Title: **Tracking sucking herbivory with nitrogen isotope labelling: Lessons from an individual trait-based approach**

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1 **Highlights**

2 - In mesocosms, sucking herbivory on grasses was tracked with ^{15}N isotope labelling
3 - Process rates (herbivory, plant growth) were strongly linked to insect identity
4 - Also, process rates depended on the interacting plant and insect species
5 - This indicates trait matching, but common plant and insect traits explained little

6

7 **Abstract**

8 Response and effect traits help to understand how changes in ecological communities (e.g. in

9 response to land use) relate to changes in ecosystem functioning. In grasslands, plants and

10 insect herbivores are involved in many ecosystem processes such as herbivory and plant

11 biomass production. Simultaneous changes in the trait composition of both plants and

12 herbivores should affect herbivory rates, with consequences for plant growth and potentially

13 biomass production. The mechanisms underlying these links are little understood for grasses

14 and sucking insects, which build a major part of grassland communities. In a mesocosm

15 experiment, we manipulated the composition of grasses and sucking herbivores (Hemiptera)

16 to study the role of plant traits, herbivore traits and their interaction on herbivory and plant

17 growth. Because sucking herbivory is generally difficult to quantify, we developed a novel

18 experimental setting, in which we labelled plants with ^{15}N isotope. This allowed to quantify

19 ^{15}N uptake and thus sucking rates of individuals. We found that herbivory and simultaneous

20 plant growth reduction are most strongly linked to herbivore species identity. Unexpectedly,

21 herbivory did not increase with herbivore size, but was highest for small species and for thin-

22 bodied Heteroptera. Additionally, herbivory and plant growth reduction depended on the

23 interacting herbivore and plant species, indicating trait matching, which could, however, not

24 be explained with commonly used traits. This indicates that mechanisms linking ecological

25 communities and ecosystem processes are highly context-specific. To understand how global

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26   change affects ecosystem functioning, studies need to cover all functionally relevant groups,

27   including plant sap suckers.

28 **Keywords:**

Introduction

31 Global change drivers such as land-use change and intensification shift the composition of

32 ecological communities across ecosystems and trophic levels, with consequences for

33 ecosystem functioning (Allan et al., 2015). In semi-natural grasslands, which are important

34 hotspots of biodiversity in temperate regions (Habel et al., 2013), intensive agricultural use

35 has been shown to be a major filter in the assembly of both plant and insect communities

36 (Neff et al., 2019; Socher et al., 2012). At the same time, the rates of different ecosystem

37 processes have been found to be strongly affected by land-use intensity (Ambarlı et al., 2021)

38 and these changes have been related to shifts in diversity or composition of ecological

39 communities (Wang et al., 2020). The underlying mechanisms by which changes in

40 community composition affect ecosystem processes are, however, understudied.

41 An important ecosystem process in semi-natural grasslands is insect herbivory, which might

42 be strongly linked to plant growth and thus affects plant biomass production. Plant biomass

43 production in these systems is an important provisioning service contributing to agricultural

44 production (Bengtsson et al., 2019). Insect herbivory might either reduce plant biomass

45 production through reduced plant growth (Crawley, 1989) or stimulate plant growth (Dungan

46 et al., 2007). Plant biomass production and insect herbivory are tightly linked to plant and

47 insect communities (Lavorel et al., 2013), but how changes in these multi-trophic

48 communities affect ecosystem processes is still poorly understood.

49 The use of effect traits, i.e. species or individual morphological or physiological

50 characteristics that affect ecosystem processes, can improve the understanding of the

51 mechanisms linking ecological communities and ecosystem processes (Lavorel & Garnier,

52 2002). For example, plants characterised by high specific leaf area (SLA) and leaf nitrogen

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53   content (LNC) tend to be associated with faster plant growth and contribute to higher plant

54   biomass production (Funk et al., 2017; Wright et al., 2004). At the same time, plant biomass

55   production was found to be more strongly reduced by larger grasshoppers (Moretti et al.,

56   2013) with stronger mandibles (Deraison et al., 2015). Additionally, traits of organisms

57   belonging to different trophic levels might have interactive effects on ecosystem processes

58   through trait matching (Schleuning et al., 2015). For example, plant biomass consumption

59   depends on the interaction between plant toughness and the grasshopper's mandible strength

60   (Ibanez, Lavorel, et al., 2013). Thus, we need to better understand how traits of organisms at

61   different trophic levels jointly affect ecosystem processes to predict how shifts in

62   communities affect ecosystem functioning.

