# Efficacy of non-electric, cut wire mesh barriers to repel *Euglandina rosea*

## INTRODUCTION

*Euglandina rosea* or the rosy wolfsnail was introduced to Hawaii in 1955 to control agricultural mollusk pests. A generalist predator, *E. rosea* proved adept at finding prey in varied habitats, including native forests, where it contributed to the decline and endangerment of endemic snail species (Hadfield & Mountain, 1980; Meyer & Cowie, 2010). *Achatinella mustelina* is an endemic, endangered species of tree snail found only in the Waianae Mountains of western Oahu. Oahu Army Natural Resources Program (OANRP) maintains five predator-proof enclosures on Oahu to protect *A. mustelina* from threats, including *E. rosea*. *Achatinella mustelina* have declined nearly everywhere except within the enclosures (Costello *et al.* 2018a, Costello *et al.* 2018b) proving they are essential to the survival of this species. An electric barrier and two physical barriers (an angle and a copper cut-wire mesh) are employed to prevent *E. rosea* incursion. All three of these barriers are described at more length in Rohrer *et al*. 2016. Unfortunately, the discovery of *E. rosea* crossing the angle barrier under their own initiative and the observation that a snail placed directly on the copper cut-wire mesh could cross, has led us to reevaluate the experiments that led us to adopt these methods. Additionally, *E. rosea* has been found several years after construction inside two of the five enclosures with all three barriers in place. In both instances it was unknown *when* the incursion occurred.

The failure rate (*i.e*. the ability of an *E. rosea* to cross within 24 hours) of three barrier types: electric, downward-facing cut wire mesh, and angle was reported to be 0, 0 and 2% respectively (Holland 2013). These values were based upon an experiment which presented each of the three barrier types to groups of 10, 10 and 30 *E. rosea* (snails) over 24 hours. It was unclear whether the same, or different, subjects were used in each trial or whether there were differences in behavior when groups of 10 were tested vs. 30. It was not stated how often snails were checked to see whether they had crossed the barrier. This was a problem because snails may move back and forth over a barrier, returning to their original positions. The snails also consume each other. A single observation may miss this barrier crossing. Barrier failure was determined by dividing the number of escapes by total snails (50) tested on any given barrier. Following barrier testing, a control group using 30 snails was presented with an easily surmountable plywood barrier. Only 40% crossed in 24 hours. The majority of the snails did not move, or were not observed frequently enough to detect movement. This suggests that the failure rates of the test barriers may have been substantially underestimated. OANRP funded this research and found it to be the best available at the time.

It has now been 6 years since the original study and barrier modifications have been suggested. Upon review, it became clear that no consistent, repeatable methodology was ever developed to compare efficacy among barrier types, nor was there a meaningful control group. When attempting to compare barrier failure rates across experiments or between different types of barriers, we found it difficult to draw any robust conclusions from the prior study. We therefore decided to not use the same methodology to test new barriers.

Here we describe our development of a repeatable method for testing the efficacy of snail barriers relative to a meaningful control (in which close to 100% of snails eventually crossed). We carried out this work to better evaluate the ability of current barriers to exclude snails as well as to test new prototypes. In addition to testing barrier efficacy, we were interested in whether snail size or source location affected behavior. In 2017 Meyer *et al.* discovered the species grouped under *E. rosea* was, in fact, two different clades. Being unable to distinguish between the two, we used the snail source location as a proxy for this

potentially important variable. We have not yet concluded testing of all barrier types, but present results for three types of cut-wire mesh barriers below.

## MATERIALS AND METHODS

### Experimental subjects

We used wild caught snails kept in captivity for a minimum of three days prior to testing, during which time they were fed raw oysters (Dietrich 2013). Test trials lasted four days during which time no food was provided. Snails are known to live up to four months without food, however, after one month, activity decreases and physiological changes become evident including slowed growth and reduced egg laying (Gerlach 1994). Satiated snails consume one prey item every three days (Dietrich 2013) while underfed snails survive well on one prey item every 14 days (Gerlach 1994). The snails in our study, therefore, were not starving during the testing period, but could be expected to begin searching for prey before the end of the trial period. A total of 132 snails were used, 88% of which were primarily from six Management Units (Figs. 1 & 2). Our observation that very small snails seldom moved and often died before testing led us to reject snails whose shell axis was < 20 mm. Experimental snails included juveniles, subadults, and adults but no hatchlings (Gerlach 1994) (Fig. 3). Calipers were employed for measurements.

