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A methodological approach to assess the small mammal community diversity in the temperate rainforest of Patagonia

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Abstract

Assessing small mammal diversity is a common procedure, which usually employs widespread standard techniques, for gathering information for a wide range of studies. Traditional methods, however, may be biased against capturing arboreal marsupials, such as *Dromiciops gliroides*, an endemic marsupial currently considered a rare species in the Patagonian temperate rainforest due to the low abundances reported previously. I tested a new capturing methodology to assess the small mammal diversity of an old-growth forest in Patagonia, based on a randomized and balanced design, which incorporated a combination of different trap types, bait types, and placement heights. The proposed methodology included four trap types (two for live-capturing: wire-mesh and Sherman traps, and two sign-recording traps for tracks and hair), two types of bait (banana and rolled oats), and two trap placements (ground level and 1.5–2.5 m above the ground). Trap type, bait type, and height of placement all had significantly different effects on capturing and detecting rodents or marsupials; environmental variables at the trap location also affected the ability to detect rodents and marsupials. Traditional methods used for sampling small mammals performed well for rodents but are not effective for capturing marsupials and vice versa, showing species-specific sampling protocols. There is no single combination of trap-bait-height capable to assess the entire small mammal community, but the combination of the most effective protocol for rodents and the most effective protocol for marsupials guarantee better results.

Keywords: Small mammals; Diversity assessment; Capture efficiency; Sampling artifact; Patagonian temperate rainforest

Introduction

Assessments of small mammal community are very common to describe biologic systems (e.g., Kelt 2000), habitat use (e.g., Simonetti 1989), estimate distribution patterns (e.g., Umetsu et al. 2008) or prey availability (e.g., Rau et al. 1995), among many others. Due to its widespread use, standard sampling protocols have been developed and used extensively with little or no adaption to the study subject (e.g., Wheatley and Larsen 2008). The traditional methods commonly employed use only classic Sherman traps, baited with oats and placed at the ground level.

The small mammal community of the Patagonian temperate rainforest is mainly composed of rodents of the genus *Abrothrix* and *Oligoryzomys longicaudatus*, and less common species such as *Geoxus valdivianus* and *Irenomys tarsalis*, and occasionally exotic rats (*Rattus rattus*, *R. norvegicus*) are captured (Meserve et al. 1999; Kelt 2000). Distinctive components of this community are the marsupials, represented by two endemic species: *Dromiciops gliroides* and *Rhyncholestes raphanurus*, both species have a restricted distribution in the temperate

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rainforest of Patagonia (Marshall 1978; Saavedra and Simonetti 2001; Lobos et al. 2006), including the Coastal Range, the Andes, and the intermediate depression (Kelt and Martínez 1989).

Dromiciops gliroides is an arboreal marsupial (Jiménez and Rageot 1979), physiologically and morphologically adapted to the cool and moist southern temperate forest (Hershkovitz 1999; Bozinovic et al. 2004). There is no consensus about the use of the vertical stratum by this species, because it is occasionally captured at ground level (Patterson et al. 1989; Patterson et al. 1990) and at a variety of canopy heights (Aizen 2003; Rodríguez-Cabal et al. 2007).

Dromiciops gliroides generally is considered rare because it is seldom detected and exhibits low numbers when small mammals are assessed with traditional methods (Meserve et al. 1988; Patterson et al. 1989; Kelt 2000). However, the low representation in the capture rate may be due to an estimation bias related to the sampling methods used, traditionally targeted for cursorial species, which may not be adequate for capturing arboreal mammals (Kelt and Martínez 1989; Rau et al. 1995; Lindenmayer et al. 1999); previous studies conducted using non-traditional sampling techniques reported high *D. gliroides* abundances (Amico and Aizen 2000; Rodríguez-Cabal et al. 2007; Rodríguez-Cabal et al. 2008; García et al. 2009).

I evaluated the effects of different trap-bait-height combinations, as well as the influence of environmental characteristics, on the capture success of different components of the small mammal community of the Patagonian temperate rainforest.