63   Such questions have rarely been studied at the level of single species or functional groups

64   (but see Ibanez, Bison, et al., 2013). Furthermore, studies addressing similar questions so far

65   never addressed herbivores that feed by sucking plant saps, which are, however, accounting

66   for a large share of herbivore communities in grasslands (e.g. Risch et al., 2015) and can

67   significantly reduce plant growth (e.g. Meyer & Whitlow, 1992). This is not least because

68   sucking herbivory rates are genuinely hard to quantify, given that feeding marks are hard to

69   see and may not well be related to uptake rates (Schowalter, 2011). However, sucking

70   herbivore communities are substantially affected by intensive land use, which changes their

71   trait composition, e.g. by filtering for smaller species (Neff et al., 2019). How these changes

72   in trait composition affect insect herbivory and relate to plant growth, and consequently plant

73   biomass production are still open questions.

74   Here, we manipulated the trait composition of plants and herbivores in a fully crossed

75   mesocosm experiment to study how traits are related to insect herbivory and plant growth and

76   whether there is indication for trait matching between the two trophic levels. We focused on

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77 hemipteran species sucking on grasses, both of which are important functional groups in

78 semi-natural grasslands (Neff et al., 2021). To overcome the difficulty of assessing sucking

79 herbivory, we developed a novel experimental setting, where plants were labelled with a

80 heavier isotope of nitrogen (^{15}N), which enabled us to track the flow of nitrogen in the system

81 (e.g. Steffan et al., 2001). Stable isotope techniques are increasingly used in insect ecology

82 (e.g. Quinby et al., 2020), and also to study nutrient flows in food webs or to assess herbivory

83 (e.g. Schallhart et al., 2012; Porras et al., 2020). Here, labelling of plants with ^{15}N allowed us

84 to quantify herbivory rates of single sucking herbivores, which has to our knowledge not

85 been done before, but provides large potential for more mechanistic studies on insect

86 herbivory. The grass species included in the experiment were chosen to cover a gradient in

87 palatability inferred from three traits (leaf dry matter content (LDMC), SLA, LNC), which

88 have commonly been used to relate plant palatability to chewing herbivory (e.g. Schädler et

89 al., 2003). Herbivore species were chosen to cover a trait space defined by three potential

90 effect traits (body volume, body shape, rostrum length). We were interested in the interplay

91 of these traits in determining insect herbivory and changes in plant growth.

92 We predicted that herbivory rates would be highest on plant species characterised by high

93 palatability and for the largest herbivore species, resulting in reduced plant growth, unless

94 there is a stimulation of compensatory plant growth by herbivory. Additionally, if there is

95 trait matching evident for the trophic relations between these two groups, we predicted that

96 highest herbivory and consequently highest plant growth reduction should be observed at

97 certain combinations of plant and herbivore traits. For example, we expect that plants with

98 thicker leaves (i.e. low SLA; Wilson et al., 1999) are better accessible to herbivores with

99 longer rostra and thus deeper leaf penetration potential, inducing trait matching.

Materials and methods

Plant and herbivore material

Plant species were restricted to Poaceae and were selected based on three traits, which are

essential determinants of the global leaf economic spectrum (Wright et al., 2004) and are

related to palatability and herbivory rates (e.g. Schädler et al., 2003): LDMC, SLA and LNC.

Three species were selected from each of three clusters of species sharing similar traits (Fig.

1A): *Agrostis capillaris*, *Arrenatherum elatius* and *Poa trivialis* in the high palatability

cluster (low LDMC, high SLA and high LNC); *Cynosurus cristatus*, *Festuca arundinacea*

and *Holcus lanatus* in the medium palatability cluster (with low LDMC, high SLA and low

LNC); and *Deschampsia cespitosa*, *Festuca ovina* agg. and *Sesleria caerulea* in the low

palatability cluster (high LDMC, low SLA and low LNC) (Appendix A for details).

Insect herbivore species were selected from grass feeding Hemiptera (suborders

Auchenorrhyncha and Heteroptera) based on three morphometric traits related to sucking

herbivore effects, i.e. body volume, rostrum length and body shape (see Appendix A for

inclusion rationales). Species were selected to cover the trait space (Fig. 2A) and included

Aelia acuminata, *Lygus* spp., *Notostira* spp., *Trigonotylus caelestialium*, *Stenodema*

laevigata, *Laodelphax striatella*, and *Deltocephalus pulicaris* (Appendix A for details).

