. 35). Plot 1 was divided into six 19 m^2 quadrats, and Plot 2 was divided into nine 24 m^2 quadrats. The plots were permanently marked using PVC pipe to delineate the boundaries of each quadrat. Each plot was searched by biologists on hands and knees for approximately 30 minutes per quadrat. The plots were monitored once a week for a total of eight weeks following the intial release efforts. By April 2012, FWS detected 6% mortality of the reintroduced snails. It was decided this rate of mortality was not of concern, and the monitoring interval for OANRP was changed to quarterly. The first quarterly interval was implemented in June 2012 to correspond with the timed-count surveys. For ease of comparison between the initial surveys and the quarterly surveys, all mortality detected prior to June was summed together.

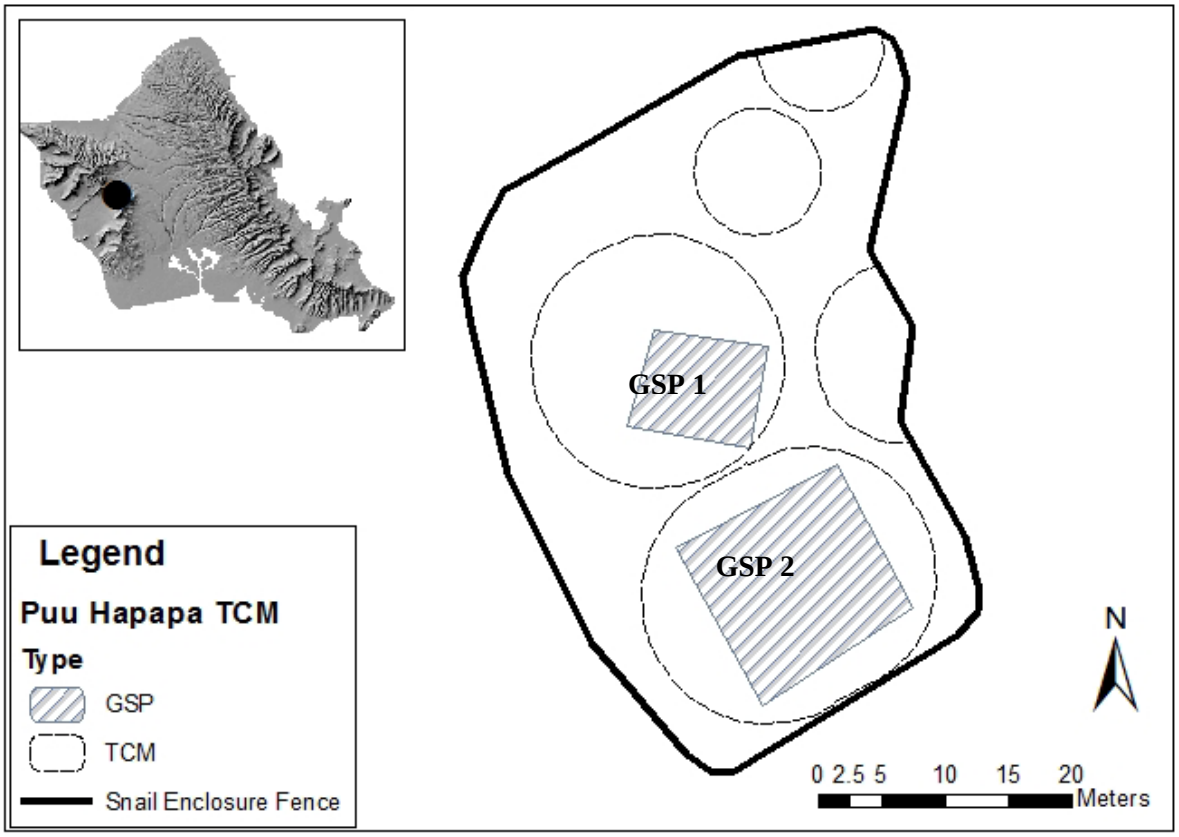

Figure 35. Map of monitoring zones at Pu‘u Hapapa snail enclosure (GSP: ground shell plot; TMC: timed-count monitoring plot).