My hypothesis was that the traditional sampling protocols may bias the representation of some species that use the vertical stratum or are not attracted by the traditional baits. The aim of this study was to determine the most appropriate protocol to assess the small mammal diversity in the temperate rainforest of Patagonia.

Material and methods

Study site

My study site (41°07′05″S, 72°36′50″W) was located closer to the Osorno Volcano and Las Cascadas town, in southern Chile. It was a ca. 20 ha well-preserved old-growth forest remnant, immersed in a complex agricultural landscape. Forest canopy was dominated by *Gevuina avellana*, *Caldcluvia paniculata*, *Eucryphia cordifolia*, and *Embothrium coccineum*, with several emergent individuals of *Nothofagus dombeyi*, intermediate overstory was dominated by juvenile trees of *E. cordifolia*, *C. paniculata*, *Weinmannia trichosperma*, *Luma apiculata*, *Lomatia ferruginea*, *Raphitamnus spinosus*, *Aextoxicon punctatum*, *Aristotelia chilensis*, and the native bamboo *Chusquea quila*. The understory was composed of tree saplings, mosses, several ferns, and a thick litter layer with abundant fallen logs.

Sampling protocol

To assess the small mammal community diversity, I used two types of live-capturing traps: the standard Sherman traps ($23\times9\times8$ cm) and wire-mesh (Tomahawk-style) traps, specifically designed for the capture of arboreal small mammals ($26\times13\times13$ cm). I also tested two types of sign-recording traps: footprint-recording tubes and hair-collecting tubes. The former were made using a PVC pipe (20 cm long, 7.5 cm diameter) with a smoked aluminum plate inside. The latter were made using a PVC pipe (20 cm long, 5 cm diameter) with double side adhesive tape affixed to its interior. The four monitoring traps were baited with banana slices or rolled oats, and placed either at ground level or above the ground (1.5–2.5 m). The monitoring design using all trap-bait-height sets rendered 16 unique combinations.

Traps were placed in six lines, dispersed in the forest area, having 32 traps in each line, each trap combination was replicated twice in each line using a full-randomized and balanced design. Trap lines were operated for six consecutive nights, with a total effort of 1152 trapnights, during the austral summer (throughout March). Traps were checked once daily early at dawn. All the animal capturing and handling procedures followed the guidelines of the American Society of Mammalogists (Gannon et al. 2007), which were also approved and authorized as well, by the Chilean Agriculture and Livestock Bureau (SAG). I followed biosafety procedures proposed by Mills et al. (1995) considering that *Oligoryzomys longicaudatus*, a known hantavirus reservoir rodent, is present in the study area and it is frequently captured.

Captured individuals were identified to species level, and marked by cutting their fur in unique patterns. Also were measured, sexed, aged (adult or juvenile), weighed, and their reproductive condition was assessed (based on the external genitalia). All animals were released in the same place where captured. For sign-recording traps, I identified visitors as rodent or marsupial, based on signs left (footprints, hairs, or fecal pellets). When feasible signs for identification were absent and bait was eaten, the event was recorded as “bait removed”.

In order to determine the optimal trap placement location, I measured the following four habitat characteristics (Rudran and Foster 1996; Brower et al. 1998): (1) height of trap placement (measuring tape, cm), (2) tree branch diameter where the trap was set (caliper, cm), (3) branch slope (for diameter and slope, a zero value was assumed for traps placed on the ground; protractor, degrees), and (4) the type of substratum where each trap was placed (litter, live tree branch, or dead tree branch).

Data analyses

I evaluated the effect of each trap-bait-height combination using logistic regression analyses with STATISTICA 7 (StatSoft 2004). I defined the response variable as a binary variable (0 = no capture, 1 = capture), the predictor variables (categorical) were trap type, bait type and height of placement.

