**The effects of squid-baiting pitfall traps for sampling wētā (Orthoptera) and other ground-dwelling forest invertebrates.**

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Short title: Efficacy of squid-baited pitfall traps

**Abstract**

Pitfall traps are commonly used to sample surface-active invertebrates, although the efficiency of the technique varies among taxa. We investigated how baiting pitfall traps with squid influenced sampling of some ground-dwelling invertebrates in New Zealand forests. The study was conducted across a total of 21 sets of seven lethal pitfall traps established between November 2012 and November 2015 in Aorangi and Remutaka forests. Four non-baited and three squid-baited lethal pitfall traps were established per set and remained active for one night during November/December and three nights during February. Squid-baited pitfall traps caught four times as many ground wētā and three times more cave wētā per unit effort than unbaited traps. Most of the ground wētā were identified as *Hemiandrus pallitarsis* (Walker, 1869). Carabidae, Scarabaeidae, Staphylinidae, Opiliones, Hymenoptera and Araneae were also more abundant in squid-baited than in unbaited traps. There was little difference in the catches of peripheral pitfall traps versus central pitfall traps, suggesting that 5 m spacing is sufficient to generate independent samples. Exceptions were Araneae and Amphipoda which were both approximately 1.5 times more abundant in central compared to peripheral unbaited traps. The attraction of ground and cave wētā to squid provides some insight into their dietary range. The higher catches obtained with squid-baiting, suggests this may be a useful modification to increase sampling rates, which is valuable where sampling effort is logistically constrained such as on islands or other remote study sites.

**Key words**

carrion, insects, lure, passive sampling, surface-active invertebrates.

**Introduction**

Invertebrates are important components of biodiversity and are often used to monitor the ecological effects of land-use practices and conservation management (Keesing & Wratten 1998; McGeoch 1998; Johns 2001). They are well represented in forest habitats and are indispensable for any ecosystem (Bowie & Frampton 2004) due to their contribution to pollination, seed dispersal, and nutrient cycling (Wilson 1987; Keesing & Wratten 1998), as well as forming part of the diet of many vertebrates (Wilson 1987; Williams 1993).

One widely used method to survey ground-dwelling invertebrates is lethal pitfall trapping (Sutherland 2006; Sherley & Stringer 2016). In terms of taxonomic diversity and the numbers of individuals caught, the effectiveness of this method can be altered by modifications such as addition of attractants and/or by modifying the sampling area using fences, polythene barriers or cages for exclusion of vertebrate and invertebrate predators (e.g. Walker 1957; Brennan et al. 2005; Woodcock 2005; Carrillo et al. 2007). Any of these, including use of baits or lures directed at attracting particular types of invertebrate, has its own bias since they may usefully increase the relative proportion of some taxa over others (Greenslade & Greenslade 1971; Woodcock 2005; Brown & Matthews 2016). Seldon & Beggs (2010) reported that Diptera and carrion Coleoptera (i.e. Agyrtidae, Hydrophilidae, Leiodidae, Scarabaeidae and Staphylinidae) were preferentially caught when squid was used as bait in lethal pitfall traps in the Waitakere Ranges, upper North Island of New Zealand. Another study, from the Czech Republic, demonstrated that fish was a useful bait when targeting Silphidae (Coleoptera) (Knapp et al. 2016). However, both studies concluded that neither squid nor fish were effective at increasing catches of Carabidae (Coleoptera), which is commonly used as an indicator of habitat changes worldwide (Vandewalle et al. 2010). Neither of these studies were attempting to capture Orthoptera.

We compare the effect of squid-baited versus unbaited lethal pitfall traps as a means of sampling wētā and other invertebrates in New Zealand forests. In particular, we examined for a bias in the effect of squid baits in attracting different body sizes of male, female or juvenile ground wētā. In addition, we tested for interference between traps due to either a positive response toward the higher odour concentration expected in the centre of the trapping array, or conversely a “shielding” effect evidenced by higher catches in the peripheral traps (Perner & Schueler 2004; Hasenbank & Hartley 2015)