Snails were numbered using a Decocolor paint pen and kept separately in two ounce plastic portion cups to prevent cannibalism. Wooden boxes with an interior ledge and screened top (to prevent snail escape) were constructed according to the dimensions shown in Figure 4. The barrier (treatment) was attached to the underside of this ledge while the control box had no attachments under the ledge. We placed moist towels on top of the ledge to encourage snails to leave the interior of the box thereby crossing the barrier (if present) (Fig. 4).

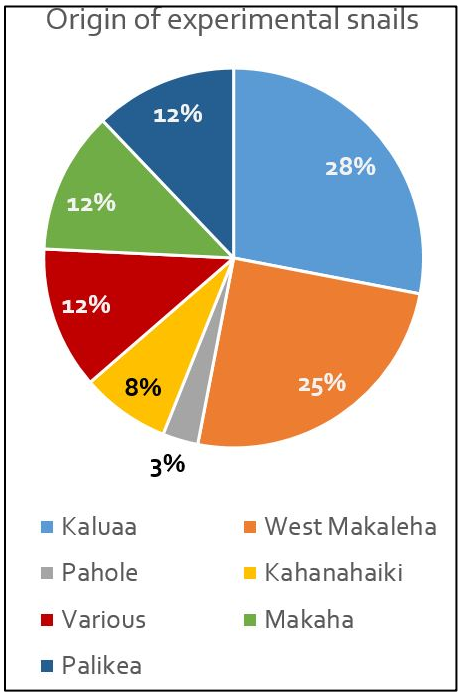


**Figure 1**. Origin of snails used in this study. Sites on Oahu where 2 or fewer snails were collected are grouped under 'Various.'