To assess the significant effects for each case, I ran separate analyses for rodents (represented mainly by *Abrothrix olivaceus*), for marsupials (represented by *D. gliroides*), and for all species combined, as well as for the live-capturing traps subset, the sign-recording traps subset and the combination of both subsets. For assessing differential effects between the live-capturing vs. sign-recording traps, live-capturing and sign-recording traps were nested in a "trap category variable", and depending on it, a "trap type" (Sherman or wire-mesh, footprint or hair) variable. For testing significant effects over the whole small mammal capture probability, I also performed an ordinal multinomial logistic regression combining marsupial and rodents' data in the response variable (1 = no capture, 2 = marsupial only, 3 = rodent only, and 4 = both). The purpose of this multiple test was to distinguish if there were differential effects when marsupials and rodents were analyzed independently and as a whole. Goodness-of-fit of the models was estimated by a Hosmer–Lemeshow test (Agresti 2007).

I used Akaike Information Criterion (AIC, Akaike 1973) to select the best model subset that explained the tendencies recorded in the study. When the sample size/number of factors (n/K) ratio was <40, the AIC values were corrected (obtaining an AIC_c estimate) following Burnham and Anderson (2002). For interpretation I present ΔAIC and AIC weights (w_i) (Burnham and Anderson 2002). Following Burnham and Anderson (2002), models are considered equivalent when $\Delta AIC<2$, and the explicative model or model subset must account $\geqslant 90\%$ of the w_i (only that model subset is presented in the tables).

To assess the site-specific variables that determine capture success of marsupials, I used data measured at each trap location to perform a logistic regression using three continuous predictors (branch diameter, branch slope, and height of placement) and one categorical predictor (substratum type). The continuous variables were tested for multicollinearity using multiple regressions. In all cases the Variation Inflation Factor was <5 and the Tolerance values were between 0.42 and 0.78 (showing no multicollinearity). I performed the same type of analysis for capture data on rodents. However, the lack of variability between the response variable and the substratum variable precluded inclusion of this variable it in the model. Consequently, only the three continuous variables were used to perform this logistic regression. For analyses on marsupials as well as rodents, the best model was selected using AIC. All procedures were run in STATISTICA 7 (StatSoft 2004).

Results

Diversity assessment and differential capturing

25 individuals of *D. gliroides* and 33 individuals of *Abrothrix olivaceus* were captured, in both cases I had a recapture rate of about 30%. Capture rates of marsupials and rodents differed with respect to the trap-bait-height combination used. Some trap combinations were more efficient in capturing marsupials whereas others were superior for rodents (Fig. 1). For *D. gliroides*, the best combination (17% success respect to the whole trap set) was wire-mesh traps, baited with banana, and placed above the ground, followed by Sherman traps, baited with banana, and placed above the ground (9%). These sets performed poorly for *Abrothrix olivaceus* (1%). Conversely, wire-mesh and Sherman traps, baited with oats or banana, but placed at ground level performed well for capturing *A. olivaceus* (14% and 17%, respectively), but poorly for *D. gliroides* (0% and 3%, respectively). Traps placed above the ground and baited with oats, were entirely unsuccessful, independently of trap type.

A similar trend was observed when using the sign-recording traps. Tubes placed above the ground and baited with banana recorded more *D. gliroides* (17 vs. 8 records) and fewer *A. olivaceus* signs (21 vs. 161 records) than those placed at ground level. Tubes placed at the ground level and baited with oats registered 97 *A. olivaceus* signs and no *D. gliroides* signs (Fig. 2). Large numbers of tracks were recorded for *D. gliroides* (14 records) and *A. olivaceus* (18 records) when baited with banana and placed above the ground. Hair-sampling tubes performed poorly compared to the other techniques (1 record vs. 162 records).

Model selection approach

Trends described above were confirmed with logistic regression and applying model selection procedures to select the most parsimonious model subsets for each case. For live-capturing data, I found two models in the $w_i \geqslant 90\%$ subset for *D. gliroides* data, four models for *A. olivaceus*' data, and three models for both *D. gliroides* and *A. olivaceus* data (Table 1).