**Methods**

**Study area**

The study was carried out in Aorangi Forest Park and Remutaka Ranges (lower North Island, New Zealand) located at 41°25′ S, 175°21′ E and 41°5' S, 175°14' E, respectively. Sampling took place on seven occasions in late-spring (November-December) and late-summer (February) from November 2012 to December 2015. The Aorangi Forest Park lies between Martinborough in the north and Cape Palliser in the south, and covers a large part of the Aorangi Ranges. The Remutaka (formerly Rimutaka) Ranges includes the Catchpool and the Orongorongo Valleys, and joins the southern end of the Tararua Ranges (Department of Conservation 2014). Both forests are protected mountainous areas with indigenous flora, dominated by broadleaf and podocarp species at lower elevations (150-500m) (e.g., *Weinmannia racemosa* (kamahi), *Hedycarya arborea* (pigeonwood), Cyatheales (tree ferns), *Pseudopanax crassifolius* (lancewood), *Melicytus ramiflorus* (whiteywood), *Dacrydium cupressinum* (rimu), *Pectinopitys ferruginea* (miro), *Podocarpus totara* (totara)). They are mixed with small trees, shrubs, ferns, mosses, lichens, lianas and epiphytes. Nothofagaceae species (beech forest) (e.g., *Fuscospora* *fusca* (red beech) and *Lophozonia menziesii* (silver beech)) are dominant at higher elevations, in dry sites and/or infertile soils. The mean annual rainfall varies from 800 mm in eastern Wairarapa to over 2400 mm in the centre of the Remutaka Range (Brockie 1992; Dymond & Shepherd 2004).

**Invertebrate sampling**

Surface-active invertebrates were caught using lethal pitfall traps (110 mm mouth diameter, 100 mm deep) containing c.25 mm depth of 35% w/v sodium chloride solution and a few drops of detergent. One set (or “line”) consisted of seven pitfall traps placed 5 m apart along the middle 35 m section of rodent and possum monitoring lines; with a maximum of 15 lines located in the Aorangi Forest Park and six in the Remutaka Ranges (each mammal monitoring line was 450 m long, after Gillies & Williams 2013) (Table 1). Invertebrate sample lines were between 150 and 800 m of elevation. Three pitfall traps per line were baited with decomposing squid (frozen fresh, then thawed for 1–2.5 days), alternated between four unbaited traps. The squid bait (~10×20×3 mm) was hung from a thin wire suspended beneath a 18×18 cm square plastic lid raised 25–30 mm over the centre of the pitfall trap (Seldon & Beggs 2010). The 5 m spacing between traps is commonly used in pitfall studies (Ward et al. 2001) and is generally considered sufficient to limit inter-trap interference. To assess this assumption, traps were categorised as Peripheral, P, (the two traps at either end of each sequence) or Central, C, (the central three traps in each sequence) as there might be preferential attraction toward the central traps (where olfactory cues may reinforce one another) versus a “shielding” effect whereby peripheral traps (P) may intercept a higher than average catch (Stenseth & Hansson 1979; Hasenbank & Hartley 2015). Traps were named as follows: four peripherals (P) comprised two baited and two unbaited traps per line, and three central traps (C) including one baited and two unbaited traps per line. Pitfall traps were active in the field for one night in late spring (mid-November to mid-December) and three nights in late summer (February) coinciding with a mammal monitoring protocol carried out in Aorangi and Remutaka ranges.

Captured invertebrates were sorted by eye to ordinal and family level (Johns 2001; Gwynne 2005; Larochelle & Larivière 2007) except for Anostostomatidae, ground wētā, which were identified to species. The abundantly sampled single species of ground wētā (*Hemiandrus pallitarsis*) were further classified as adult female, adult male or juvenile based on the presence of the secondary copulatory organ for females (Gwynne 2002; 2005) and the body length (>18mm being the threshold for adults).

**Statistical analyses**

To study the effect of squid baiting (present vs absent), pitfall position (peripheral vs central positions) and season (November vs February) on the number of wētā or other invertebrate taxa in a trap, we used Generalized Linear Mixed Models (GLMMs) with a Poisson family error distribution and log link (Eqn 1). Pitfall position and its interaction with squid baiting were included to test for effects of interference among traps such as shielding and attraction-spillover on the adjacent unbaited traps along lines. We also tested for an effect of squid baiting, pitfall position and season upon average body sizes of wētā and the ratio of adult to juvenile ground wētā, using equivalent Linear Mixed Effect (lmer) models with Gaussian error distribution. In both sets of models, monitoring lines (max n= 21 in total) nested within broader sites (n=7) were included as random effects (Table 1). We included an offset into the glmer model for the number of trap-nights (TN). The offset allows us to model “counts per trap-night” while retaining the strength of the Poisson analysis with integer counts data as the input (Parry 2018).