By July of 2014, a total of 194 shells (including all size classes) were detected in the ground shell plots. The smallest size class was harder to detect than larger ones, and some mortality could have been missed. No evidence of rat predation was observed among the shells collected from the plots. Mortality rates were calculated from the total number of ground shells divided by the population estimate. Population estimates were derived from the total number of snails added to the enclosure minus the total number of snail shells recovered in ground shell plots at a given time. The population estimates do not account for the number of births or unknown deaths, though these values are presumed to be relatively small. Nine percent mortality was detected in the first four months after the initial reintroduction (Fig. 36). After this time, the quarterly mortality rate varied, ranging from less than one, to five percent, with the lowest mortality in April and July 2014. Initial higher mortality, though likely due in part by the longer time interval preceding ground shell plot monitoring (four months) as compared with subsequent monitoring (three months between monitoring events), could also be attributed to the inclusion of laboratory-reared snails (33%) among the first series of augmentations, as compared with introductions after June 2012 that only included wild snails (Table 10).

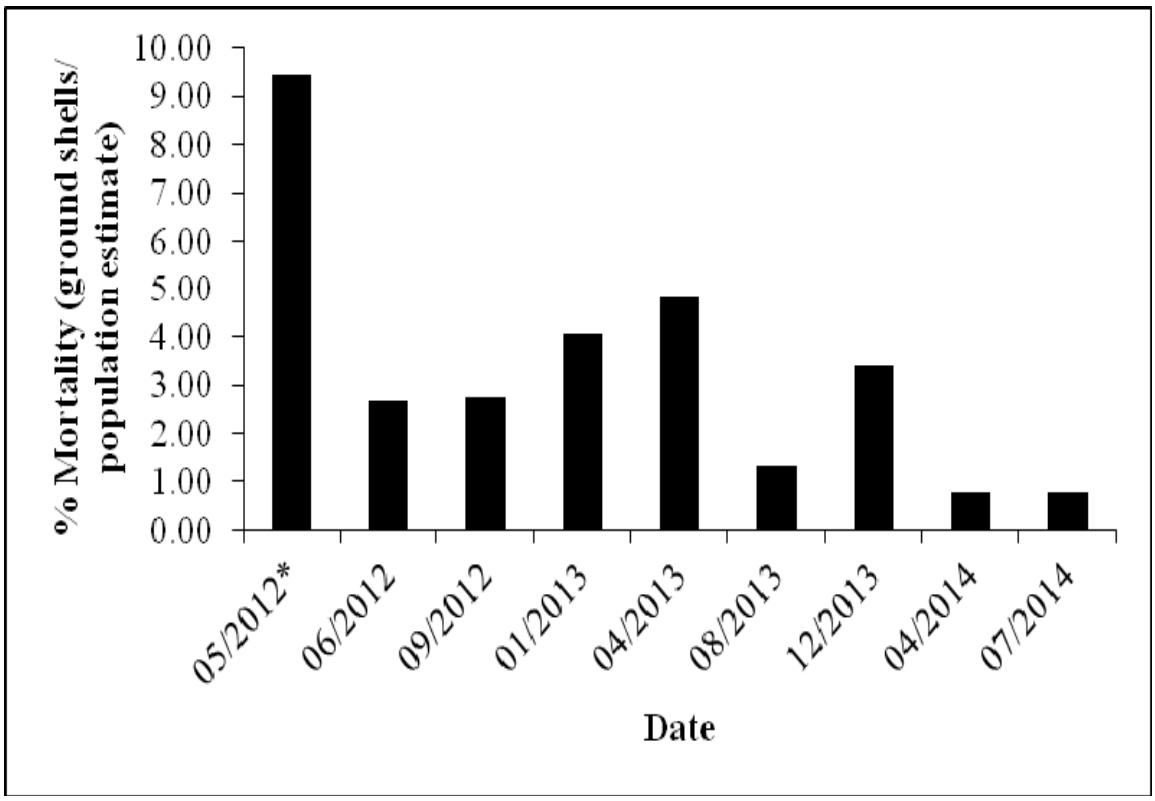

Figure 36. Mortality rates for *A. mustelina* detected in ground shell plots at Pu'u Hapapa. Population estimate is derived from total number of augmented snails minus total shells recovered from ground shell plots. *FWS and OANRP ground shell plot monitoring from February to May 2012; all other dates represent quarterly intervals.

