

1 **Experimental evidence that even minor livestock trampling has severe**  
2 **effects on land snail communities in forest remnants**

3

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## 27 **Summary**

28 **1.** Land-use intensification is increasing dramatically in production systems worldwide.

29 Livestock production is an important component of production land-use and increases in  
30 livestock densities have had a wide range of negative consequences. The off-site effects of  
31 livestock grazing and trampling on native vegetation adjacent to pastoral land have received  
32 less attention than on-farm effects. Moreover, where significant ecological effects of  
33 livestock spillover have been identified, the mechanistic determinants of these effects have  
34 not typically been investigated.

35 **2.** Here, we tested the mechanistic drivers of livestock trampling effects on land snail  
36 communities in forest remnants using simulated trampling under field conditions. We used a  
37 factorial combination of leaf-litter manipulation and trampling treatments to partition  
38 different causal drivers of livestock impacts on land snail communities, and related them to  
39 five environmental variables that are altered by livestock.

40 **3.** We show that even very low frequency trampling caused severe changes to land snail  
41 communities. Land snail density, even under the lowest trampling frequency, declined by an  
42 average of 42 individuals m<sup>-2</sup> and land snail species richness decreased by an average of 10  
43 species per plot compared with control plots.

44 **4.** The underlying drivers of changes in land snail communities varied, but were primarily  
45 linked to leaf-litter mass, rather than soil compaction.

46 **5. *Synthesis and applications.*** Overall, these results suggest that even minimal disturbance by  
47 livestock has large effects on land snail communities but the underlying drivers of these  
48 effects require further investigation in longer duration and more intensive studies. Our results  
49 provide strong support for livestock exclusion as an important management tool for native  
50 forest remnants embedded within production landscapes.

51

52 **Keywords:** pulse disturbance, land snails, livestock exclusion, fencing, forest fragmentation,  
53 trampling, spillover

54

## 55 **Introduction**

56 Land-use intensification is increasing dramatically in production systems worldwide, with  
57 significant effects on biodiversity and ecosystem functioning (Green *et al.* 2005; Cunningham  
58 *et al.* 2013). In particular, there are growing concerns about the ecological impacts of  
59 livestock production given it has expanded greatly in land area and intensification in recent  
60 years (Tilman *et al.* 2002). Over 45% of the world's land surface area is now occupied in  
61 some form by livestock systems, including both permanent grazing pastures and off-site  
62 production of livestock feed crops (Herrero *et al.* 2009). Intensification of agri-chemical  
63 inputs, greater reliance on irrigation, and increases in imported feedstock have led to a  
64 dramatic rise in production capacity (Dorrough, Moll & Crosthwaite 2007). For example, in  
65 the 40 years prior to 2002, global per capita meat production increased by >60% (Tilman *et*  
66 *al.* 2002). Furthermore, these trends are only predicted to increase with a projected doubling  
67 of global meat demand by 2050 (e.g. Tilman *et al.* 2002; Herrero *et al.* 2009).

68         Livestock production has contributed significantly to land degradation (Pietola, Horn  
69 & Yli-Halla 2005) and greenhouse gas emissions (Herrero *et al.* 2009). Even in systems  
70 where large herbivores have historically contributed to ecosystem dynamics (e.g. the African  
71 savanna and the North America prairies), increases in livestock densities have had many  
72 negative effects through grazing and trampling (Biondini, Patton & Nyren 1998; Savadogo,  
73 Savadogo & Tiveau 2007). For example, overstocking can lead to unsustainable decreases in  
74 vegetation biomass, which increases the need for imported feedstock, and can facilitate  
75 establishment of invasive plants (Roques, O'Connor & Watkinson 2001). High-intensity  
76 livestock trampling can also have severe direct and indirect effects on the soil, causing

77 erosion, changes in nutrient availability, altered soil microbial communities, and negative  
78 effects on invertebrate communities (Bromham *et al.* 1999; Pietola, Horn & Yli-Halla 2005;  
79 Jeddi & Chaieb 2010).

80         The off-site effects of livestock grazing and trampling in native habitat remnants  
81 within pastoral land have been less well-studied than effects on the pasture itself, but  
82 nevertheless can be severe (e.g. Close, Davidson & Watson 2008; Dodd *et al.* 2011). The  
83 importance of these off-site effects for understorey plant communities (Naeth *et al.* 1991;  
84 Burns *et al.* 2011) and soils (Pietola, Horn & Yli-Halla 2005; Jeddi & Chaieb 2010) is  
85 reflected in the extensive evidence for differences in biodiversity between fenced and  
86 unfenced forest remnants (e.g. Bromham *et al.* 1999; Didham *et al.* 2009). Consequently,  
87 worldwide, fencing to exclude livestock is an increasingly common management option for  
88 the conservation of native habitat remnants embedded within production landscapes  
89 (Hayward & Kerley 2009; Burns *et al.* 2011). However, in many other regions where  
90 livestock still have access to native remnants, the off-site or ‘spillover’ effects of livestock  
91 grazing and trampling are only likely to become more significant in the future as livestock  
92 densities increase.

93         Surprisingly, in the majority of cases where significant ecological alterations from  
94 livestock spillover have been identified, the mechanistic determinants of these changes have  
95 not been investigated. Loss of vegetation and changes to soil structure are the most obvious  
96 changes to native remnant ecosystems caused by livestock, but it can be difficult to separate  
97 these factors from other potential correlates, such as variation in soil and litter moisture  
98 levels, amount of litter, or nutrient deposition (e.g. Didham *et al.* 2009). Moreover, there  
99 might be interactions between multiple components of livestock effects which could  
100 exacerbate ecosystem-level perturbation as livestock density increases. For example, it is not  
101 known whether the effects of livestock trampling scale proportionately with variation in

102 livestock density per unit area per unit time, or whether there are thresholds of livestock  
103 trampling above which more substantial effects on ecosystems might occur. This information  
104 is vital to assess the environmental consequences of intensifying livestock production, and to  
105 devise mitigation strategies to limit livestock impacts in native remnant ecosystems  
106 embedded within production landscapes.