(Eqn 1)

When an effect of pitfall position was found for a particular taxon (position: *p*<0.05 or position \* squid baiting: p<0.05), we fitted the following model (once for squid-baited traps and once for unbaited traps) to confirm whether the interference was affecting baited and/or unbaited pitfall traps (Eqn 2):

(Eqn 2)

We analysed counts of the most abundant ground-dwelling invertebrates collected in Aorangi and Remutaka forests including Orthoptera (ground and cave wētā), Coleoptera (Carabidae, Scarabaeidae and Staphylinidae), Hymenoptera, Araneae, Opiliones, Diplopoda, Amphipoda and Gastropoda. Wētā were collected, counted and measured from November 2012 to December 2015 (seven sampling events and 1435 trap-nights in total) while other invertebrates from November 2013 to December 2015 (five events and 1190 trap-nights in total). Statistical analyses were performed using R base functions (Version 0.99.902, © 2009-2016 RStudio) and the lme4 package (Bates et al. 2015).

**Results**

**Wētā**

A large number of ground wētā (n = 528) and fewer cave wētā (n = 195) were collected in the Aorangi and Remutaka forests during 1435 trap-nights between November 2012 to November 2015. Most of the ground wētā were positively identified as *Hemiandrus pallitarsis*, with only one confirmed individual of *H. luna* (Taylor-Smith et al. 2016). The sex ratio of the ground wētā in the samples, considered across all seasons, was male-biased, 1.8:1 in Aorangi and 1.4:1 in Remutaka.

Four times as many ground wētā (*Hemiandrus* spp.) were caught in squid-baited pitfall traps compared to unbaited pitfall traps (mean of 1.39±0.32 individuals per trap-night, versus 0.35±0.11; glmer: χ²(1)=93.44, *p*<0.001) (Table 2, Figure 1). Significant differences persisted when samples were analysed according to sex and life stage: more than five times more females (glmer: χ²(1)=23.63, *p*<0.001), almost five times as many as males (glmer: χ²(1)=42.10, *p*<0.001) and more than twice as many juveniles (glmer: χ²(1)=29.72, *p*<0.001) in squid-baited pitfall traps compared with unbaited traps (Table 2). Cave wētā were three times as abundant in squid baited pitfall traps (mean=0.47±0.10 individuals per trap night) compared with unbaited traps (mean=0.22±0.07 individuals per trap night) (glmer: χ²(1)=12.97, *p*<0.001) (Figure 1).

The ratio of adult to juvenile ground wētā (*Hemiandrus* spp.) found in baited pitfall traps (1.5:1 = adult based) was significantly different from the ratio caught in unbaited traps (0.7:1 = juvenile based) (lmer: χ²(1)=24.94, *p<*0.001) (Figure 2). Ground wētā were more abundant in late summer (February) than late spring, and the ratio of adult to juvenile ground wētā changed with season, being 1.5:1 in February, compared to 0.9:1 in November (lmer: χ²(1)=37.19, *p<*0.001). Ground wētā did not show any significant difference in the ratio of adult to juvenile for the interaction between squid baiting and season (Figure 2). When separated by age-class and sex the average body size of females, males and juveniles did not differ due to squid-baiting or season.

**Other invertebrates**

Significant positive effects of squid-baiting were also found for the catches of most other surface-active invertebrates (Table 2, Figure 1). Scarabaeidae (Coleoptera) were more than twenty-two times more abundant (glmer: χ²(1)=162.75, *p*<0.001) while Staphylinidae (Coleoptera) were four times more abundant (glmer: χ²(1)=26.04, *p*<0.001) in squid-baited than in unbaited pitfall traps. Carabidae (Coleoptera) were 1.4 times more abundant in baited compared with unbaited pitfall traps (glmer: χ²(1)=6.47, *p*=0.011). Hymenoptera (mostly ants) showed almost three-fold increase (lmer: χ²(1)=16.41, *p*<0.001), Opiliones more than two-fold (lme; χ²(1)=66.97, *p*<0.001), and Araneae showed almost two-fold increase (glmer: χ²(1)=44.59, *p*<0.001) in squid-baited pitfall traps compared with unbaited traps. Diplopoda, Gastropoda and Amphipoda did not show any significant difference in abundance due to squid-baiting, while Amphipoda showed evidence of an interaction between trap position and squid-baiting (Table 2, Figure 1).