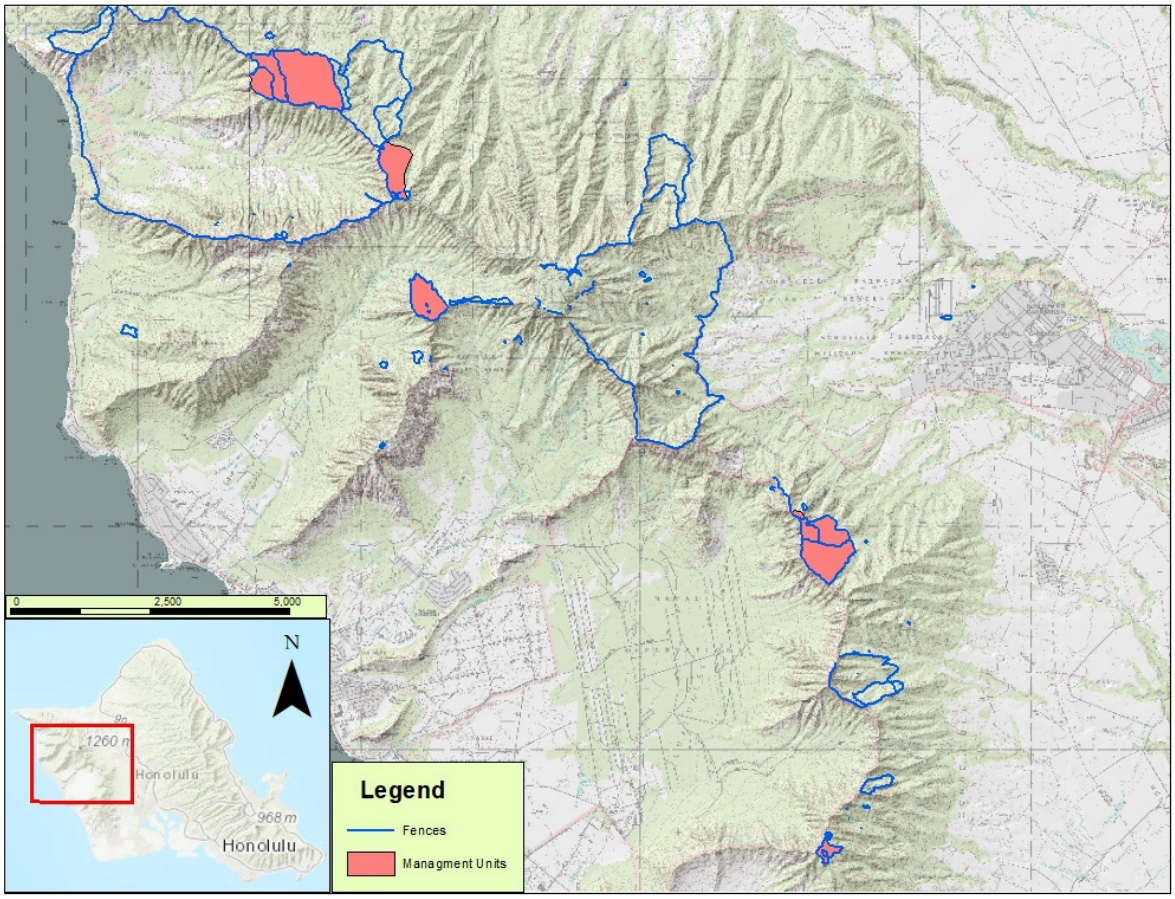


**Figure 2.** Map of snail collection locations.

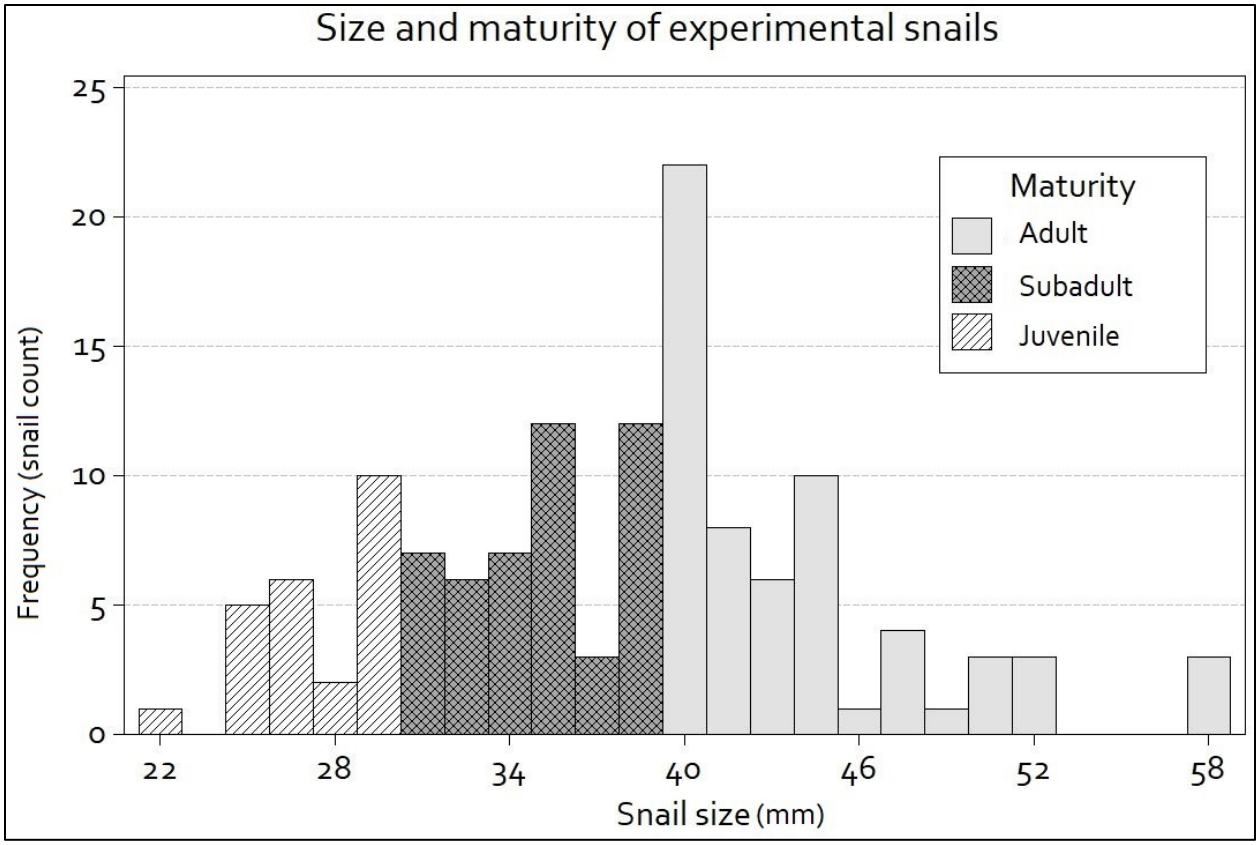


**Figure 3.** Frequency of snails in each size class (Gerlach 1994) used in the experiment. This likely doesn't reflect frequencies in the wild because large snails are easier to find and collect.