107         In this study we test the mechanistic determinants of livestock trampling effects on  
108 land snail communities in native habitat remnants embedded within production landscapes,  
109 using a simulated trampling experiment conducted under field conditions in New Zealand  
110 (NZ). Land snails were studied because they are key components of the detritivore fauna  
111 critical for litter decomposition in mesic forests. In NZ, land snail communities have high  
112 micro-scale sympatric abundance (100–6000 individuals m<sup>-2</sup>) and richness (10–50 species m<sup>-2</sup>)  
113 (Barker & Mayhill 1999), similar to other key detritivore taxa such as oribatid mites and  
114 Collembola. Land snails are highly responsive to environmental disturbance (Baur & Baur  
115 1995; Boschi 2007, Douglas, Brown & Pederson 2013), therefore the diverse communities in  
116 NZ systems provide excellent models for testing livestock effects that manifest through  
117 modification of the forest litter layer and underlying soil properties.

118         We use a combination of litter manipulation and trampling intensity treatments to  
119 partition different drivers of changes in land snail communities due to livestock, and relate  
120 treatment differences to covariance in five proximate measures of litter structure and  
121 microclimate (leaf-litter mass, leaf-litter complexity, leaf-litter moisture content, soil  
122 moisture content and soil compaction) which, typically change in relation to intensity of  
123 livestock trampling (Naeth *et al.* 1991; Close, Davidson & Watson 2008). Our experiment  
124 provides a quantitative test of the scaling of ecological change with increasing intensity of  
125 livestock trampling, and discriminates the dominant mechanistic pathways through which  
126 these effects operate.

127

## 128 **Materials and methods**

### 129 *Experimental Site*

130 The experiment was conducted in a remnant native forest on farmland in the Waipa District,  
131 which is located in the Waikato Region of the North Island, NZ. The majority of forest cover  
132 in NZ was cleared for farming in the early 20<sup>th</sup> century, leaving most lowland native  
133 vegetation as small remnants embedded within production systems, most of which were open  
134 to livestock for the first 50 or more years of farming (Burns *et al.* 2011). The experimental  
135 site was typical of this situation, and was selected because it has been reliably fenced to  
136 exclude livestock for over 20 years, ensuring time for recovery of leaf-litter invertebrates  
137 (Didham *et al.* 2009). All areas sampled were at least 20 m from the remnant edge.

138

### 139 *Experimental Design*

140 The mechanistic basis for livestock effects on land snail communities was tested in a Before-  
141 After-Control-Impact (BACI) design (Appendix S1 in Supporting Information), using a  
142 factorial combination of four levels of simulated trampling (0, 2, 4, or 6 trampling events)  
143 crossed with three levels of litter manipulation (litter not exposed to trampling or reduced in  
144 volume, litter exposed to trampling but not reduced in volume, and litter exposed to trampling  
145 and reduced by 50% in volume). The 12 treatment combinations were applied to separate 1.2  
146 × 3.0 m plots, and were replicated three times in a randomised block design (36 plots in  
147 total). The three blocks were determined by spatial location within the forest remnant and by  
148 percentage ground cover of thread fern *Blechnum filiforme* (Fig. S1), a common species at the  
149 site which varied from 0–95 % cover among the blocks.

150 A mechanical hoof (Appendix S1) was used to simulate trampling, enabling

151 randomised treading but high repeatability of trampling intensities. In addition, a live cow

152 could have had confounding effects through browsing and nutrient enrichment which were  
153 not the focus of the study. The mechanical hoof consisted of a compressor-driven pneumatic  
154 ram with a cow hoof attached (Fig. 1). The trampling pressure was set at 220 kPa to simulate  
155 treading by an adult Friesian cow (Di *et al.* 2001). The mechanical hoof was mounted on a  
156 steel frame that allowed us to move the hoof to any point within the plots. To ensure variation  
157 in spatial offset, or overlap, of individual hoof compressions, we marked 5-cm intervals on  
158 the trolley and frame assembly to project a  $5 \times 5$  cm grid of 720 possible compression points  
159 onto the plot surface, and randomly allocated 360 hoof compressions within the grid  
160 (Appendix S1). In practice, individual points were trampled between 0 and 4 times at each  
161 trampling event, equating to a small herd of approximately 45 adult cows traversing a  
162 remnant without stopping. This level of intensity was chosen by selecting a realistic range of  
163 livestock density for the region (based on surveys across a much wider range of farms in the  
164 same area, Appendix S1, Table S1, S2). Then we scaled from only two trampling events over  
165 a six-week period (paddock rotations are typically every 5–6 weeks) through to a more  
166 intensive multiple trampling regime (weekly trampling events) that could occur through  
167 continued incursions or a larger cow herd (Appendix S1).

168 Three litter manipulation treatments were designed to discriminate the effects of soil  
169 compaction from the effects of changes in leaf-litter volume. First, we combined trampling of  
170 both the soil and the litter (SL treatment), without manipulating litter cover. The plots within  
171 this treatment where trampling was not applied (SL0) represent the experimental controls. To  
172 separate soil compaction effects from other treatment effects, we included a soil-only  
173 trampling treatment (S). In this treatment, all leaf-litter and friable humus was removed from  
174 within the plot before trampling and immediately spread evenly back onto the plot  
175 afterwards. Finally, the effect of reduced litter volume, in combination with trampling, was  
176 determined using a ‘half litter removed’ treatment (SLR), where all of the litter was removed

177 from the plots as above, and then only half was spread evenly back onto the plots. We chose a  
178 50% litter reduction treatment based on the positive relationship observed between litter mass  
179 and time since livestock exclusion in an earlier study within the same region (Didham *et al.*  
180 2009), with unfenced forest remnants having approximately 50% of the litter mass than in  
181 remnants that had been fenced for 20–40 years. Litter reduction was carried out once before  
182 the first trampling event in all of the SLR plots, including the plots that were not trampled  
183 (SLR0).

184

### 185 *Sampling methods*

186 In each of the 36 plots, samples were collected both pre-treatment and post-treatment as per  
187 the BACI design. On each occasion, leaf-litter samples, penetration resistance measurements  
188 and soil samples were taken at four random points within each plot. Pre- and post-treatment  
189 samples were taken from different sampling points within each plot.