Carabidae, Scarabaeidae, Opiliones, Diplopoda, Amphipoda and Gastropoda were more abundant per trap night in samples from late spring (November) than late summer (February) (Table 2). Only two out of eleven taxa showed effects of positional interference among pitfall traps on their catch rates. Araneae (Z=2.57; *p*<0.05) and Amphipoda (Z=3.54; *p*<0.001) were 1.4 and 1.5 times, respectively, more abundant in unbaited traps located in the centre of sampling lines (mean= 0.69±0.1 and mean= 0.58±0.19, respectively) compared to the ones placed in the periphery (mean= 0.49±0.07 and mean= 0.38±0.08, respectively). No positional effects were found for invertebrates collected in baited pitfall traps.

**Discussion**

*Hemiandrus pallitarsis* which is widely distributed across the North Island (Johns 2001; Chappell et al. 2012; Trewick & Bland 2012) was far the most common anostostomatid wētā collected in pitfall traps from Aorangi and Remutaka forests. Previous studies have reported the presence of at least two species of cave wētā in Aorangi Forest: *Pachyrhamma edwardsii* (Scudder, 1869) and *P. longipes* (Colenso, 1887) (Cook et al. 2010), and three species in the Orongorongo catchment of the Remutaka Ranges: *Pallidoplectron* sp., *Pleioplectron hudsoni* Hutton, 1897 and *Pachyrhamma longipes* (Moeed & Meads 1983). However, we could not confirm the presence of these or other species as it was not possible to reliably identify our specimens due to taxonomic uncertainty (Fitness et al. 2015; Fitness et al. 2018). In Aorangi and Remutaka forests the sex ratio of ground wētā caught in pitfall traps was male-biased suggesting either sex-baised populations, greater dispersal activity by males compared to females and/or females were hidden in burrows with eggs/nymphs, emerging infrequently (see Gwynne 2004 for description of maternal care).

The efficiency of pitfall traps is known to be sensitive to population density, environmental factors affecting individual behaviour, and surrounding ground vegetation that impede or facilitate the catch (Greenslade 1964). Although pitfall traps have limitations, the method is the most frequently used approach to monitor ground-dwelling invertebrates and offers a standard and quick solution to collect high numbers of individuals and species at low cost (Woodcock 2005; Knapp et al. 2016; Brown & Matthews 2016). Fitness et al. (2015) suggested that capture rates of cave wētā are species-dependent and can depend on habitat and time of year. Our findings confirm that lethal pitfall traps are useful in catching ground and cave wētā.

The use of squid baits in pitfall traps improves the catch of ground wētā (*Hemiandrus* spp.) and cave wētā (multiple species) by four and three times, respectively. Both, adult and juvenile ground wētā, were attracted to baited pitfall traps but adults were more strongly attracted to squid baits than juveniles. This may imply that adult ground wētā are more responsive to the volatiles of carrion than juveniles. Female ground wētā showed a slightly stronger response than males which may be because they have greater need for protein in their diet to develop eggs, as is common in many insect species (e.g. Kasper et al. 2015).

Ground wētā species have been classified as predominantly predators and scavengers (Cary 1983) but they have also been reported to feed on fruits (Burns 2006; Morgan-Richards et al. 2008). A diverse dietary range has been reported for the opportunistic *Hemiandrus maia* Taylor-Smith et al. (2013), with this species preferring dead insects over vegetable matter (Taylor-Smith et al. 2013). Attraction to squid baits in our study suggests *H. pallitarsis* detect and actively seek carrion and other foods rather than being passive predators.

The use of baits or lures in pitfall traps, such as carrion and dung, has been reported to increase the catch of flies (Diptera), carrion beetles (Seldon & Beggs 2010) and dung beetles (Halffter & Favila 1993; Andresen 2008; Jones et al. 2012), and this probably indicates sensory response to volatiles from bait (Stavert et al. 2014). Similarly, we demonstrated that Scarabaeidae (*Saphobius* spp., dung beetles), Staphylinidae (rove beetles), Hymenoptera (mainly ants), Opiliones (harvestmen), Araneae (spiders) and Carabidae (ground beetles) were attracted to decomposing squid. Carabidae (predominantly *Ctenognathus*, *Holcapsis*, *Mecodema* and *Megadromus* spp. in our samples) were significantly more abundant in baited traps, contrary to what was observed by Seldon & Beggs (2010) who found similar but small numbers of *Ctenognathus*, *Holcapsis* and *Mecodema* in baited and unbaited traps.

The present study was limited by the use of only one attractant. It would be interesting to investigate other bait types and their comparative efficacies. It is also possible that some taxa (e.g. spiders) were not attracted to squid baits directly, but may nevertheless have shown an elevated catch if they were attracted to the movements of struggling invertebrates more often found in baited traps.