Experimental design and setup

The mesocosm experiment was performed in experimental cages in August/ September 2019

with a completely randomized design with two crossed treatment factors (Appendix B: Figs

1–3): plant palatability (three factor levels) and herbivore species identity (seven species and

one control treatment without herbivores). Each treatment combination was replicated five

times (3 plant treatments $\times$ 7 herbivore treatments $\times$ 5 replicates = 105 cages), except for the

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123   control treatments, to which some additional cages originally planned to contain further

124   herbivore species were added, resulting in up to eight replicates (3 plant treatments $\times$ 1

125   herbivore control $\times$ 7–8 replicates = 23 cages). Because some cages were built with a

126   plexiglass that was unexpectedly preventing plant growth, these cages were excluded from

127   analyses. These cages had been randomly assigned to the study treatments and together with

128   other, minor incidents, we ended up with two to five (seven for some controls) replicates per

129   treatment combination and a total of 94 experimental cages (see Appendix B: Table 1 for a

130   complete overview of replicates), which still enabled robust analyses given the fully crossed

131   experimental design.

132   At the start of the experiment, each cage contained an individual of each of the three plant

133   species selected for the palatability cluster, which were labelled with ^{15}N to track the flow of

134   nitrogen in the system, and two individuals of a herbivore species (Appendix B: Fig. 1).

135   Different measures were taken on plants and herbivores at the start and the end of the

136   experiment (Appendix B: Table 2), which were used to quantify traits, plant growth and

137   herbivory rates. For details on the experimental setup, see Appendix A.

138   *Estimation of ecosystem processes*

139   For each plant individual, we predicted dry mass at the end of the experiment ($m_{end,pred}$) that

140   would have been expected in the absence of herbivores from estimated dry mass at the

141   beginning of the experiment and growth observed for control plants not affected by

142   herbivores (Appendix A). Predicted dry mass was related to measured dry mass at the end of

143   the experiment (m_{end}) to determine relative deviation from expected growth g_{off} as

144    $$g_{off} = \frac{m_{end,pred} - m_{end}}{m_{end}}$$ (1)

145   with positive values representing lower than expected growth and negative values

146   representing higher than expected growth. These values were used as proxies of plant growth

147   reduction.

148   Insect herbivory was estimated based on uptake of ^{15}N by herbivores (u_{abs}) and mean ^{15}N

149   concentration of the available plants ($\overline{c_{15_N}}$), which were determined from $\delta^{15}N$ ratios,

150   nitrogen content and biomass of plant and herbivore samples (Appendix A). ^{15}N uptake by

151   herbivores relative to available ^{15}N in plants (u_{rel}) was determined as

152                              $$u_{rel} = \frac{u_{abs}}{c_{15_N}}$$ (2)

153   and was used as a proxy of insect herbivory.

154   *Statistical analyses*

155   All analyses were conducted in R v3.5.2 (R Core Team, 2018). Linear mixed effects models

156   were used to relate herbivory and plant growth reduction to *(i)* herbivore species and plant

157   palatability cluster identities and *(ii)* herbivore traits and plant traits. The identity models

158   contained herbivore species identity, plant palatability cluster identity, their interaction as

159   well as the potentially confounding variables herbivore survival and distance to light (integer

160   denoting the row at which the cage was positioned; Appendix B: Fig. 4) as fixed effects and a

161   random effect for the cage. Herbivory was analysed at the level of individual herbivores, with

162   survival indicating whether the individual was found alive at the end of the experiment (0/1),

163   whereas plant growth reduction was analysed at the level of the individual plants, with

164   survival indicating the number of individual herbivores that were found alive at the end of the

165   experiment (0–2). Herbivory was log-transformed prior to analyses to meet distributional

166   assumptions. The trait models had the same structure as the identity models, but herbivore

167   species and plant palatability cluster identities were replaced with herbivore and plant PC

168   axes. Based on the principal component analyses that were used for the selection of herbivore

169   and plant species, study specimens were placed on the same PC axes based on their measured

170   trait values. PERMANOVA from the package 'vegan' (Oksanen et al., 2018) was used to

171   check whether plant palatability clusters for the study plants were also represented by their

172   PC axis values based on measured traits (9999 permutations). The two PC axes per trophic

173   level were then included in the models. Additionally, all possible interactions between

174   herbivore PC axes and plant PC axes ($n = 4$) were included in the model. Backward model

175   selection (based on χ^2 tests) was used to find the optimal interaction structure for each model.

176   Only interactions but no main effects were excluded during model selection. As for the

177   identity models, herbivory was analysed at the level of individual herbivores, whereas plant

178   growth reduction was analysed at the level of individual plants. Trait values of the respective

179   other level were aggregated at cage level by taking mean values. The effect of sex on

180   herbivory was tested in both the identity and trait models, but was not found to be significant,

181   which is why it was excluded from the final models. All linear mixed effects models were run

182   through the package 'glmmTMB' (Magnusson et al., 2020).