Timed-Count Monitoring: To quantify long-term population trends and assess if the reintroducted population was self-sustaining over time, OANRP implemented a timed-count monitoring methodology. Due to an extremely patchy distribution of *A. mustelina* within the enclosure (as snails were released into five small areas) and relatively small population size, standard quadrat plots and belt transect methodologies had limited statisical power for detecting long-term trends. Given this, a timed-count survey methodology was determined to be the most appropiate monitoring technique. To standardize search efforts, five plots were established in the highest density areas (corresponding with the snail release zones) within the enclosure. Vegetation gaps in the canopy stratum were choosen as natural plot boundaries for deliniation purposes, eliminating the need to incorporate immigration/migration into the current data analysis and synthesis. As the vegetation gaps between survey plots fill in with native canopy over time, it is anticipated that snails will emigrate into new locations within the enclosure. The influence of emigration on the timed-count data is expected to be minimal, as known emigration rates are low for *A. mustelina* (ranging from 0.7% to 4.6%) (Hall *et al*., 2010). Over time, once the change in canopy cover stabilizes, it is presumed that emigration/migration rates will become equivocal, due to the artificial barrier of the enclosure wall.

In June of 2012, the timed-count plots were established and baseline surveys were conducted. Each plot was systematically surveyed by a team of two people, and the total number of snails

within each plot was recorded by size-class (small: < 8 mm; medium: 8-18 mm; and large: > 18 mm). The length of time necessary to survey each zone was established during the first timed-count survey. To ensure consistency between survey periods, a minimum of one person on each monitoring team had previous experience conducting the timed-count. The size-class and location of each snail was recorded and communicated between the surveyors to minimize double counting. Spot lights and binoculars were utilized to detect and accurately identify snails in the upper canopy. Timed-count surveys were conducted three times per quarter over three consecutive weeks from June 2012 to August 2013, then once quarterly beginning in December 2013. Because lower than expected numbers of snails were found during the December 2013 survey, an additional survey was conducted in January 2014.

Because *A. mustelina* were continually introduced into the enclosure from June 2012 to March 2014, it was not feasible to assess if the population was stable and self-replacing. The recurrent augmentation of snails also prevented the program from separating the effect of population recruitment from augmentation. It did, however, enable OANRP managers to verify that the timed-count monitoring technique was a robust method for detecting a true change in the total population size using correlation analyses, and to calculate the detection rate of snails in the enclosure.

There was a significant correlation between the estimated total number of snails within the enclosure (the total number of snails augmented into the population minus the total number of shells recovered from ground shell plots) and the number of snails detected during timed-count surveys (r^2 = 90.0%, $p < 0.001$) (Fig. 37). The detection rate (the timed-count total divided by the population estimate) ranged from 17 to 39%, with a mean of 27%, and a 95% confidence interval for the mean of 24% to 30% (Fig. 38). Repeating the timed-count surveys three times per quarter helped control for variability in detection rates. The highest detection value was observed during a timed-count survey that was implemented one day after snails had been moved into the enclosure and placed in the lower branches of a *Freycinetia arborea*. The monitoring field crew believed the augmented snails did not have time to disperse, making them easier to detect during timed-count surveys and most likely contributed to the high rate of detection during that survey period. The lowest detection values coincided with the night with the lowest relative humidity. The canopy vegetation has become considerably denser since the initial clearing of non-native vegetation from the reintroduction site, which could make snails more difficult to detect. Mean hourly Intelesense Technologies measurements for air temperature, relative humidity, wind speed, wind direction, rainfall and wind gust speed during the 17 monitoring events between June 2012 and July 2014 did not vary greatly among observations (Fig. 39). These variables did not clearly influence detection rates (Table 11).