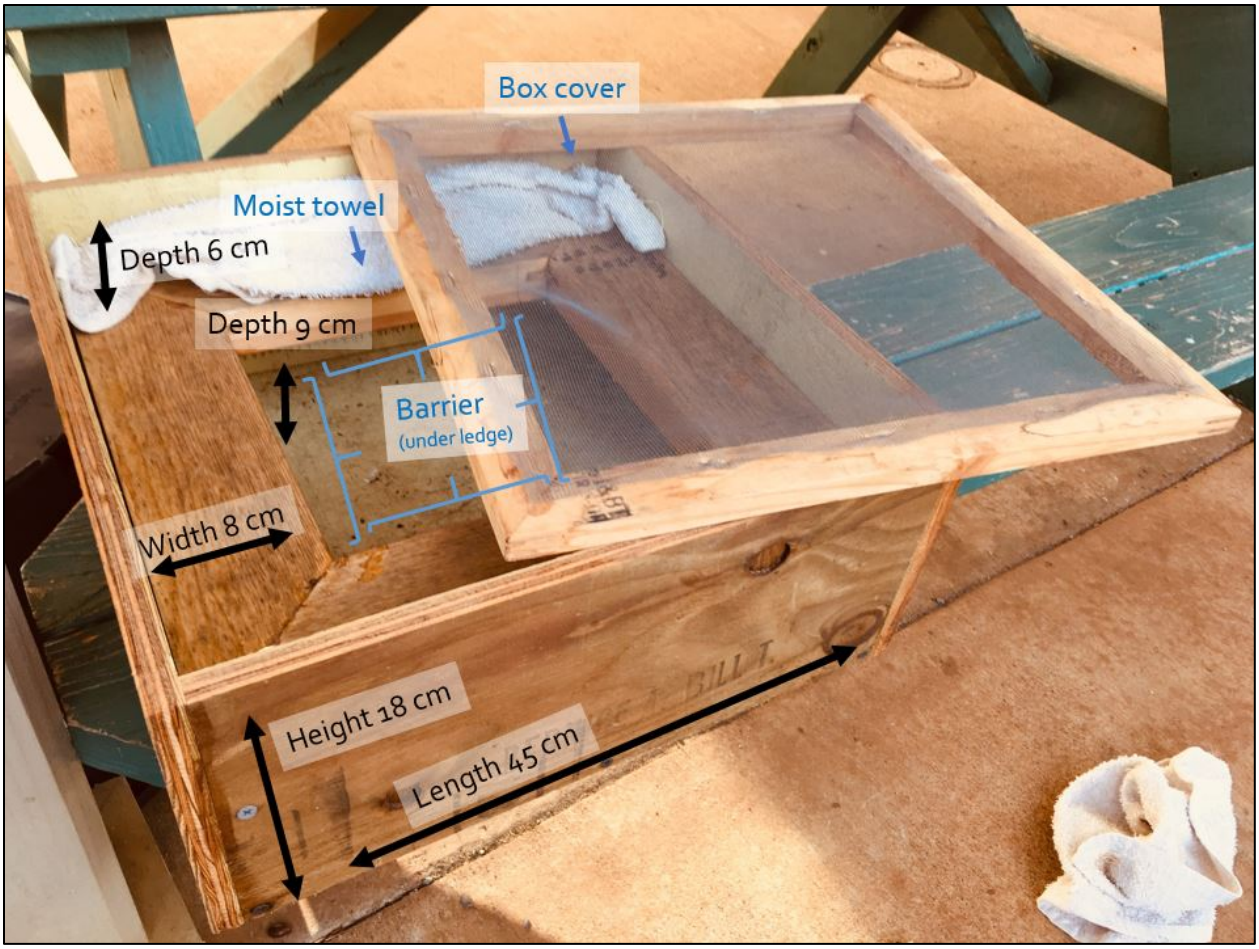


**Figure 4**. Diagram showing dimensions of the experimental box.

Barrier treatments

All barriers consisted of 11 rows of the test mesh 15 mm high secured such that each formed rows spaced 5 mm apart using construction adhesive (Figs. 5 & 6). The three barrier treatments were:

1. Copper mesh with a wire diameter of 0.7 mm and a mesh density of 6 squares per $cm^2$ (referred to as the CM6 treatment)

2. Copper mesh with a wire diameter of 0.7 mm and a mesh density of 3 squares per $cm^2$ (referred to as the CM3 treatment)

3. Stainless steel mesh with a wire diameter of 0.18 mm and a mesh density of 22 squares per $cm^2$ (referred to as the SSM treatment)

Our justification for selecting these treatments varied. The CM6 treatment is currently employed at predator proof enclosures (Rohrer et al. 2016). We hypothesized that the CM3 treatment would provide less attachment area for an inverted snail. The SSM treatment, if successful, is approximately 25% cheaper than copper and may prove more durable in harsh environments (Diamond Spas 2015).