190 First, the litter samples were collected from each sampling point using a 33-cm  
191 diameter circular frame (0.086 m<sup>2</sup>) (Didham *et al.* 2009). All leaf-litter and friable humus was  
192 scraped from inside the frame and placed into a large cloth bag. The samples were weighed to  
193 obtain total wet litter mass, and then sieved through a 10-mm sieve to obtain a fine ‘bottom  
194 fraction’ containing land snails, and a coarse ‘top fraction’. Both the top and bottom fractions  
195 of litter were dried and weighed to obtain a total dry litter mass, which was converted to mass  
196 m<sup>-2</sup> for analysis. Moisture content of the litter was calculated as a percentage of the dry litter  
197 mass (Appendix S1). The “bottom fraction” of litter was then passed through three more  
198 sieves of mesh sizes 4.75 mm, 2.00 mm and 0.50 mm, resulting in four size classes that were  
199 weighed. These weights were used to obtain a measure of litter complexity calculated as  
200 Simpson’s diversity in Biodiversity Professional software (BD Pro, version 2.0).



201 The 'bottom fraction' of litter was searched for land snails with a stereomicroscope.  
202 The total number of individuals found in each plot (snail abundance) was converted to  
203 number m<sup>-2</sup> before analysis. All land snails were identified to species where possible, by  
204 reference to authoritatively-identified material (including vouchers for undescribed species)  
205 held in the Museum of New Zealand Te Papa Tongarewa (NMNZ). Nomenclature was  
206 standardised to that used by Spencer *et al.* (2009). Only snails that were live when sampled  
207 (determined by the presence of dried tissue within the shell) were included in analysis.

208 Penetration resistance (soil compaction) was measured at the centre of the bare  
209 ground left after sampling the pre-treatment litter. Measurements were taken to a depth of 10  
210 cm using a penetrometer (Eijkelkamppetrologger) with a 2-cm<sup>2</sup> cone and 60° top angles.  
211 The post-treatment measurement was taken as near as possible to the pre-treatment  
212 measurement (unlike the other measurements which were taken at a different sampling point)  
213 as there can be high variability in penetration resistance over small distances. Lastly, a 2 cm  
214 diameter soil core was taken to a depth of 10 cm at the centre of the patch made bare by litter  
215 removal. The samples were weighed immediately, and then dried to a constant weight at  
216 105°C. Moisture content was calculated as a percentage of the dry soil weight (Appendix S1).

217

## 218 *Statistical Analysis*

### 219 *Determining completeness of sampling effort pre- and post-treatment*

220 Across both pre-and post-treatment samples, the completeness of land snail community  
221 sampling and species richness estimation was evaluated using sample-based species  
222 accumulation curves, re-scaled to number of individuals, in EstimateS version 8.2.0 (Colwell  
223 2005).

224

225 *Determining variation in land snail community composition pre- and post-treatment*

226 Post-treatment plot-level measures of land snail abundance  $m^{-2}$  and land snail richness were  
227 standardised using post-treatment minus pre-treatment values to account for spatial variation  
228 within plots and temporal fluctuations between sampling periods. Plot-standardised values  
229 were then converted to treatment minus control differentials. While it is possible there could  
230 have been lateral movement of land snails between control and treatment plots, we consider  
231 that the magnitude of this effect is likely to be small (movement rates for land snails of the  
232 average size of our species are ca 100 mm  $day^{-1}$ ; Baur & Baur 1988), and would only serve to  
233 make the measured treatment effects more conservative.

234 To compare land snail community composition between treatment plots we calculated  
235 variation in community dissimilarity to control plots based on an ordination of snail species-  
236 abundance distributions. Non-metric Multi-dimensional Scaling (NMDS) ordination was  
237 performed on square root transformed abundance data using the Bray-Curtis measure of  
238 similarity in Primer v.6 (Carr 1994). Formal significance tests for differences between pre-  
239 and post-treatment samples were conducted using the ANOSIM permutation test (Clarke  
240 1993). Variability (i.e. dispersion) of snail community composition between pre- and post-  
241 treatment samples was also tested in Primer using permutational analysis of multivariate  
242 dispersions (PERMDISP: Anderson 2006). Subsequently, community dissimilarity to control  
243 plots was determined from the underlying Bray-Curtis distance matrix, using the average  
244 dissimilarity between each treatment plot and the three control plots. Post-treatment measures  
245 of snail community composition were standardised using post-treatment minus pre-treatment  
246 values.

247

248 *Determining mechanisms of change in land snail communities associated with trampling and*  
249 *litter treatments*

250 We constructed structural equation models (SEM) in Amos version 19.0 (Appendix S1) to  
251 partition the direct versus indirect mechanistic pathways through which trampling intensity  
252 and changes in litter volume might influence land snail communities. All SEM analyses were  
253 carried out on plot-standardised treatment minus control differentials as described above.

254 A causal hypothesis of relationships among variables was created for each response  
255 variable (i.e. the ‘full’ SEM models, Fig. S2), with litter treatments, trampling treatments and  
256 their interaction (‘exogenous’ predictors) having both direct and indirect effects on the  
257 response variables. We hypothesised that the treatments could have had indirect effects  
258 through a series of habitat structure and microclimatic variables (‘endogenous’ predictors)  
259 that were predicted to be important for land snail abundance and diversity: leaf-litter mass,  
260 leaf-litter moisture content, soil moisture content, penetration resistance (soil compaction),  
261 and leaf-litter complexity. In the SEM of land snail species richness, land snail abundance  
262 was also entered as a mediating variable to account for bias in richness estimates due to  
263 variation in abundance among plots (Fig. S2).

264 To determine the most parsimonious SEM model, with the minimum number of  
265 pathways necessary to explain variation in the three response variables, all possible  
266 combinations of direct and indirect pathways between the experimental treatments and  
267 response variables were tested using the specification search option in Amos. The model with  
268 the lowest Akaike Information Criteria (AIC) was chosen as the best fit. We used  
269 bootstrapping with 1,000 random samples generated from the observed covariance matrix to  
270 estimate 90th percentile confidence intervals and significance values for the standardised  
271 direct, indirect and total effects in the final model (Appendix S1). Model fit to the data was  
272 considered good if standardised residual covariance values between variables were below 2,

273 and the root-mean-square error of approximation (RMSEA) values were below 0.05  
274 (Appendix S1). Models were assessed for multivariate normality and when the critical ratio  
275 for kurtosis exceeded 2.0 we also validated model fit by obtaining Bollen-Stine bootstrap *P*-  
276 values (Appendix S1).