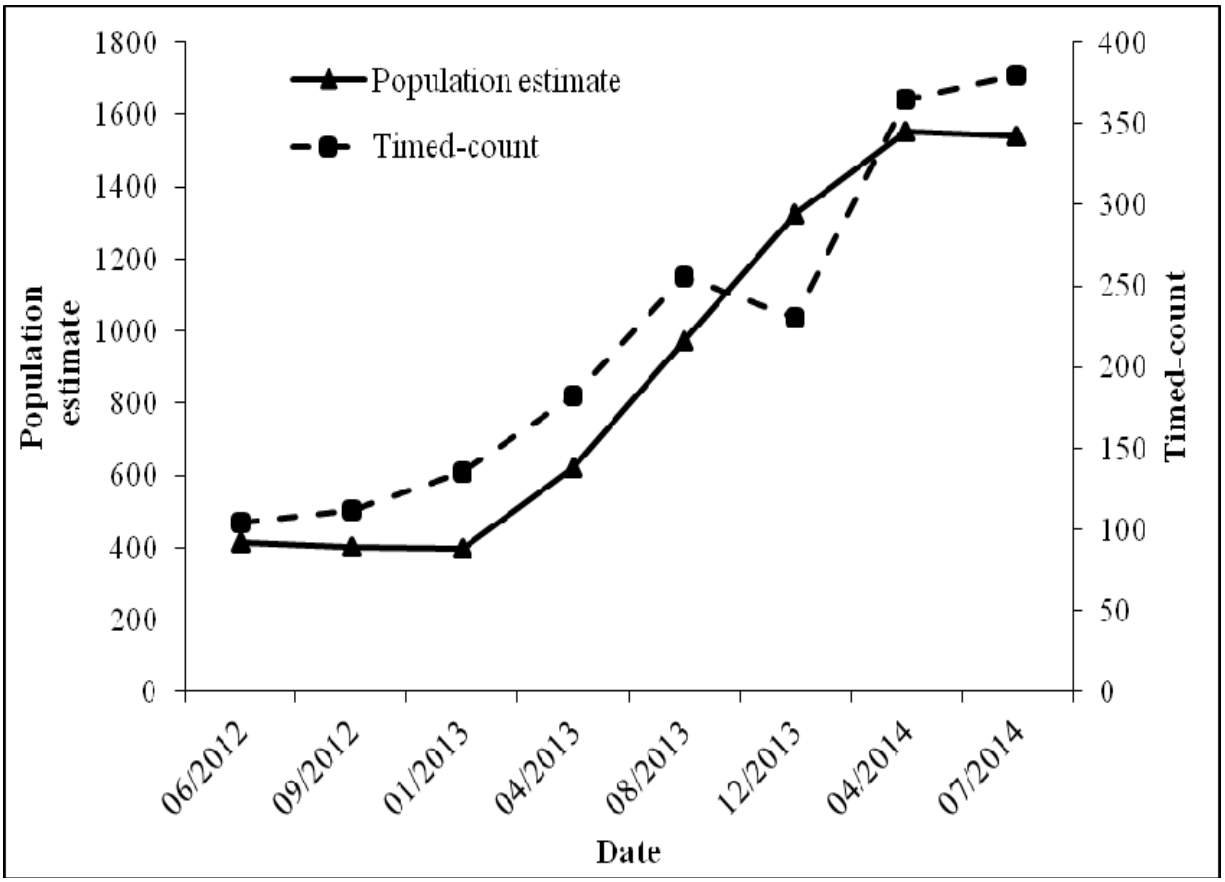

Figure 37. Timed-count monitoring of *A. mustelina* in relation to population estimate. Population estimate is derived from total number of augmented snails minus total shells recovered from ground shell plots. Timed-count data represents mean values for multiple surveys per quarter.