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Results

184 Plant palatability clusters were represented by traits measured for the study plants

185 (PERMANOVA: $P < 0.001$ for all pairwise comparisons based on PC axes), although

186 variation within the clusters was quite large (Fig. 1B). On average, the study plants had

187 higher LNC than plants in the data base, indicating a fertilisation effect caused by the ^{15}N

188 labelling (Appendix B: Fig. 5). All study plants were strongly enriched in ^{15}N compared to

189 plants of the same species that were not included in the experiment (Appendix B: Fig. 5). Dry

190 mass of control plants without herbivores present increased by 230% ± 16% (mean ± SE)

191 relative to predicted dry mass at the start of the experiment, while dry mass of plants with

192 herbivores present increased by 160% ± 8%, which was significantly less than for control

193 plants (LMM: $\chi^2 = 14.36$, $P = 1.5e\text{-}04$; Appendix B: Fig. 6).

194 The traits measured on the study herbivore specimens matched closely the expected trait

195 ranges (Fig. 2B). Mortality among retrieved study specimens was 52.7% ($n = 68$).

196 Additionally, 14.0% ($n = 21$) of individuals could not be retrieved at the end of the

197 experiment and were thus recorded as dead, resulting in an overall survival rate of 40.7%

198 ($n = 61$), which differed greatly among study species (Appendix B: Table 3). All specimens,

199 including the ones that had died, had clearly elevated ^{15}N concentrations, indicating

200 (premortem) feeding activity of all specimens (Appendix B: Fig. 7). Average absolute ^{15}N

201 uptake by herbivores was estimated to 0.590μg (0.006 – 2.939μg [5% and 95% quantiles]),

202 which relative to plant content of ^{15}N corresponds to 97.7μg (1.09 – 476.8μg) of dry plant

203 material that was taken up (Appendix B: Fig. 8). Average dry mass of herbivores was 3.73mg

204 (0.214 – 16.44mg; Appendix B: Fig. 9). Differences in relative ^{15}N uptake between herbivore

205 species were ranging from 27.1μg (1.68 – 103.2μg) of dry plant material for *Stenodema*

206 *laevigata* to 188.9μg (0.294 – 518.0μg) for *Trigonotylus caelestialium* (Appendix B: Fig. 8).

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207 Signs of herbivory on the plants were recorded on 25 plants (11.3%), 12 of which were on

208 plants that were with *T. caelestialium*.

209 *Effect of plant and herbivore species on herbivory and plant growth reduction*

210 Herbivory (^{15}N uptake of herbivores relative to average plant ^{15}N content) was strongly

211 affected by the interaction of plant palatability cluster identity and herbivore identity (LMM:

212 $\chi^2 = 41.40$, $P = 4.2\text{e-}05$; Appendix B: Table 4), while plant growth reduction (relative

213 deviation in plant growth from control) was marginally significantly related to the interaction

214 (LMM: $\chi^2 = 18.55$, $P = 0.10$; Appendix B: Table 5). Also, there was a significant effect of

215 herbivore identity on herbivory (LMM: $\chi^2 = 52.09$, $P = 1.8\text{e-}09$; Appendix B: Table 4). Apart

216 from the interactive effects, plant palatability cluster identity did neither show a significant

217 relation to herbivory nor plant growth reduction. Herbivory but not plant growth reduction

218 was higher for surviving individuals (Appendix B: Table 4). Model predictions from both

219 process models indicate that the higher herbivory, the higher plant growth reduction (Fig. 3).

220 Highest predicted herbivory and plant growth reduction were observed for *T. caelestialium*

221 on plants of the medium and high palatability cluster and for *Notostira* spp. on plants of the

222 low palatability cluster (Fig. 3). While for *Notostira* spp., no difference in herbivory rates

223 were found between the two species *N. elongata* and *N. erratica* (student's t-test: $P = 0.39$),

224 there was a tendency for higher herbivory rates in *Lygus rugulipennis* compared to *L.*

225 *pratensis* (student's t-test: $P = 0.068$). Accounting for the different *Lygus* species in the

226 analyses of herbivory rates did, however, not change the overall picture (Appendix B: Fig.

227 10).

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228 *Effect of plant and herbivore traits on herbivory and plant growth reduction*

229 Herbivory was highest for specimens with small body volume (low herbivore PC axis 1

230 values) and thin bodies (high herbivore PC axis 2 values) (Fig. 4, Appendix B: Table 6).