277

## 278 **Results**

### 279 *Variation in land snail communities within and between pre- and post- treatment samples*

280 The pre-treatment leaf-litter samples contained a total of 1,660 land snails, comprising 91  
281 species, with abundances ranging from 0 to 56 individuals per sample (mean  $\pm$  95% CL  
282 densities of  $134.77 \pm 21.70$  individuals per  $m^2$ ) and species richness per sample ranging from  
283 0 to 29 species (mean  $\pm$  95% CL of  $7.79 \pm 1.04$  species) (Table S3). Post-treatment samples  
284 contained a total of 2,105 individuals, comprising 78 species, with abundances ranging from  
285 0 to 59 individuals per sample (mean  $\pm$  95% CL densities of  $169.98 \pm 24.46$  individuals per  
286  $m^2$ ) and species richness per sample ranging from 0 to 34 species (mean  $\pm$  95% CL,  $10.57 \pm$   
287  $1.30$  species) (Table S3).

288 Species accumulation curves calculated for pre-treatment and post-treatment samples  
289 approached an asymptote and no additional land snail species were captured in the post-  
290 treatment sampling that were not captured in the pre-treatment sampling. Together, this  
291 suggests that sampling was representative of the community at the experimental site (Fig.  
292 S3). Despite a 27 % increase of land snail individuals in the post-treatment samples, there  
293 was a 16% decrease in rarefied species richness in post-treatment samples (at a standardised  
294 sample abundance of  $n = 1,660$  individuals) (Fig. S3).

295 In an NMDS ordination analysis of variation in land snail community composition  
296 among experimental plots, there was significant variation in species' relative abundances  
297 between pre-treatment and post-treatment plots (ANOSIM,  $R = 0.111$ ,  $P = 0.001$ , Fig.2).

298 Moreover, variation among plots was also higher in pre-treatment samples than post-  
299 treatment samples (PERMDISP, t-statistic=5.13,  $P = 0.001$ ), suggesting that communities  
300 became more similar post-treatment (Fig. 2).

301

### 302 *The direct and indirect effects of trampling and litter treatments on land snail communities*

#### 303 *Land snail density*

304 Treatment contrasts (Fig.3a) showed that all levels of trampling intensity and litter  
305 manipulation had negative effects on land snail density, compared with control plots, with an  
306 average reduction in density of 72 individuals per  $m^2$  across all treated plots (Fig. 3a). The  
307 magnitude of these effects increased monotonically with increasing trampling intensity in the  
308 trampling-only treatments (S and SL) (Fig. 3a). However, trampling intensity effects were not  
309 evident in the trampling combined with litter removal treatments (SLR) because all plots  
310 showed very high reductions in land snail density (declines of up to 100 individuals per  $m^2$ ,  
311 on average) (Fig. 3a).

312 The most parsimonious SEM model for variation in land snail density explained 72 %  
313 of variance in the data ( $R^2 = 0.72$ ) and had acceptable goodness of fit indices, with all residual  
314 covariance values  $< 2$ , a non-significant Bollen-Stine value ( $P = 0.944$ , Table S4), and  
315 RMSEA value below 0.001 (Fig. 4a). Both the trampling intensity and litter manipulation  
316 treatments, as well as their interaction, caused significant changes to land snail density  
317 (Fig.4a). Intensity of trampling had a significant negative (direct) effect on land snail density,  
318 but this effect could not be attributed to any indirect (mediating) effects of altered soil  
319 penetration resistance or litter structure (Fig.4a). The trampling intensity effect was  
320 significantly diminished by its interaction with litter removal (i.e. the positive trampling by  
321 litter interaction effect indicates that the negative effect of trampling on land snail density  
322 diminishes when half the litter was removed before the first trampling event (SLR); Fig. 3a).

323 In contrast, the litter manipulation treatment operated through a mediating effect of decreased  
324 litter mass, rather than substantial alteration of any other measured litter variables, and  
325 contributed strongly to decreases in land snail density (Fig. 3a, 4a, S4). Litter mass was the  
326 most important explanatory variable for changes in land snail density (standardised total  
327 effect = 0.680, Table S5). In addition to litter mass, several other endogenous predictors had  
328 significant positive effects on snail density independent of the experimental treatment effects:  
329 soil penetration resistance, litter and soil moisture contents (Fig. 4a).

330

### 331 *Land snail species richness*

332 Similar to land snail density, relative species richness declined at all levels of trampling  
333 intensity and litter manipulation (Fig. 3b). The average decrease in land snail species richness  
334 across all treatment plots was 10 species and up to an average of 23 species in the plots where  
335 changes were most severe (Fig. 3b). Within the soil-only trampling (S) and the trampling  
336 with litter removal (SLR) treatments, land snail species richness generally decreased with  
337 increasing trampling intensity. However, within the soil-and-litter (SL) trampling treatment  
338 the results were more variable.

339 The final SEM model for land snail species richness had acceptable goodness of fit  
340 indices (as above) ( $P = 0.968$ , Table S4). The model explained substantially less variation in  
341 land snail species richness ( $R^2 = 0.35$ ) than was observed for land snail density, and most of  
342 the important treatment effects in the SEM analysis were mediated by land snail density (Fig.  
343 3b, Table S5). Land snail species richness decreased with increasing trampling intensity  
344 (mediated by declining land snail density) and decreased with increasingly severe treatment  
345 alteration of litter structure (mediated by the effect of declining litter mass on land snail  
346 density) (Fig. 3b, 4b). There were also weak (non-significant) indirect effects of the trampling

347 by litter interaction term (mediated by litter complexity), and weak direct negative effects of  
348 soil moisture on land snail species richness (Fig.4b).

349

#### 350 *Land snail community composition*

351 All treatment combinations substantially altered land snail community composition, as  
352 measured by dissimilarity relative to the control plots (Fig. 3c). These effects on community  
353 composition were remarkably consistent across all levels of trampling and leaf-litter  
354 manipulation, with an average post-treatment increase in snail community dissimilarity  
355 relative to control plots of 18.6 % (Fig. 3c).

356 The final SEM model for land snail community dissimilarity explained 32 % of  
357 variance ( $R^2 = 0.32$ ) and had acceptable goodness of fit indices ( $P = 0.959$ , Table S4).