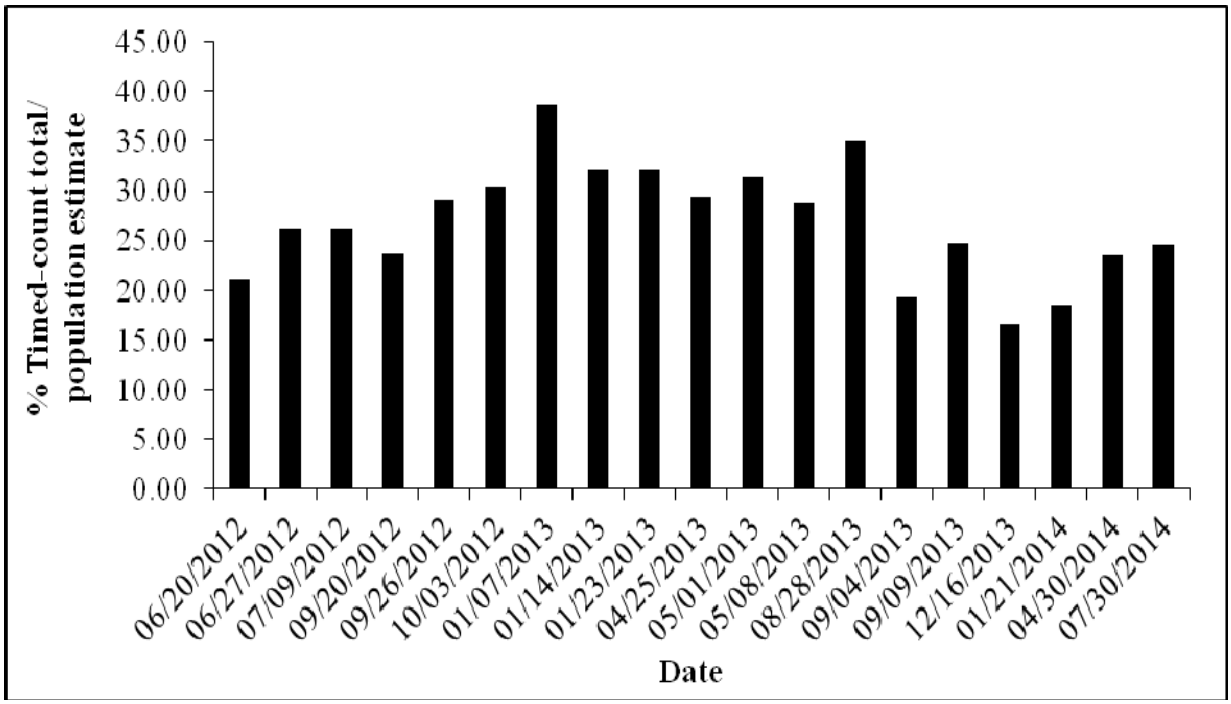

Figure 38. Detection rates for *A. mustelina* at Pu‘u Hapapa. Population estimate is derived from total number of augmented snails minus total shells recovered from ground shell plots.