Araneae and Amphipoda showed differences in catch due to the position of a pitfall trap relative to other traps. Both taxa were more abundant in the central unbaited traps compared to the peripheral unbaited traps. This could be explained by a generalised attraction to the centre of the line where the squid odour is expected to be concentrated, but with individuals of these taxa being intercepted in nearby unbaited traps as they move up the concentration gradient. All other taxa were unaffected by trap position suggesting that trap placement 5 m apart was sufficient to achieve independence of samples for these taxa. Ward et al. (2001) found that abundance and species richness of ants and beetles to be unaffected when trap spacing increased from five to ten metres apart, but somewhat lower when reduced to 1 m spacing.

In summary, our results provide support for squid baiting as a technique to increase the efficacy of catching a wide range of invertebrates in pitfall traps, with little evidence of interference or shielding when traps are 5 m apart. This could make it a useful method for biodiversity research and monitoring, especially in situations where logistics constrain the amount of time that pitfall traps can be left open. In this study, for example, we were able to combine monitoring of surface-active invertebrates with the timing of standard mammal monitoring protocols.

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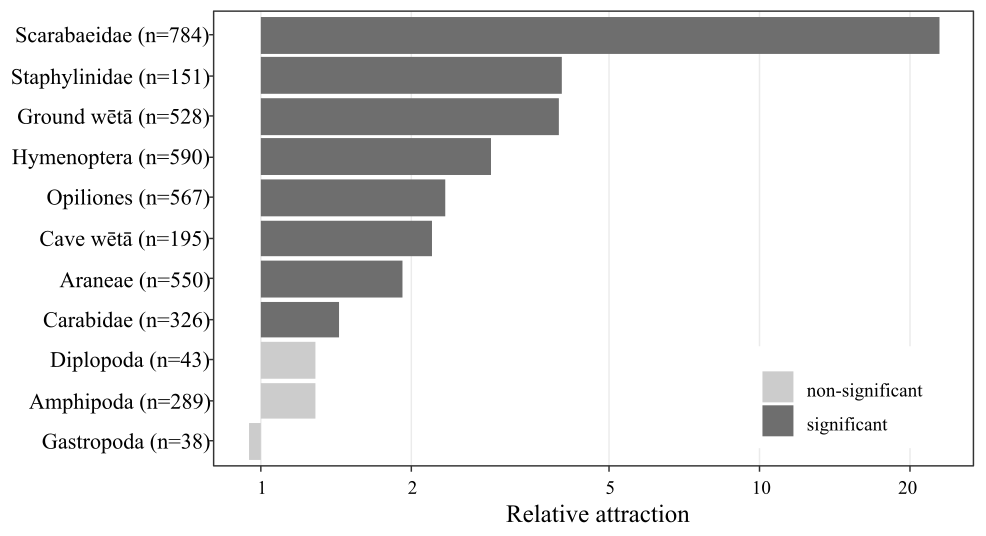
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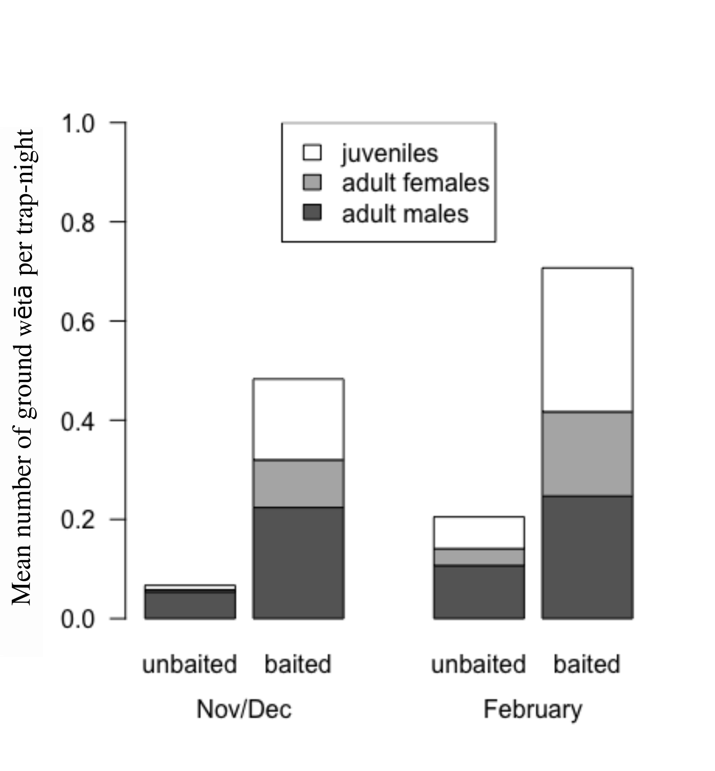
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**Figure 1.** Relative trap-catch of individuals in squid-baited pitfall traps relative to unbaited pitfall traps in the Aorangi and Remutaka ranges, ordered by strength of effect. Dark grey bars denote significant differences in the average number of individuals as a function of baiting while light grey bars represent non-significant differences. Ground wētā = Anostostomatidae, *Hemiandrus* spp. and Cave wētā = Rhaphidophoridae.