**Figure 5**. Alignment of mesh on the underside of the ledge. The CM3 treatment is pictured.

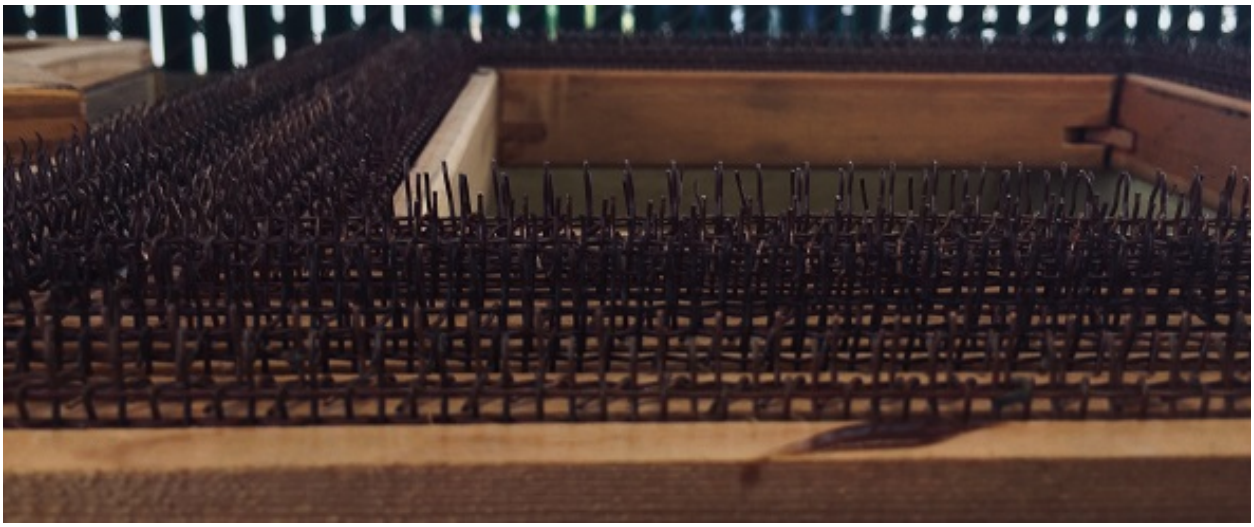

**Figure 6.** A side view of the same treatment (CM3).

Experimental design

Trials took place between February and June 2019. In each trial, we placed five snails at a time within a box. Boxes were checked two times a day for four days at 0700 and 1700 hours. At each time we recorded the identification number of each snail and their position in the box. Snails found atop the ledge (often underneath the towels) were considered ‘escaped’ as they had crossed the barrier. Those remaining in the box were considered ‘trapped.’ Snails were left in as they were found unless they died or were consumed by other snails (in other words, an escaped snail was not moved back inside the box). Data for

snails that died prior to the conclusion of the 4-day trial were omitted regardless of their position inside the box. Towels were kept moist and the interior of the box was spritzed with water once per day to provide trapped snails with moisture. At the conclusion of the trial, most snails were killed. The remainder were put into clean containers and fed. This minority was reused in up to three trials. Though we attempted to use fresh snails in each trial, we had difficulty finding sufficient numbers. Because an individual able to cross a barrier may be more likely to surmount that same barrier than a naïve snail, reused subjects were never exposed to the same treatment twice.

Though trials of different treatments were run concurrently when there were sufficient subjects, only a single control was run at each time regardless of the number of treatments. A minimum of 10 snails (5 for each group) were used to conduct a trial containing one treatment and one control. When possible, we tested multiple treatments concurrently, up to a maximum of three treatments and one control (20 snails).

If a snail escaped (*i.e.* crossed the barrier onto the ledge), data was recorded on the number of hours it took to escape the first time. Though that snail may cross the barrier again over the ensuing days, any escape during the four days was recorded as a single escape.

RESULTS

Here, barrier failure rate is percent of snails escaped. Time constraints and lack of snails led to unequal sample sizes with the control group having the most replicates followed by CM6, CM3 and lastly SSM. As more snails are found, we plan to continue testing CM3 and SSM. These are shown in Table 1. Also shown are total numbers of snails escaped and the failure rate based on those values. Among those snails that escaped, the mean number of hours to first escape is shown alongside the standard error. It is clear from these that the 24 hour length used in Holland 2013 was of insufficient duration.

**Table 1**. Barrier treatment summary.

| Barrier treatment | Sample size | Snails escaped | Snails trapped | Barrier failure rate | Mean hours to first escape (± SE) |
|---|---|---|---|---|---|
| Control | 75 | 71 | 4 | 95% | 26 $\pm$ 2 |
| CM6 | 75 | 12 | 63 | 16% | 30 $\pm$ 5 |
| CM3 | 45 | 3 | 42 | 7% | 35 $\pm$ 19 |
| SSM | 10 | 7 | 3 | 70% | 21 $\pm$ 9 |

Results from a logistic regression demonstrated that the probability of snail escape was significantly affected by treatment (P = <.0001, Fig. 7), but not by snail size (P = 0.6256, Fig. 8) or origin (P = 0.4475, Fig. 9). Among the treatment groups, the CM3 and CM6 barriers had significantly lower failure rates (Tukey's HSD, P = 0.0011) than the SSM barrier and the control, but did not differ significantly from one another; the SSM barrier was not significantly different from the control (Fig. 7).