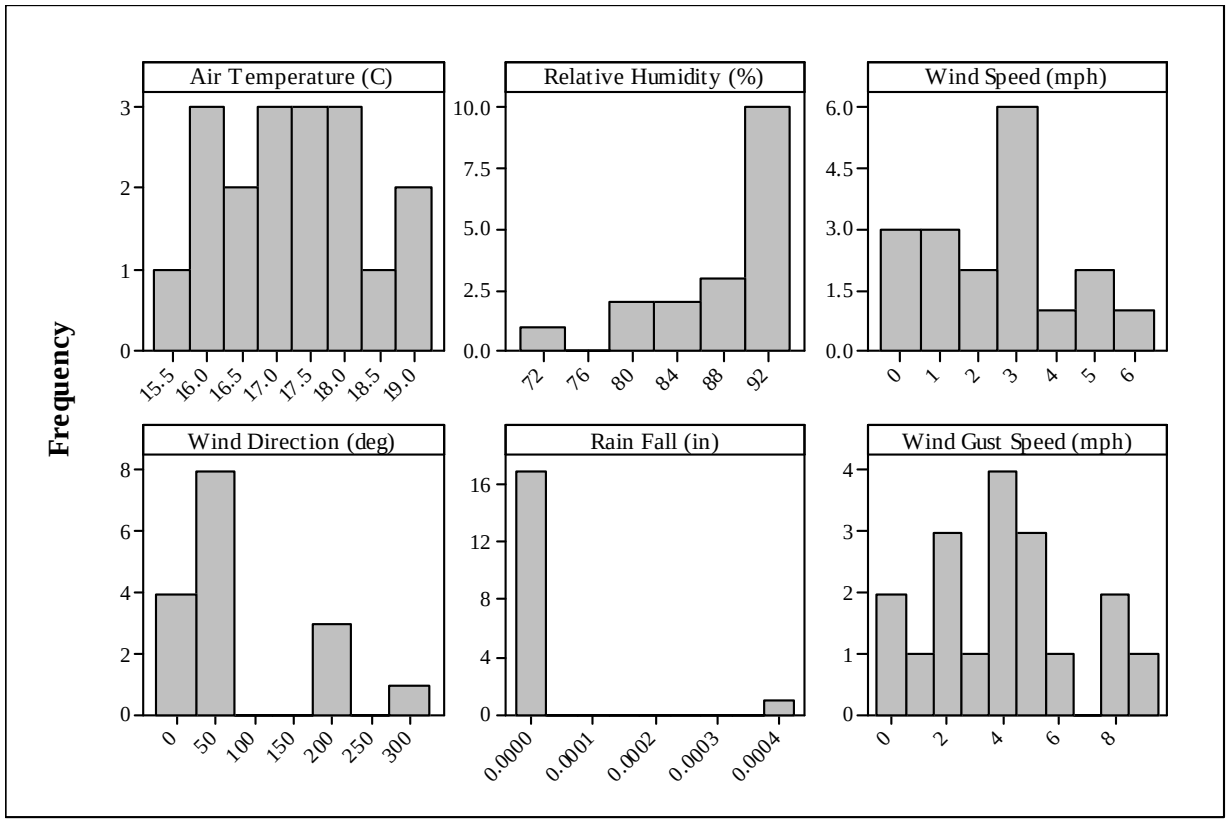

Figure 39. Distribution of environmental variables during timed-count monitoring at Pu‘u Hapapa. Data is derived from mean hourly measurements for each of the 17 monitoring events between June 2012 and July 2014.

Table 11. Correlation of environmental variables with snail detection rates at Pu‘u Hapapa. Spearman’s correlation was used for relative humidity, wind direction, and rainfall due to non-normality. Because wind speeds averaged 0 mph during two monitoring events, there were fewer samples for wind direction.

	r^2	ρ	p	df
Air Temperature (°C)	12.6	-	0.148	16
Wind Speed (mph)	11.0	-	0.178	16
Wind Gust Speed (mph)	16.2	-	0.122	16
Relative Humidity (%)	-	0.319	0.196	16
Wind Direction (deg)	-	-0.127	0.640	14
Rainfall (in)	-	0.164	0.516	16

By July 2014, the population reached an estimated total of 1542 (total number of snails added to the enclosure minus the number of snail shells recovered from ground shell plots). This far exceeds the MIP numerical goal for the ESU. Because there was continual augmentation of snails into the population, OANRP managers were unable to use the data to determine if intensive threat control was enough to reverse negative population trends and achieve the stable and self-replacing goal. OANRP managers were, however, able to detect lower mortality rates after the first four months following the initial release of large numbers of snails from the laboratory, and to assess the efficacy of utilizing timed-count monitoring to quantify long-term population trends. Following completion of augmentation efforts, the timed-count monitoring methodology may be used to assess if the population is stable and self-replacing. However, because *A. mustelina* birth rates are low, and small snails are more difficult to detect than larger ones, it may take several years before population trends become apparent.

In order to reduce the total effort spent monitoring, timed-count surveys were reduced to one per quarter instead of three per quarter. For this reason, it will be of greater importance to control for variability between surveys by maintaining a consistent monitoring crew over time, and avoiding conducting monitoring during unusually dry nights, when snails may be less active. If lower, or higher, than expected numbers occur during timed-counts, an additional timed-count survey should be conducted to minimize the influence of variation in detection.

Enclosure Habitat Restoration

Line clearing and site preparation for introduction of snails into the Pu‘u Hapapa enclosure had a drastic impact on vegetation cover. The native forest trees were largely connected by interspersed *S. terebinthifolius*, and this invasive weed tree was removed along the fenceline corridor, and wherever found inside the enclosure. The twisted trunk and thick bark of this species is favorable mollusk habitat and its removal was deemed necessary to clear the enclosure of *E. rosea.* The habitat inside the enclosure was further disturbed physically during intense surveys for *E. rosea* that began shortly after fence completion. Leaf litter and dense

understory was raked and removed from the entire enclosure regularly to enhance the detection *E. rosea,* particularly juveniles.

The urgency to fill the light gaps and maintain a more constant humidity prompted restoration actions that could be done easily while staff worked in the area. As a result, early restoration focused on transplanting native saplings from the surrounding area into the enclosure, conducting seed sows of *Pipturis albidus* and *Bidens torta,* and outplanting any local plants available in the greenhouse. Fortuitously, plantings conducted by The Nature Conservancy Hawai‘i years earlier that were inside the enclosure responded dramatically to the removal of *S. terebinthifolius.* These plants were stunted by the dense alien canopy, however, within a couple of years after removal, these stands increased dramatically in size and density, and began to reproduce prolifically.

Formal restoration plans were developed in 2012 and prioritized outplanting of hundreds of snail host species, establishing cover on areas of bare ground, and creating a connected native canopy across the enclosure. Over the course of three years, cuttings and or fruit/seed were collected from source plants from the surrounding area for propagation in the OANRP Nursery and then outplanted during the winter months (see table 12 for outplanting summary). The major effort of revegetating the enclosure is now completed.