231 Neither plant PC axes nor the interactions between herbivore and plant PC axes were

232 significantly related to herbivory. Plant growth was reduced most strongly by large

233 herbivores (herbivore PC axis 1) on plants with high LNC (plant PC axis 2) or by small

234 herbivores on plants with low LNC, as was indicated by a significant interaction between the

235 two PC axes (Fig. 5, Appendix B: Table 7).

236 **Discussion**

237 Plant growth was clearly inhibited by herbivore presence and tended to be most strongly

238 reduced in settings that showed highest herbivory rates, supporting the potential of sucking

239 herbivores to affect plant biomass production. Because sucking herbivores withdraw

240 photosynthates from the plants, they potentially reduce their ability for growth. Furthermore,

241 herbivory can lead to plant stress-responses such as lowered photosynthesis (Sulaiman et al.,

242 2021), also resulting in lowered plant growth. Alternatively, plants may hold their C uptake

243 constant but invest a large part of their photosynthetically obtained C into defence (and thus

244 respiration) or store it in the roots, which would reduce the relative amount of C available for

245 aboveground growth (Dyer et al., 1991; Walling, 2000). Although these different

246 mechanisms can explain the observed plant growth reduction, it might still be unexpected,

247 given that in non-outbreak situations, insect herbivory is often expected to increase plant

248 productivity (Dyer et al., 1993). However, such stimulation in growth might only be apparent

249 once herbivory pressure is reduced again (Hawkins et al., 1986), which was not the case here

250 with herbivores being present during the whole experiment. Also, the study design only

251 allowed us to study the plants for two weeks after infestation with herbivores, which might

252 not be long enough to observe compensatory growth. Thus, although the observed reduction

253 in plant growth with increasing herbivory was considerable, more work needs to be done to

254 understand its quantitative impact in real-world ecosystems.

255 Process rates differed between herbivore species, but the observed relations did not match our

256 expectation that large herbivore species would consume more and reduce plant growth more,

257 as is the case for grasshoppers (Moretti et al., 2013). Although survival was included in our

258 models, this result may still have been partly influenced by differences in survival rates

259 between herbivore species. As survival rates were high for very different species such as

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260　large bugs (e.g. *Aelia acuminata*) and small leafhoppers (e.g. *Deltocephalus pulicaris*), we

261　expect other factors to be more important in explaining the observed species differences.

262　Increasing consumption rates and thus herbivory increasing with body size are generally

263　expected due to higher metabolic rates (Brown et al., 2004). While the positive relation

264　between body size and metabolic rates in herbivores is undisputed (e.g. Ehnes et al., 2011),

265　other factors can affect metabolic rates of herbivores. For example, species that are engaged

266　in regular activities with high metabolic demand (e.g. flying, producing sounds) tend to have

267　higher metabolic rates (Reinhold, 1999). The smaller species included in our study have

268　shorter generation times (Biedermann & Niedringhaus, 2004; Wachmann et al., 2004–2012),

269　which might require more activities with high metabolic demand in a shorter time to fulfil

270　their life cycle. Measures such as metabolic rate should be further addressed as potential

271　effect traits related to herbivory and plant growth reduction.

272　Mechanical plant palatability traits such as LDMC and SLA have been related to leaf

273　toughness and are thus regularly postulated to be negatively related to plant palatability for

274　chewing herbivores (e.g. Descombes et al., 2020). The lack of clear relationships in this study

275　suggests that those traits are less related to accessibility of leaf tissue and transport vessels for

276　sucking herbivores and that other traits such as nutrient contents could be stronger

277　determinants (Prestidge, 1982). Because LNC is of essential value for sucking herbivores,

278　given it is generally a major limiting nutrient in their diet (Elser et al., 2000), the lacking

279　relation between LNC and herbivory in our study is surprising. It might, however, be related

280　to the elevated LNC of all study plants compared to values reported in previous studies,

281　which was a consequence of the fertilization imposed by the labelling. Thus, the plant

282　palatability clusters that were defined based on literature traits were partly blurred.

283　Consequently, all herbivores might have met their nitrogen demand in all palatability

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284   clusters, such that differences in consumption rates rather reflect differences in physiological

285   needs of herbivores than of plant palatability defined by the three investigated plant traits.

286   While the results of our study question the usefulness of commonly used plant palatability

287   traits for sucking herbivores, further work needs to investigate which traits might be more

288   relevant for this important group of insect herbivores.