358 Increased trampling had significant direct positive effects on community dissimilarity to  
359 control sites, but this result could not be attributed to any specific mediating processes (Fig.  
360 3c, 4c). The dominant factors explaining variance in land snail community dissimilarity were  
361 increasing penetration resistance and increasing litter moisture content, independent of the  
362 effects of the experimental manipulations (Fig. 4c, S4).

363

## 364 **Discussion**

365 In many parts of the world, there is extensive evidence that unfenced forest remnants  
366 embedded within farmland can be severely damaged by livestock trampling (Close, Davidson  
367 & Watson 2008; Hayward & Kerley 2009; Burns *et al.* 2011). However, the relationship  
368 between the intensity of livestock trampling and the magnitude of ecological effect has been  
369 poorly quantified. Here, we show experimentally that even low-intensity trampling is  
370 sufficient to cause severe effects on native land snail communities within a forest remnant  
371 with 20 years of livestock exclusion. Our lowest-intensity trampling treatment was sufficient

372 to cause a large reduction in the abundance and richness of land snails compared with control  
373 plots, even though this was only a minimal ‘pulse’ disturbance over a six-week time period.  
374 These effects increased dramatically with trampling intensity, even though they still  
375 underestimate the real-world ‘press’ disturbance effects of repeated livestock access over  
376 months or years in field situations. We discuss the implications of these findings for the  
377 conservation management of native forest remnants embedded within production landscapes.

378

### 379 *Effects of livestock on land snail density*

380 Experimental manipulation of trampling intensity had a highly significant negative effect on  
381 land snail density, and was one of the main contributors to the very high overall explanatory  
382 power (72 % variance explained) in the SEM model for land snail density. On average, land  
383 snail density in the lowest intensity treatment plots decreased by 42 individuals m<sup>-2</sup> compared  
384 with the control, and up to 100 individuals m<sup>-2</sup> on average across higher-intensity treatments.  
385 The trampling effect (and the interaction effect between trampling and litter manipulation  
386 treatments) operated entirely through direct effects on land snail density, rather than through  
387 any indirect mediating influences of the measured proximate variables. The most  
388 parsimonious explanation for this result is a direct physical effect of trampling on the  
389 mortality of land snails due to crushing. Typically, the dominant environmental variables  
390 influencing land snail communities are one or more of three key variables reflecting soil  
391 moisture levels and leaf-litter structure (both of which we measured here), as well as soil pH  
392 (or calcium levels) which we did not measure (Martin & Sommer 2004; Barker 2005; Müller,  
393 Strätz & Hothorn 2005; Ruckli, Rusterholz & Baur 2013).

394 For the first of these three potential determinants of land snail distribution, we found  
395 no effects of trampling on either soil or litter moisture in the experiment. This lack of effect is  
396 perhaps somewhat surprising, considering there is substantial evidence showing that grazing



397 intensity influences soil compaction and moisture levels (e.g. Bromham *et al.* 1999; Chaichi,  
398 Saravi & Arash 2005). However, this discrepancy may be due to the short-term nature of our  
399 experiment and the relatively low-intensity treatments we applied compared with other  
400 studies. Here, we found no significant link between trampling treatments and soil compaction  
401 (penetration resistance). In the longer term, repeated trampling treatments might be expected  
402 to have a greater influence on soil and litter moisture, through increased soil compaction (e.g.  
403 Chaichi, Saravi & Arash 2005; Savadogo, Sawadogo & Tiveau 2007). Despite the lack of a  
404 trampling effect on soil or litter moisture, each variable significantly influenced land snail  
405 density in its own right. This result supports other studies worldwide suggesting moisture  
406 levels as one of the key drivers of land snail community structure (Martin & Sommer 2004;  
407 Čejka & Hamerlík 2009), particularly in NZ where a reliable moisture regime is one of key  
408 factors postulated as favouring high land snail density in native forests (Solem 1984). The  
409 strong link we observed between variation in moisture content and land snail density suggests  
410 that any long-term effects of livestock trampling on moisture levels would only exacerbate  
411 the already severe effects of trampling measured in this experiment.

412         Second, we found that the trampling treatments had a weak (marginally non-  
413 significant) effect on litter complexity, but over the short time-course of the experiment these  
414 effects did not mediate changes in land snail density. Other aspects of habitat structure that  
415 were not measured and could mediate trampling effects on land snail density include root  
416 mass and amount of dead wood in the plots. Amount of dead wood in particular can be  
417 important for land snail communities (Müller, Strätz & Hothorn 2005; Boschi 2007),  
418 although it is unlikely to be important here due to the minimal amount of dead wood present  
419 at the study site. Third, we did not measure variation in soil pH or calcium levels which could  
420 have mediated some of the direct trampling effects that we observed (Martin & Sommer

421 2004; Jeddi & Chaieb 2010). Overall, the most parsimonious explanation for short-term  
422 declines in land snail density due to trampling is through direct mortality effects.

423 In contrast to the trampling treatments, we found that the effects of our litter  
424 manipulation treatments were entirely mediated by the measured proximate variables, and  
425 there were no residual (unexplained) direct influences of litter manipulation on land snail  
426 density. Litter mass reduction had a very large negative effect on land snail density, and was  
427 the single most important explanatory variable in the SEM model for land snail density. The  
428 dominant influence of litter mass on land snail density makes it all the more surprising that so  
429 few studies investigating land snail community dynamics consider the potential causal  
430 influence of variation in litter mass in their analyses (but see Barker & Mayhill 1999; Barker  
431 2005).

432

### 433 *Effects of livestock on land snail species richness and community composition*

434 Land snail species richness and community composition were negatively affected by all  
435 treatment combinations of trampling intensity and litter manipulation. Overall, species  
436 richness per plot declined dramatically (average of 10 species per plot) and community  
437 dissimilarity relative to control plots was consistently higher (from 12 to 32 %) across all  
438 levels of trampling and litter manipulation. These severe and consistent negative effects on  
439 species richness and community composition suggest that NZ land snails are highly sensitive  
440 to disturbance resulting from the presence of livestock.