Table 12. Pu‘u Hapapa Outplanting Summary. Plants were planted from February, 2012 through January, 2015 avoiding the hot summer months. Plants were monitored in September 2014 and the last reintroduction (not monitored) followed in early 2015.

Species	**Total Planted**	**Remaining September 2014**	**Survival**	**Planted 2015**
Acacia koa	11	unknown	unknown	
Antidesma platyphyllum	43	41	91%	12
Cyanea membranacea	4	1	25%	
Freycenetia arborea	11	1	9%	31
Labordia Kaalae	27	22	81%	3
Myrsine lessertiana	114	103	90%	
Perrottetia sandwicensis	73	57	78%	
Pisonia sandwicensis				11
Planchonella sandwicensis	7	6	83%	5
Urera glabra	70	62	89%	
Urera Kaalae	40	18	44%	
TOTAL	400	311	80%	Total: 62

Outplants were monitored at least once every 6 months to determine survival and replacement needs, and for an overall understanding of restoration applicability of the species used.

Catchment water was readily available onsite to water plants when visibly dry. Outplantings were deemed complete when staff anecdotally noted that enough plants were established to create a connected canopy in the near future. Continual assessments will continue to determine if more outplants are needed to contribute to canopy cover or to enhance the suite of snail host trees, or the diversity of the forest patch inside the enclosure.

Early seed sows of *P. albidus* proved very beneficial in establishing canopy cover in light gaps between forest patches, and creating shady areas for outplants. Similarly, continued sowing of *Bidens torta* provided dense cover for areas of bare ground and re-established leaf litter on the ground. Seed sows of other species were trialed, and some success was found with the sedge *Carex wahuensis*. Transplants made early on in the enclosure did not fare as well as greenhouse outplants ultimately did. Many divisions taken from parent plants at Pu‘u Hapapa of *Dianella sandwicensis* and *Microlepia strigosa* did establish inside the enclosure.

The barrier is also effective against other mollusks such as slugs. Three introduced species are present in the area and a serious threat to native plant regeneration. Though the angle barrier is ineffective alone, the bed of nails and the electrical barriers are effective. The prevention of slug ingress is important to enable continuing natural native habitat restoration within the enclosure.

Figure 40. *Urera glabra* outplants with *Achatinella mustelina* present on leaves. As of spring 2015, snails are regularly found on a variety of outplanted species.

CONCLUSIONS

The enclosure has been successful in excluding snail predators. One chameleon has been found within the enclosure after possibly bridging vegetation crossing the fence from the inside to the outside (since trimmed). We have not cut back the surrounding trees completely for fear of creating a large dry, barren zone around the enclosure.

Since there was continual augmentation of snails into the population, OANRP managers were unable to use the data to determine if intensive threat control was enough to reverse negative population trends and achieve the stable and self-replacing goal. Once the augmentation effort is completed it will allow management to use the timed-count monitoring (TCM) methodology to assess if the population is stable and self-replacing.

OANRP managers were, however, able to detect a decrease in the mortality rates over time as well as assess the efficacy of utilizing TCM to quantify long-term population trends. The total number of snails within the enclosure reached 993 snails by September of 2013, which far exceeds the Makua Implementation Plan goal pf 300 snails for the ESU. However, it is unclear whether or not the numerical goal will ensure a stable snail population.

In efforts to reduce the total effort spent monitoring, OANRP reduced the monitoring effort to one survey per quarter instead of three surveys per quarter. It would be important to maintain a consistent monitoring crew over time and *only* conduct monitoring during evenings when snails are the most active. If unusual observations, such as a significantly low number of snails are detected, an additional timed count survey should be conducted to assess if the observation is reflective of a true change in the population or is due to introduced variability.

Management Implications

The monitoring framework and results of this study revealed several considerations:

1) Only a fraction of snails are detected during surveys - implying that the reported number of snails within each ESU is an underestimate of the total number of snails actually present.
2) Timed-count monitoring is an effective method of quantifying long-term population trends for *A. mustelina*.
3) Detection rates are variable and may change over time; caution should be used in estimating population size based on timed-count surveys.
4) Environmental variables should continue to be considered when analyzing monitoring results due to uncertainty of effects on snail detection rates.
5) Timed-count monitoring should not be conducted too soon following augmentation, to allow time for dispersal.
6) It may take several years before population trends become apparent.

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