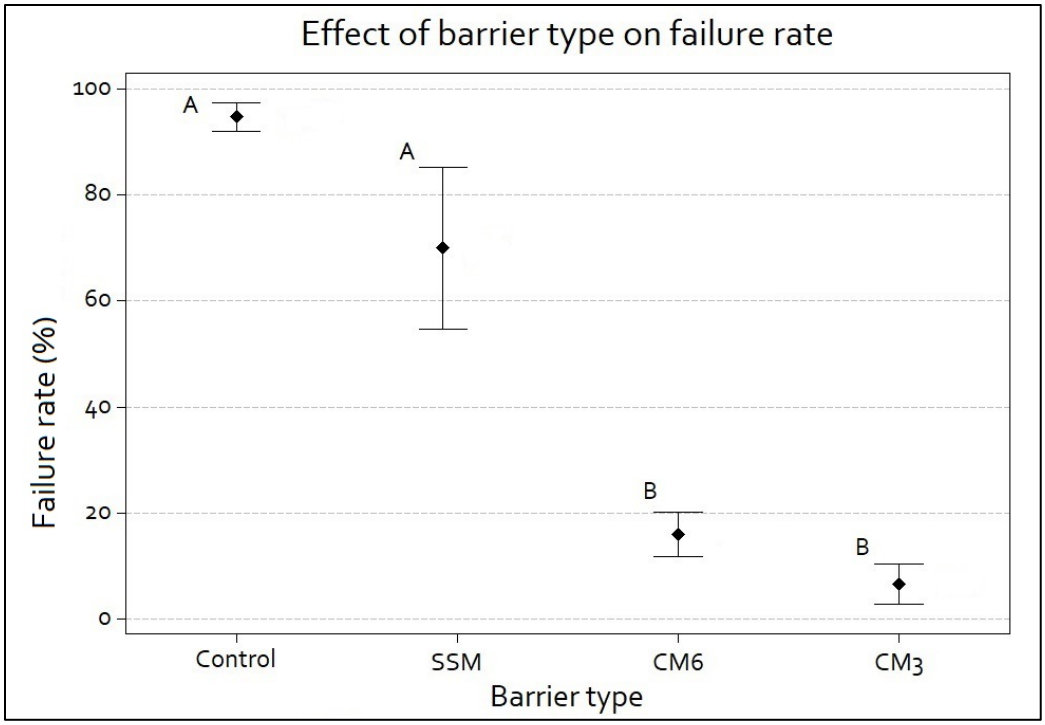


**Figure 7**. The effect of barrier type on the probability of failure. Bars are ± one standard error of the mean (SEM). Letters indicate groups that differ significantly from one another. SSM = stainless steel mesh; CM3 = copper mesh with 3 squares per $cm^2$; CM6 = copper mesh with 3 squares per $cm^2$