**Figure 2.** Comparison between the mean number of adults (males and females) and juvenile related to squid baiting (unbaited and baited-pitfall traps) and season (late-spring (November-December) and late-summer (February)) for ground wētā (*Hemiandrus* spp.) collected in Aorangi and Remutaka ranges over a three-year period (n= 1435 trap nights and 528 individuals sampled).

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**Table 1.** Total number of lines each of seven pitfall traps, by site and season across Aorangi and Remutaka ranges. \*extra sampling for weta species in Aorangi Forest.

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Site** | **Forest** | **Nov-**  **12\*** | **Feb-**  **13\*** | **Nov-**  **13** | **Feb-**  **14** | **Nov-**  **14** | **Feb-**  **15** | **Nov-**  **15** | **No. of traps** | **No. of trap-nights** |
| Bull Hill | Aorangi | 3 | 3 | - | 3 | 3 | 3 | 3 | **126** | **252** |
| Mangatoetoe | Aorangi | - | 2 | 2 | 2 | 2 | 2 | 3 | **91** | **175** |
| Waihora | Aorangi | 1 | 2 | 2 | 2 | 2 | 2 | 3 | **98** | **182** |
| Pinnacles | Aorangi | - | - | 3 | 3 | 3 | 3 | 3 | **105** | **189** |
| Whawanui | Aorangi | 1 | 3 | 3 | 3 | 3 | 3 | 3 | **133** | **259** |
| Orongorongo | Remutaka | - | - | 3 | 3 | 3 | 3 | 3 | **105** | **189** |
| Wairongamai | Remutaka | - | - | 3 | 3 | 3 | 3 | 3 | **105** | **189** |
| **No. of traps** | | **35** | **70** | **112** | **133** | **133** | **133** | **147** | **763** | **-** |
| **No. of trap-nights** | | **35** | **210** | **112** | **399** | **133** | **399** | **147** | **-** | **1,435** |

**Table 2.** Summary of fitted generalized linear mixed models and analysis of variance (Type III, chi-square test) for the effects of squid baiting, position of the pitfall trap and sampling season on the mean abundance of ground-dwelling invertebrates. Values shown are beta coefficients (=natural log of relative difference) with significance of anova indicated by asterisks. Positive main effects of squid, pitfall position or season indicate higher abundance in baited-pitfall traps, central pitfalls or November samples, respectively. NS = non-significant effect (p>0.05), \**p*<0.05, \*\**p*<0.01 and \*\*\**p* < 0.001. Ground wētā=*Hemiandrus* spp., and cave wētā= Rhaphidophoridae.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Taxa | Squid baiting | Pitfall position | Season | Squid\*Pitfall position |
| Ground wētā (all life stages) | 1.326\*\*\* | NS | -0.547\*\*\* | NS |
| adult female | 1.634\*\*\* | NS | -0.769\*\* | NS |
| adult male | 1.599\*\*\* | NS | -0.834\*\*\* | NS |
| juvenile | 1.026\*\*\* | NS | -0.328\* | NS |
| Cave wētā | 0.693\*\*\* | NS | NS | NS |
| Carabidae | 0.348\*\* | NS | 1.118\*\*\* | NS |
| Scarabaeidae | 2.982\*\*\* | NS | 0.531\*\*\* | NS |
| Staphylinidae | 1.529\*\*\* | NS | NS | NS |
| Hymenoptera | 1.193\*\*\* | NS | NS | NS |
| Araneae | 0.828\*\*\* | 0.349\* | NS | NS |
| Opiliones | 0.973\*\*\* | NS | 0.714\*\* | NS |
| Diplopoda | NS | NS | 0.816\*\* | NS |
| Amphipoda | NS | 0.514\*\*\* | 0.466\*\*\* | -0.578\* |
| Gastropoda | NS | NS | 0.915\*\* | NS |