289   Both herbivory and plant growth reduction depended on the combination of herbivore species

290   and plant palatability cluster. This indicates trait matching, but because the interactive effect

291   could at best weakly be explained by the investigated traits, other traits might be involved to

292   explain the specialisation of sucking herbivores to certain grasses. In dicotyledons,

293   specialisation is often explained by the highly diversified composition in terms of secondary

294   compounds, which is postulated to be an evolutionary response to herbivores (Ehrlich &

295   Raven, 1964). Grasses, however, lack this diversity in secondary compounds (Tscharntke &

296   Greiler, 1995), posing the question of what is mainly driving specialisation. A probable factor

297   are once more varying nutrient levels among grass species and individuals, with herbivores

298   being physiologically adapted to very specific host stoichiometries (Denno & Roderick,

299   1990). Furthermore, grasses are known to use elevated silicon concentrations as defence

300   against herbivores (Vicari & Bazely, 1993). Thus, differences in silicon concentrations could

301   explain the observed patterns, although their efficacy against sucking herbivory is not well

302   understood so far (Keeping & Kvedaras, 2008). Investigating trait matching by assessing host

303   and herbivore stoichiometries and additional host defence structures could be a way forward

304   to extend this concept to sucking herbivores.

305   By labelling plants with ^{15}N isotope, we successfully quantified sucking herbivory at the

306   level of single individuals, which is otherwise hard to observe. As such, the method provides

307   great potential for future mechanistic studies on insect herbivory. We show that different

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308 herbivore species differently affect herbivory and plant growth and find indications for

309 interactive effects between herbivores and plants in determining process rates, which suggest

310 trait matching. Such relationships are in line with previous studies from grasslands with

311 grasshoppers and indicate the importance of plant and herbivore community shifts for

312 ecosystem functions such as plant biomass production. However, the traits generally

313 recognised to be involved in the relationships among plants, grasshoppers and ecosystem

314 processes had little explanatory power in our model system. This suggests that new traits

315 should be addressed to understand the consequences of changes in multi-trophic community

316 composition, e.g. in response to land-use intensification, for ecosystem functioning.

Acknowledgements

Appendix A. Supplementary materials and methods

Appendix B. Supplementary figures and tables

Supplementary data associated with this article can be found, in the online version, at

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329 **Declaration of interests**

330   The authors declare that they have no known competing financial interests or personal

331 relationships that could have appeared to influence the work reported in this paper.

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503

Figure captions

Fig. 1. (A) Allocation of Poaceae species in a two-dimensional trait space derived from principal component analysis (PCA) on LDMC, SLA and LNC (derived from a trait data base; Kattge et al., 2020). Each point is a species, colours represent assignment to three different palatability clusters (low, medium, high palatability). Arrows show PCA loadings of the three trait variables. Study species are indicated by strong colours and labels. **(B)** Allocation of study plants in the same two-dimensional trait space as in (A). Each point is an individual, colours show the palatability cluster and shapes represent the species. Ellipses show cluster allocations based on a multivariate normal distribution for a confidence level of 0.66. Agr_cap: *Agrostis capillaris*, Arr_ela: *Arrhenatherum elatius*, Poa_tri: *Poa trivialis*, Cyn_cri: *Cynosurus cristatus*, Fes_aru: *Festuca arundinacea*, Hol_lan: *Holcus lanatus*, Des_ces: *Deschampsia cespitosa*, Fes_ovi: *Festuca ovina*, Ses_cae: *Sesleria caerulea*

Fig. 2. (A) Allocation of Heteroptera and Auchenorrhyncha species that feed on Poaceae in a two-dimensional trait space derived from principal component analysis (PCA) on body volume, rostrum length and body shape. Species selection is based on a study by Neff et al. (2019), from which also species-level trait values are derived. Arrows show PCA loadings of the three trait variables. Colours indicate the two suborders. The herbivore species that were selected for the experiment are indicated by strong colours and labels. **(B)** Allocation of study specimens in the same two-dimensional trait space as in (A). Each point is an individual, colours show the species. Ellipses show cluster allocations based on a multivariate normal distribution for a confidence level of 0.66. Here and in other figures, insect icons in the legend show the outline of the study species true to scale. Ael_acu: *Aelia acuminata*, Lyg_spp: *Lygus* spp., Ste_lae: *Stenodema laevigata*, Not_spp: *Notostira* spp., Tri_cae: *Trigonotylus caelestialium*, Del_pul: *Deltocephalus pulicaris*, Lao_str: *Laodelphax striatella*

528 **Fig. 3.** Predictions from models analysing the joint effect of plant and herbivore treatment

529 and their interaction on herbivory and on plant growth reduction. Predictions are shown for

530 different plant palatability clusters (panels; high, medium, low palatability) and for different