I obtained similar results for sign-recording data (Table 2), where trap type, bait type, and placement

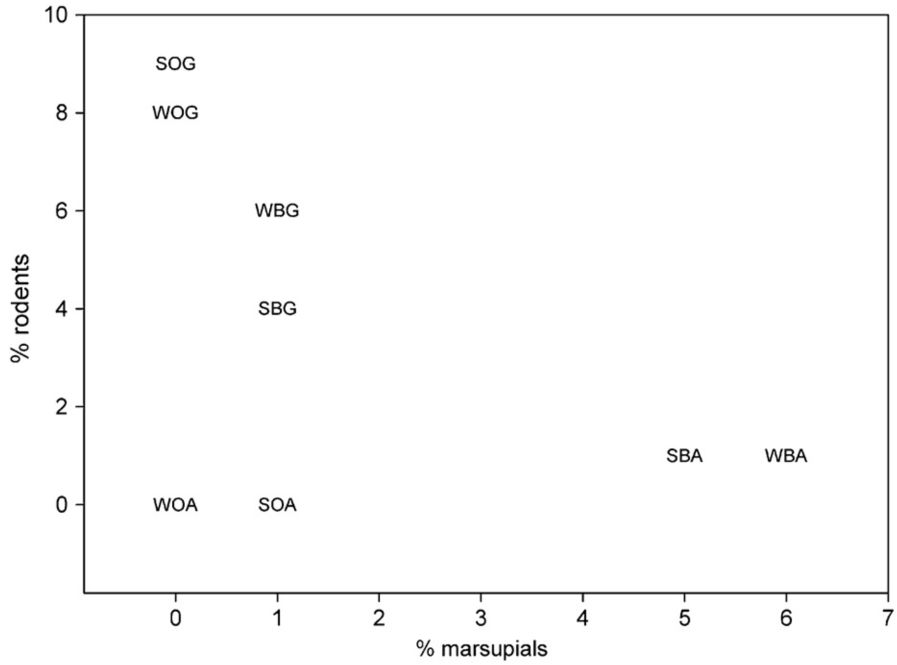

Fig. 1. Marsupials vs. rodents live-capturing rates in relation to each trap-bait-height combinations. Traps are W = wire-mesh, S = Sherman; baits are B = banana, O = oats; and heights are G = ground level, A = above the ground.

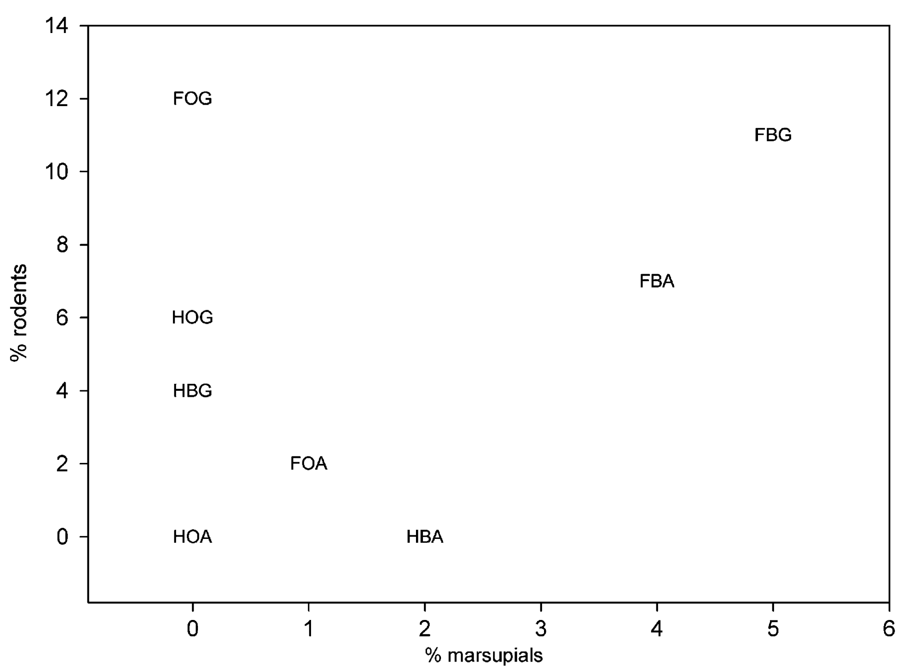

Fig. 2. Marsupials vs. rodents sign-recording rates in relation to each trap-bait-height combinations. Traps are F = footprint tube, H = hair-collecting tube; baits are B = banana, O = oats; and heights are G = ground level, A = above the ground.

height effects combined provided the best explanation for success. Conversely, when the logistic regression was run for the whole trap set, I obtained four models in the $w_i \geqslant 90\%$ subset for *D. gliroides*, five models for *A. olivaceus*, and four models for the combined data (Table 3).