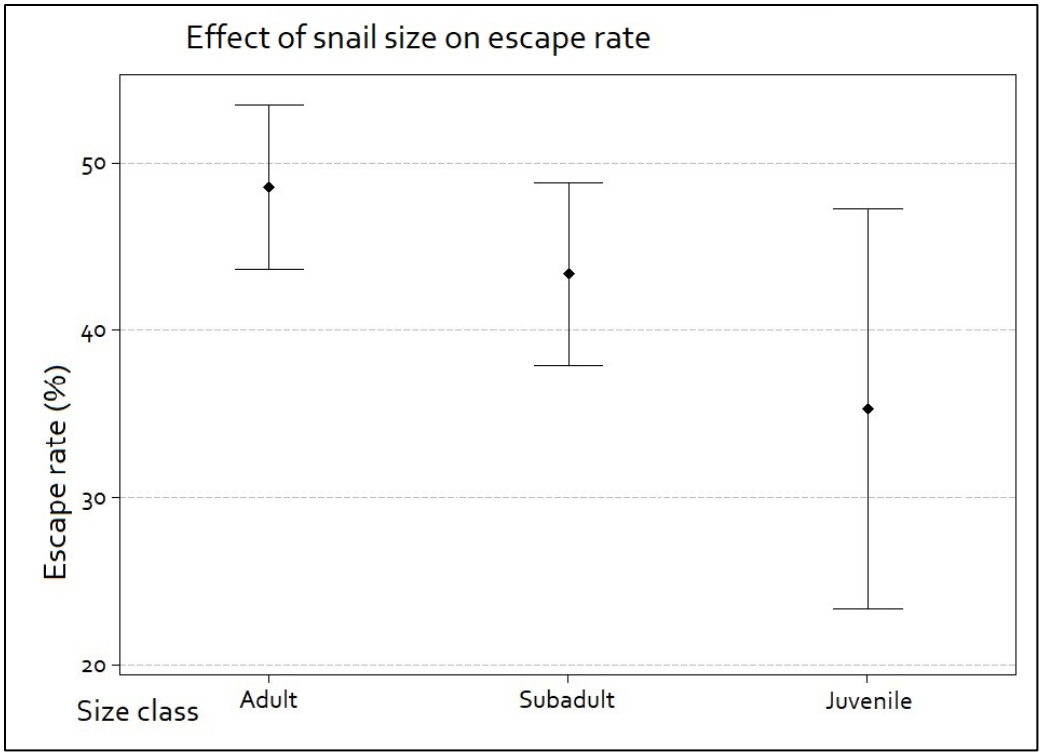


**Figure 8**. Effect of snail size on escape probability. Bars are ± 1 SEM. Size did not significantly influence escape probability. For ease of viewing, the graph shows size as a categorical variable when, in fact, a continuous measure of snail size was used in analysis.

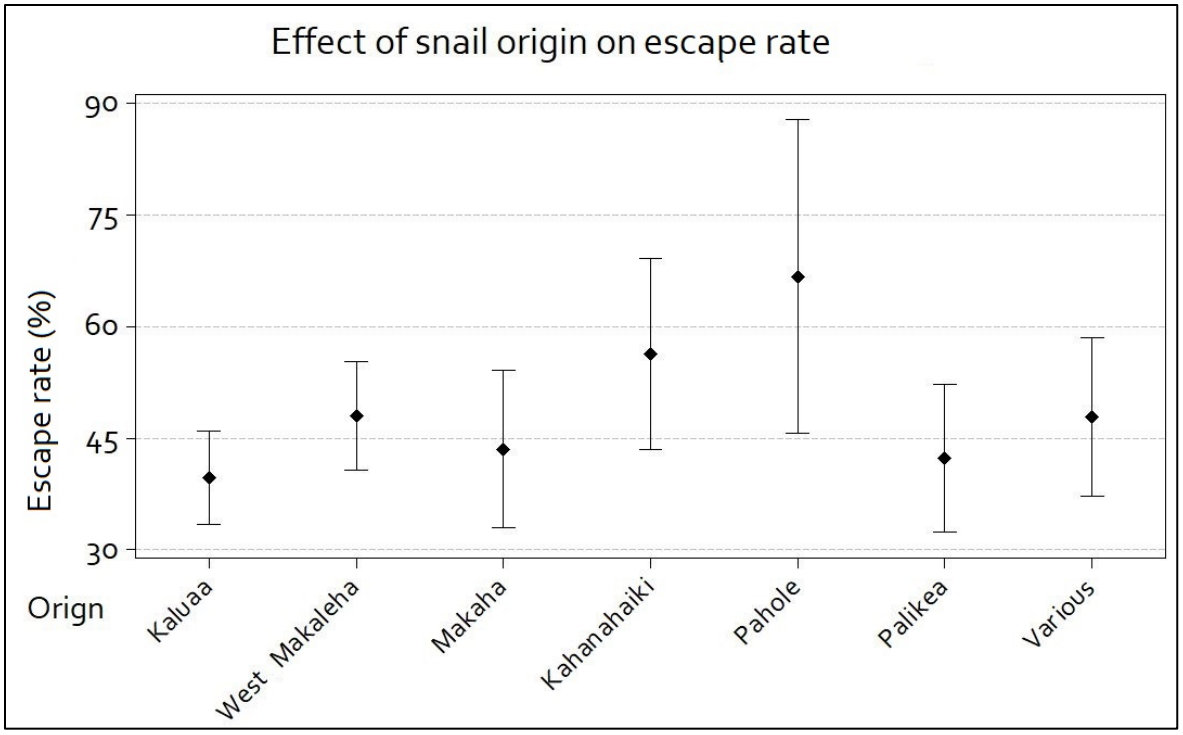


**Figure 9**. Effect of snail origin (site collected) on escape rate. Bars are ± 1 SEM. The site where the snail was collected did not significantly affect whether a snail was able to escape.