441 In the species richness analysis, the strong treatment effects observed for land snail  
442 density also had a significant cascading effect on species richness (through the mediating  
443 effect of density on richness), with few additional direct or indirect influences on species  
444 richness itself. However, there was still a very weak direct negative effect of soil moisture on  
445 species richness. This is surprising considering there was a positive direct effect of soil

446 moisture on snail density, and that previous studies have linked soil moisture positively with  
447 land snail abundance and richness (Martin & Sommer 2004; Čejka & Hamerlík 2009). Also  
448 independent of the significant indirect land snail density effect was a weak positive effect of  
449 trampling on land snail species richness mediated by litter complexity. This result is  
450 supported by numerous studies linking habitat complexity with species richness, for land  
451 snails and other taxa (e.g. Solem 1984; Boschi 2007). Diverse habitats offer a variety niches,  
452 more physical refugia and increased ways of exploiting available resources (Boschi 2007).

453 In comparison, trampling intensity increased land snail community dissimilarity to  
454 control plots through direct rather than indirect effects, and none of the observed effects on  
455 community dissimilarity were mediated by changes in litter mass. In addition, both  
456 penetration resistance and litter moisture increased dissimilarity to the controls, though  
457 neither was linked to the trampling or litter treatments. The strong link between penetration  
458 resistance and community composition, without a significant effect of trampling treatment on  
459 penetration resistance, implies that higher intensity, longer term trampling that would cause  
460 changes in soil compaction, could have large effects on land snail community composition.

461 The results from all of the SEMs taken together suggest that the alteration of litter  
462 mass and complexity were the most important factors mediating changes in land snail species  
463 richness and density. Conversely, factors associated more directly with trampling such as  
464 snail mortality, penetration resistance and litter moisture content were more influential in  
465 changing patterns of land-snail community composition.

466

#### 467 *Conservation management implications*

468 The number of landowners fencing native forest remnants in NZ is continuing to rise  
469 (Anonymous 2013b), as farmers become more aware of the conservation significance of  
470 remnant vegetation and the important role that livestock exclusion can play in habitat

471 restoration (Didham *et al.* 2009). Nevertheless, many farmers (as many as 77% of farmers in  
472 the region where this study was conducted) value their forest remnants for livestock shelter as  
473 well as for native species conservation (Jay 2005). For example, farmers that have fenced  
474 their forest remnants for biodiversity conservation may allow livestock into the remnants  
475 once or twice a year during bad weather or during lambing and calving (LHD, pers. obs.).  
476 Conflicting trade-offs between production and conservation goals are common in production  
477 landscapes and will become more difficult to avoid if we are to ensure food security into the  
478 future (Cunningham *et al.* 2013). How conflicts are managed is often context dependent. For  
479 example, in landscapes with a long history of land-use such as the calcareous grasslands of  
480 Europe, light to medium intensity grazing actually promotes species richness (Boschi 2007).  
481 Nevertheless, even in these systems, overstocking can lead to land degradation and declines  
482 in native biodiversity (Boschi 2007). In landscapes with relatively short land-use histories,  
483 however, livestock trampling is more likely to have a negative effect, and the results of our  
484 trampling experiment calls into question the sustainability of even minimal levels of livestock  
485 access to forest habitats. In NZ in particular, the absence of ungulate trampling in the  
486 evolutionary history of ecosystems may have made invertebrate communities more  
487 vulnerable. Our results support increased action to maintain and improve livestock exclusion  
488 as a priority conservation management action for native forest remnants embedded in  
489 production landscapes.

490

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498

#### 499 **Data Accessibility**

500 -All sample details: DRYAD entry doi:10.5061/dryad.60g61

501

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603

604 **Supporting Information**

605 Additional Supporting Information may be found in the online version of this article:

606

607 **Appendix S1.** Supplementary methods

608

609 **Fig. S1.** Experimental plot layout

610

611 **Fig. S2.** Full SEM models tested in Amos v.19

612

613 **Fig. S3.** Sample based species accumulation curves

614

615 **Fig. S4.** Average effects of experimental litter manipulation and trampling treatments on  
616 habitat structure and environmental variables

617

618 **Table S1.** Typical stocking rates for farms in the Waikato region, New Zealand

619

620 **Table S2.** Standardised measures of stocking units for different livestock classes on farms

621

622 **Table S3.** List of the snail species identified

623

624 **Table S4.** Results from structural equation model multivariate normality tests

625

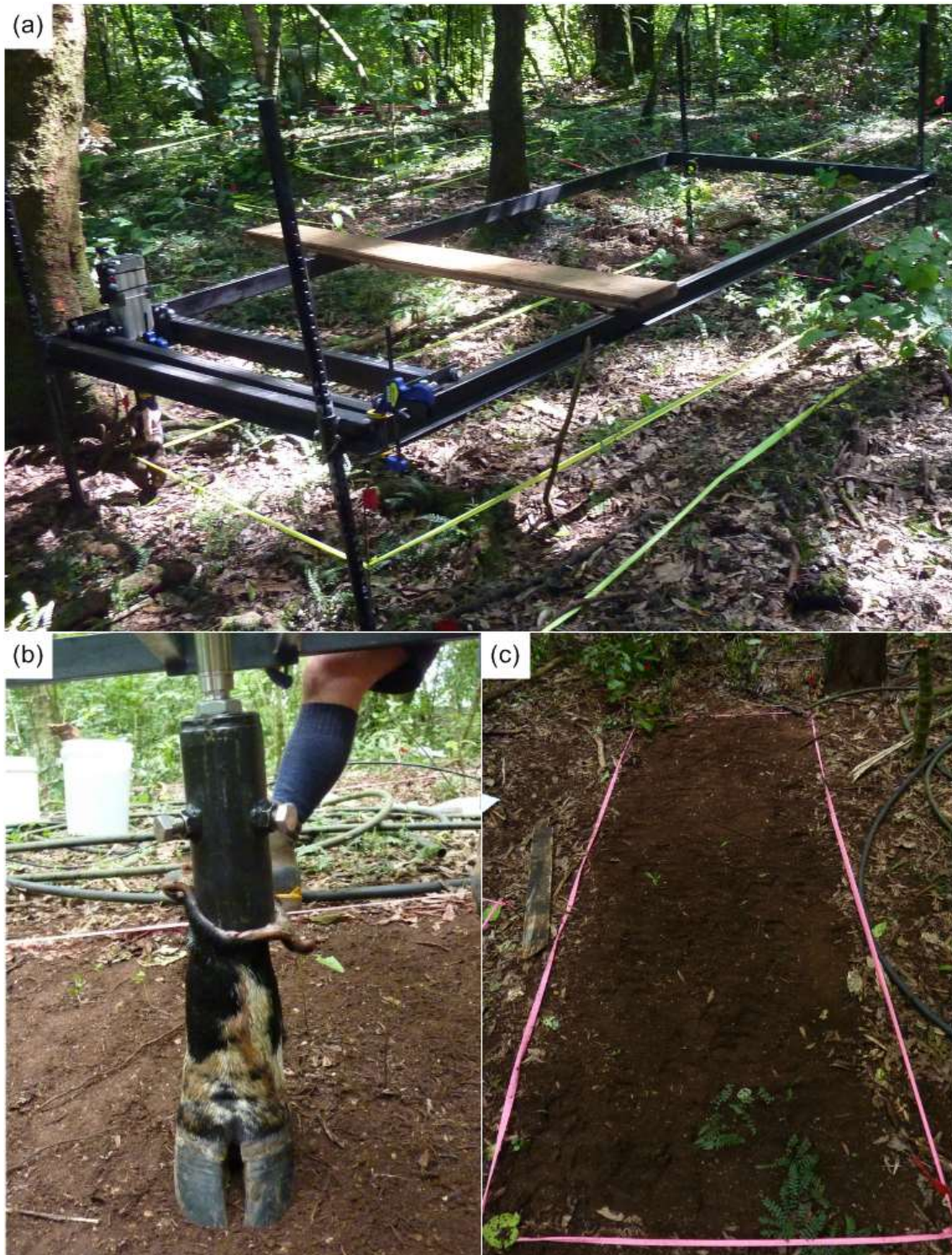
626 **Table S5.** Standardized path coefficients for final structural equation models