Site-specific variables assessment

Logistic regression applied to the site-specific environmental variables showed that trap height provided the best explanation for *D. gliroides* capture success (AIC = 292.4848, $w_i = 0.3567$), and branch inclination

Table 1. Model selection for live-capturing data.

Par 1	Par 2	Par 3	df	AIC_c	ΔAIC_c	Weight AIC_c
Live-capturing data for marsupials						
B	H		2	61.5201	0.0000	0.7545
T	B	H	3	63.6519	2.3927	0.2281
Live-capturing data for rodents						
H			1	86.4604	0.0000	0.3962
B	H		2	86.7117	0.2513	0.3494
T	H		2	88.4740	2.0136	0.1448
Live-capturing data for marsupials and rodents						
H			1	177.1440	0.0000	0.4738
B	H		2	178.4924	1.3484	0.2414
T	H		2	178.8830	1.7389	0.1986

Parameters in the models are T = trap, B = bait, and H = placement height.

Table 2. Model selection for sign-recording data.

Par 1	Par 2	Par 3	df	AIC_c	ΔAIC_c	Weight AIC_c
Sign-recording data for marsupials						
T	B		2	60.2457	0.0000	0.6649
T	B	H	3	61.8964	1.9116	0.2556
Sign-recording data for rodents						
T	H		2	79.5004	0.0000	0.7376
T	B	H	3	81.3067	2.0673	0.2624
Sign-recording data for marsupials and rodents						
T	B	H	3	159.6862	0.0000	0.7937
T	H		2	162.6425	2.6955	0.2062

Parameters and acronyms as in Table 1.

(AIC = 293.7319, $w_i = 0.1898$) and diameter (AIC = 294.4666, $w_i = 0.1324$) in a second instance. For rodent capture success, trap height and branch diameter provided the best explanation (AIC = 240.8785, $w_i = 0.5705$), whereas the interaction between height, diameter and inclination were important in a second instance (AIC = 242.2179, $w_i = 0.2860$).

Discussion

Capture results indicated a spatial segregation between arboreal marsupials and rodents, the former tended to be found above the ground, whereas the latter were found at the ground level. That differential capturabilty suggest resource partitioning (Meserve 1981; Simonetti 1989; Kelt et al. 1994), in which *D. gliroides* exploits mainly the canopy resources, whereas rodents exploit mainly the understory resources, resulting in a reduced spatial (vertical) overlap and lower interspecific competition levels (Meserve et al. 1988; Kelt et al. 1995).

Apart from the vertical segregation, the bait used also had a differential effect on the capturability of marsupial and rodents, because the former preferred banana and the latter consumed both banana and oats as bait. Bait preference by *D. gliroides* partially explains the lower capture rates previously reported. (e.g., Meserve et al. 1982; Patterson et al. 1989; Meserve et al. 1999; Kelt 2000), in which the sampling protocols used only oats as standard bait. I recorded only one case of a *D. gliroides* captured in an oat-baited trap, which seemed to be a chance event, as the captured individual did not eat the bait.

Table 3. Model selection for live-capturing and sign-recording data, with trap type (wire-mesh or Sherman, and footprint-recording or hair-collecting) variable nested into a trap category (live-capturing or sign-recording) variable.

Par 1	Par 2	Par 3	Par 4	df	AIC	ΔAIC	Weight AIC
Live-capturing and sign-recording data for marsupials							
T	B	H		3	124.4947	0.0000	0.4518
B	H			2	125.9657	1.4710	0.2165
C	T	B	H	4	126.2736	1.7789	0.1856
C	B	H		3	127.7517	3.2570	0.0886
Live-capturing and sign-recording data for rodents							
C	T	H		3	180.4053	0.0000	0.6118
C	T	B	H	4	182.0826	1.6773	0.2645
T	H			2	184.3233	3.9180	0.0863
Live-capturing and sign-recording data for both marsupials and rodents							
C	T	B	H	4	357.1905	0.0000	0.4244
T	B	H		3	358.4136	1.2230	0.2302
C	T	H		3	358.6400	1.4494	0.2056
T	H			2	359.4154	2.2248	0.1395

Parameters in the models are C = trap category, T = trap type, B = bait, and H = height of placement.