DISCUSSION

The CM6 barrier is currently used at all predator-proof enclosures. Though results show that it is not 100% effective, relatively few snails crossed this barrier. The CM3 barrier had less surface area than the CM6 barrier, and we hypothesized that this may make it more difficult for snails to adhere to the barrier. Though not significantly different, the failure rate of CM3 was less than half that of the CM6, suggesting that this hypothesis might be true. There have been comparatively few trials of the CM3 to date, and more testing will resolve whether CM3 is in fact a more effective barrier than CM6. The SSM barrier was ineffective at preventing snail incursion. If our speculation regarding surface area and barrier performance proves true, this may be because SSM had the most surface area of the three barriers tested. It would be interesting to test stainless steel at similar wire thickness and density as the copper mesh in order to see whether differences due to material exist. If not, it may be more cost effective to use stainless steel or some other cheaper alternative to copper.

Widespread implementation of this barrier by OANRP and partner agencies was informed by findings in Holland (2013). Like Holland (2013), we observed close to a 0% failure rate of CM6 over the first 24 hours. However, this failure rate increased to 16% over the course of four days. Similarly, failure rates were 2% and 42% and for the CM3 and SSM treatments (and only 44% for the control) over the first 24 hours, but subsequently increased over four days, in some cases dramatically. We do not know why snails were slow to move initially. It was not due to moisture, as they were thoroughly saturated twice a day. Nor was it due to starvation since they were well fed prior to testing. Healthy snails in moist conditions move, on average, 8 mm/second over 12 hours (night time only) (Gerlach 1994). By this measure, a snail could cross the length of our box diagonally (636 mm) in less than two minutes. Regardless of the explanation, our results indicate that barrier trial periods should be longer than 24 hours. The ideal trial period for simulating snail pressure in the field is unknown and perhaps unknowable. In nature, snails can

opt to move away from the barrier in order to find food and shelter. This could mean that any amount of time that snails are placed in a small box might overestimate actual rates at enclosures. On the other hand, snails at enclosures have unlimited time to attempt entry and are influenced by a myriad of factors such as the presence of slime trials (Cook 1985; Gerlach 2001; Shaheen *et al*. 2005; Davis-Berg 2012) not tested here. Given these uncertainties, it seems prudent to use trial durations long enough to allow a high percentage of control snails to escape, to ensure that snails are sufficiently active and thus make inter-barrier comparisons of efficacy meaningful.

In this respect, our experiment provides a robust relative measure of barrier effectiveness. The methods reported can be used to compare different barriers objectively using standardized measures, providing reliable data to inform improvements to barrier design. Though failure rates at the enclosures remain unknown, we believe that a barrier outperforming another under our protocol should reflect real differences between the two in the field.

## CONCLUSION

To avoid confusion, here we refer to the native tree snail as *Achatinella mustelina* and the rosy wolfsnail as *Euglandina rosea*.

Though not tested using our methods, Holland's finding that the electric barrier was 100% effective seems likely to be accurate. The voltage used prevents *E. rosea* from attaching to the wires. This is fortunate as the physical barriers may not be as effective as previously thought. The downside of the electric barrier is that it is sensitive to temperature and moisture fluctuations and can break. We are just beginning to keep records on the duration and frequency of these events, so do not know how often enclosures are without an electric barrier. Some breaks result in only a partial loss of electronics with two of the four wires remaining active. We don't know if this is an effective barrier. There are also constant improvements being made to the gauge and tension of the wires to prevent future breaks. We hope that by closely monitoring those enclosures with remote systems, we will have a clearer idea of how often and for how long physical barriers are the only prevention to *E. rosea* ingress.

*Euglandina rosea* are responsible for 1/3 of all native snail extinctions on islands (Régnier 2009). As of 2018, only 3,608 *A. mustelina* were found in the wild, 33% of which are protected within enclosures (Costello *et al.* 2018b). Populations outside of enclosures are becoming scarce due to predation and climate change. Rescue by conservation agencies, including our own, involves moving snails either to enclosures or into captive propagation. It is likely that within the next few years no wild populations will exist outside enclosures. A single *E. rosea* can consume 350 prey items in 16 months (Chiu & Chou 1962). This means that barrier failure and breach of enclosures by *E. rosea*, if not rapidly detected, could result in substantial *A. mustelina* mortality.

Given the importance of these enclosures, it is perhaps unsurprising that considerable labor and resources are dedicated to their construction and maintenance. Rohrer (*et al.* 2016) reports that over 1,000 person hours were required to clear a 160 m perimeter for the enclosure at Puu Hapapa. This did not include labor associated with its construction which amounted to $100,000 in 2009.

Further testing of enclosure barriers is needed, especially the angle, as it is evident that existing research is insufficient to demonstrate its level of efficacy relative to the mesh barrier. Given the importance of enclosures to the survival of *A. mustelina*, the enormous cost they require to build and maintain, as well as our plans to build more, it is wise to make certain they really *are* predator proof to the extent possible, and to understand any barrier efficacy limitations.

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