531 herbivore species (colours). Model predictions and standard errors are shown. Herbivory was

532 log-transformed prior to modelling to meet distributional assumptions, but transformed back

533 for the illustration of this figure. Models also account for herbivore survival and cage

534 position. Ael_acu: *Aelia acuminata*, Lyg_spp: *Lygus* spp., Ste_lae: *Stenodema laevigata*,

535 Not_spp: *Notostira* spp., Tri_cae: *Trigonotylus caelestialium*, Del_pul: *Deltocephalus*

536 *pulicaris*, Lao_str: *Laodelphax striatella*. Detailed model results in Appendix B: Tables 4 and

537 5

538 **Fig. 4.** Predicted herbivory from a model analysing the joint effect of plant and herbivore

539 traits and their interactions (colour gradient). Plant and herbivore traits were represented by

540 PC axes (Fig. 1 and Fig. 2), whose meaning is indicated by the arrows below and right to the

541 graphs. Points show the spread of underlying observations in trait space. All non-significant

542 interactions were excluded from the model, leaving no interaction between plant and

543 herbivore traits in this model. Models also account for herbivore survival and cage position.

544 Significance of variables is indicated next to axis labels (***: $p \leq 0.001$, **: $p \leq 0.01$,

545 *: $p \leq 0.05$). ^{15}N uptake was log-transformed prior to modelling to meet distributional

546 assumptions, but transformed back for the illustration of this figure. Detailed model results in

547 Appendix B: Table 6

548 **Fig. 5.** Predicted plant growth reduction from a model analysing the joint effect of plant and

549 herbivore traits and their interactions (colour gradient). Plant and herbivore traits were

550 represented by PC axes (Fig. 1 and Fig. 2), whose meaning is indicated by the arrows below

551 and right to the graphs. Points show the spread of underlying observations in trait space. All

28

552   non-significant interactions were excluded from the model, leaving only the interaction

553   between herbivore PC axis 1 and plant PC axis 2 in the model. Models also account for

554   herbivore survival and cage position. Significance of variables is indicated next to axis labels