Models obtained through logistic regressions indicate that trap type had a secondary effect on capture of *D. gliroides*, and no effect for *A. olivaceus* capturing. Empirical evidence suggested that more *D. gliroides* were captured with wire-mesh traps than with Sherman traps, in agreement with Hershkovitz's (1999) statements, who described that *D. gliroides* had a strong dislike to closed traps (such as the Sherman traps). When analyzing the combined effects of trap type, bait type, and placement height, I distinguished species-specific sampling protocols, derived from the tested combinations. Wire-mesh traps baited with banana and placed above the ground was the most efficient combination for capturing *D. gliroides* (but placing the traps above the ground resulted in the exclusion of capturing of the majority of the rodents), whereas the traditional Sherman traps, baited with oats and placed in the ground performed well for rodents but not for marsupials. In order to study the entire small mammal community in the Patagonia rainforest, the combination of trap combinations is essential.

Live-capturing and sign-recording traps are different in ability to accurately sample small mammals, these findings are in agreement with previous reports (Taylor and Raphael 1988; Carey and Witt 1991; Mortelliti and Boitani 2008) that showed that live-capture and sign-capture data or abundance estimates obtained from them are not comparable. In this study, I focused on live-capturing traps because they gave a high level of confidence for the identity of the captured animal (including individualized recognition when recaptured). Additionally, sign-recording techniques provided many more records than live-capturing traps. We must interpret such information with caution, as a higher number of footprints, hair, or other signs records do not necessarily imply a higher abundance or an under-representation of the live-capture sampling. It is possible that the same individual may visited several traps in one night.

Although the above, the use of sign-recording traps may allow us to improve our understanding of the three-dimensional use of the forest by rodents and marsupials. The occurrence of many *D. gliroides* sign records at banana-baited ground traps suggest that this species also searches for food at the ground level, as confirmed with four live-captures at ground level. Comparing the two sign-recording traps, the track plates performed much better than the hair traps, because in the latter the identification turned out to be more difficult and inaccurate.

The differential capture success may depend on environmental variables, analyzing these variables may help to improve trap placement and improve the capturing success. For capturing marsupials, the "best" model in the subset derived from logistic regression showed that the height of the trap was the principal factor influencing capture success, in agreement with the sampling combinations discussed previously. The following "best" model, however, considered the interaction between height and branch slope ($w_i = 0.1898$), and between height and branch diameter ($w_i = 0.1324$), suggesting that both variables may have a secondary effect in the captures. The model-derived information is consistent with my field observations, as I observed that *D. gliroides* preferably used thin and horizontal branches to move through the forest, instead of using larger and vertical trunks.

For *A. olivaceus*, trap height and branch diameter effects significantly influenced capture success but the second "best" model also included the branch slope. We should be cautious when interpreting these models, because the assessed environmental variables had different meanings for *D. gliroides* and rodents, as the former is arboreal and the latter are either cursorial (e.g., *Abrothrix olivaceus*, the most common species in the study site) or scansorial (e.g., *Oligoryzomys longicaudatus*). Cursorial rodents may occasionally fall in traps placed above the ground when they had an "easy access" through a low-slope and medium-diameter branch. Scansorial rodents can reach traps placed >1 m above the ground level as found by Gallardo-Santis et al. (2005) and Rau et al. (1995).

Considering many trap-bait-height combinations in the diversity assessment protocols may improve the capturing success of small mammal because these combinations are complementary and species-specific. There is no single combination capable of sampling the entire small mammal community of the Patagonian temperate rainforest, because there are ground-dwelling rodents, and arboreal marsupials and/or rodents that do not respond in the same way than the former. Additionally, considering some key environmental variables when deciding the most appropriate placement trap location may also improve the capture success, and consequently, improve the small mammal community sampling, resulting in better species richness and abundance estimations.

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