627

628

629



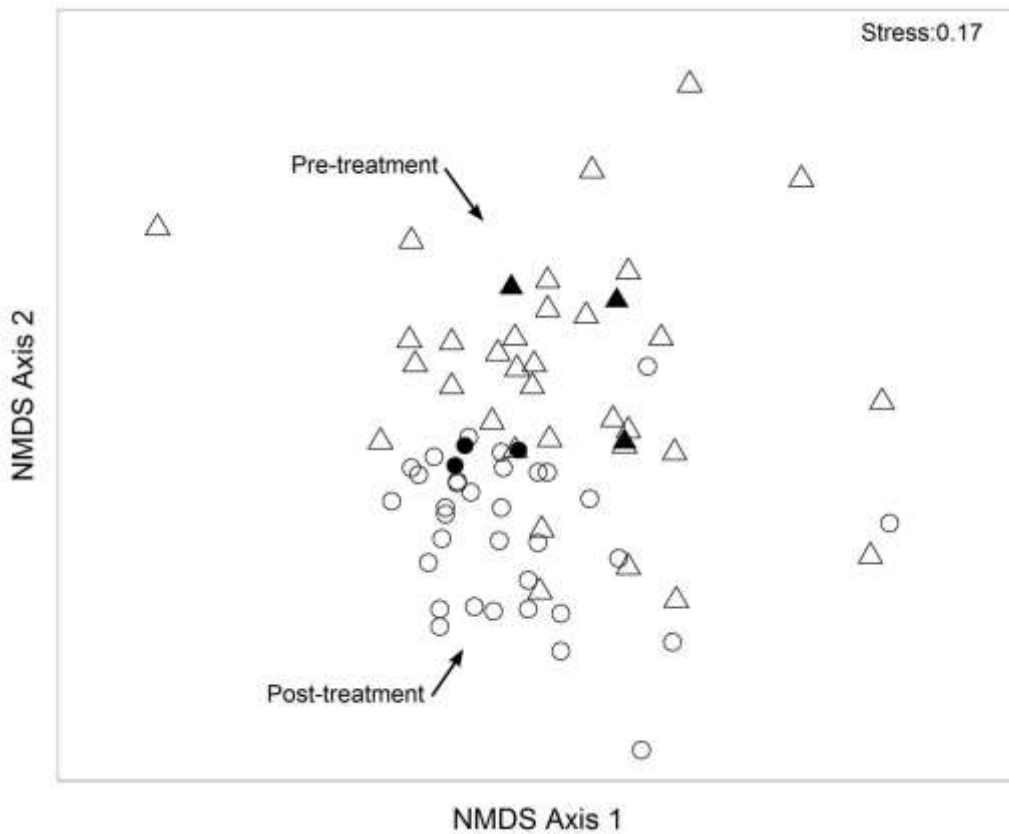
630 **Figures**

631

632 **Figure 1.**Experimental hoof design, showing (a) mechanical frame set up for each trampling

633 event, (b) cow hoof attached to the air ram, and (c) a plot after a trampling event (S

634 treatment), showing impressions made by the mechanical hoof.



635

636 **Figure 2.** Variation in land snail community composition within and between pre-treatment

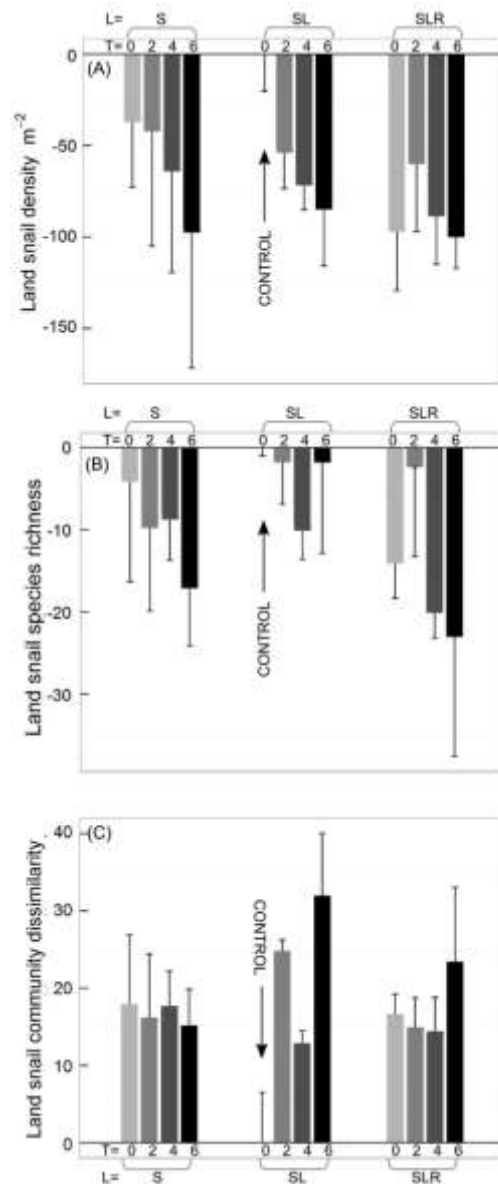
637 and post-treatment samples in a NMDS ordination (based on square-root transformed land

638 snail abundance data and a Bray-Curtis dissimilarity). Pre-treatment plots are denoted by

639 triangles and post-treatment plots by circles. Control plots are indicated by filled symbols.