555   (***: $p \leq 0.001$, **: $p \leq 0.01$, *: $p \leq 0.05$). Detailed model results in Appendix B: Table 7
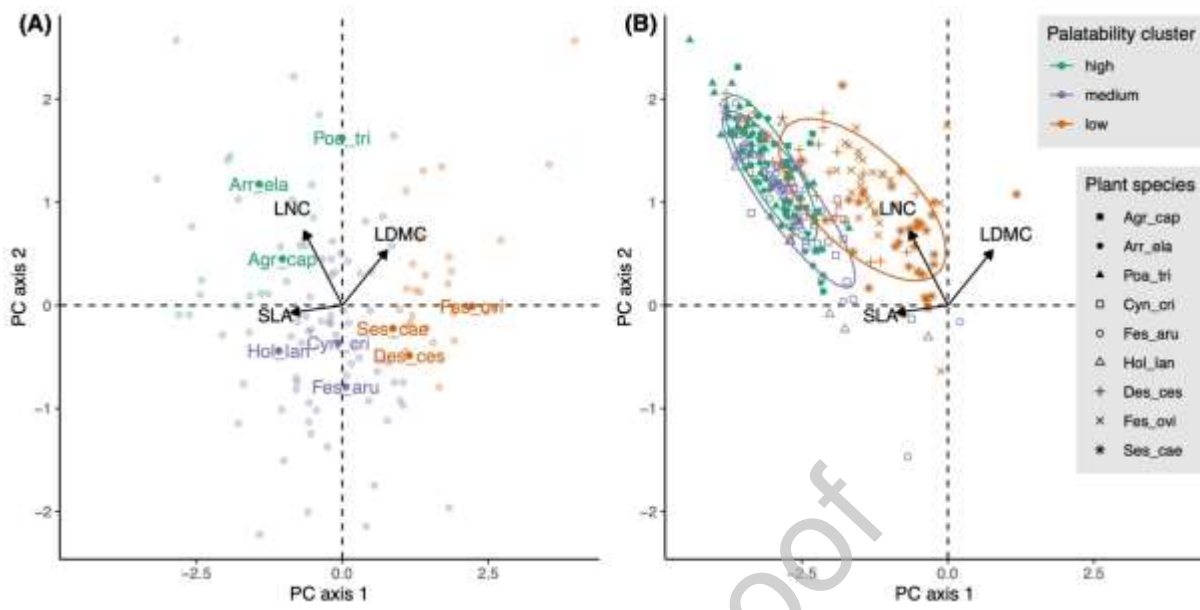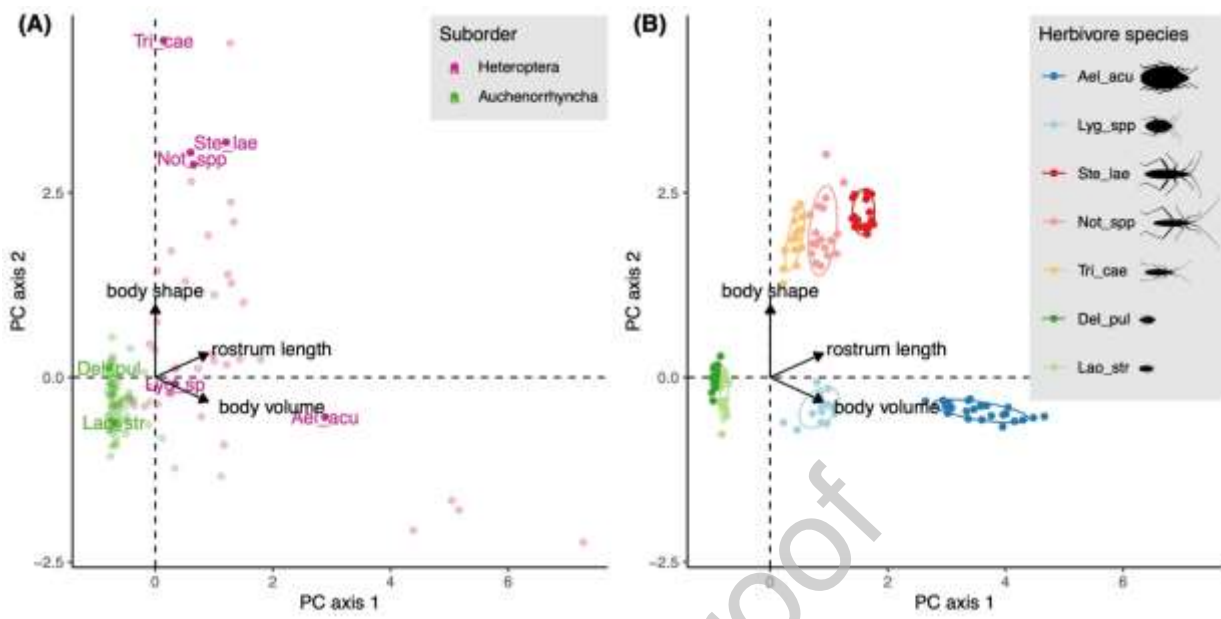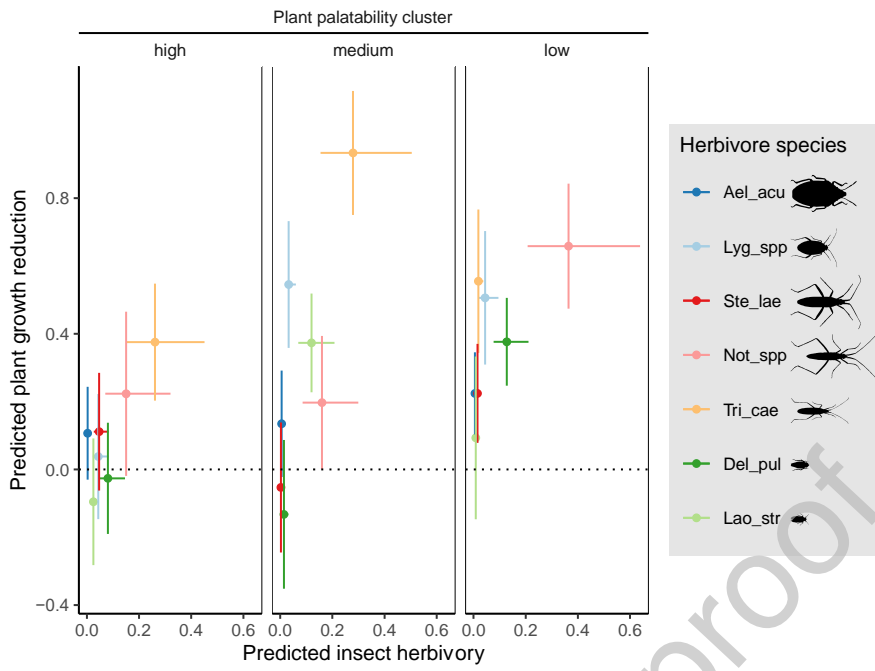
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Figure 1

Figure 2

Figure 3

Figure 4

Figure 5