640 The stress value is for the 3D solution of this 2D NMDS.

641



642

643 **Figure 3.** Average effects of experimental litter manipulation (L) and trampling intensity (T)644 on land snail community responses: (a) land snail density  $m^{-2}$ , (b) species richness per plot,

645 (c) community dissimilarity relative to control plots. Response variables were standardised

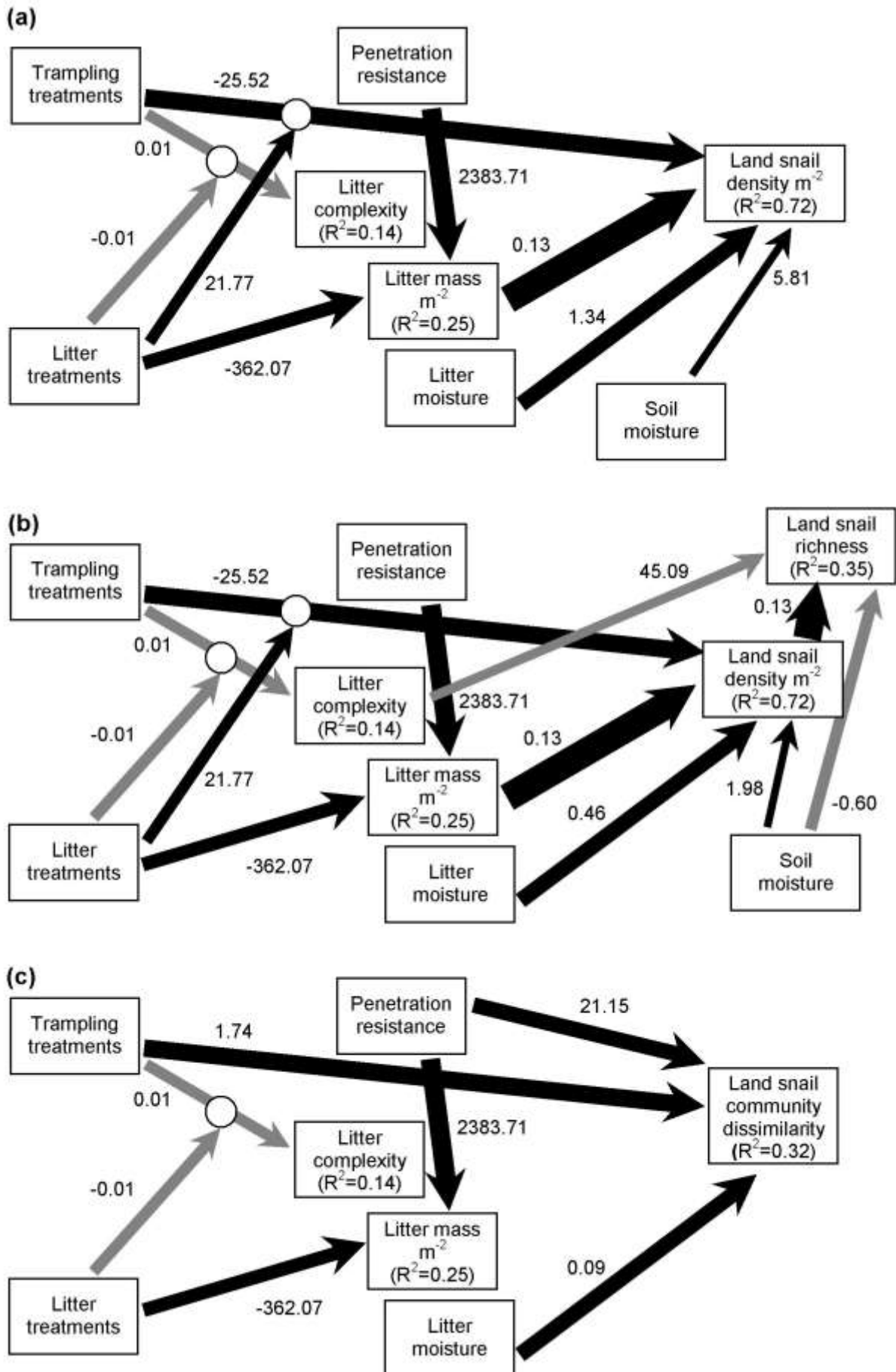
646 using post-treatment minus pre-treatment values and expressed as treatment minus control

647 differentials (control=SL0). Responses are grouped by litter treatment: S=only soil trampled,

648 SL=soil and litter trampled, and SLR=soil and litter trampled after removing half of the litter

649 mass. Within litter treatments, bars indicate response to number of trampling events (T) over

650 a six-week period.



652 **Figure 4.**Structural equation models discriminating direct versus indirect pathways through  
653 which trampling and litter manipulation influence snail communities. Path diagrams show the  
654 most parsimonious models for **(a)** land snail density m<sup>-2</sup>, **(b)** land snail species richness, and  
655 **(c)** land snail community composition. Arrows represent causal paths from predictor to  
656 response variables. A circle where two paths intersect indicates the interaction effect between  
657 the two predictors. The number on each path is the unstandardised partial regression  
658 coefficient, indicating whether the relationship is positive or negative. Statistical significance  
659 of coefficients is indicated by line colour (black,  $P \leq 0.05$ ; grey,  $> 0.05$ ). Line thickness  
660 indicates the magnitude of standardised path coefficients (Table S5).  $R^2$  represents the  
661 variance explained for endogenous variables.