Accepted Manuscript

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1 *Solidago canadensis* **impacts on native plant and pollinator communities in different-**

2 **aged old fields**

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22

23   **Abstract**

24

25   Secondary succession in former arable fields (i.e. old fields) might be altered by the

26   colonization of invasive alien species, with possible community-wide impacts, hindering the

27   ability old fields to become species-rich communities. However, the effects of invasive

28   species on local communities have rarely been addressed in the light of secondary succession.

29   Therefore, we studied the impact of the highly invasive *Solidago canadensis* on plant and

30   pollinator communities along a gradient of invasion severity in old fields with different ages

31   (1–20 years since last ploughing) in Southern Transylvania, Romania. We asked whether the

32   invasion of *S. canadensis* causes shifts in (1) the composition and diversity of plant

33   communities, and (2) pollinator communities along the successional gradient. Further, we

34   asked (3) to what extent the presence of *S. canadensis* affected flower visitation of native

35   plant species by pollinators. According to our results, the invasion reduced the native plant

36   species richness throughout succession, although the most profound negative effect on plant

37   diversity and vegetation naturalness was exerted in older successional communities. The

38   invasion of *S. canadensis* had a negative effect on the abundance of bees irrespective of the

39   old field age; however, there was no similar negative effect on hoverflies. Native flowers

40   experienced reduced visitation by wild bees, honey bees and hoverflies due to the augmented

41   presence of *S. canadensis*. Therefore, the invasion of this perennial plant species diverts the

42   trajectory of vegetation succession, alters the mutualistic links between the native elements of

43   these old fields, and causes a non-desired alternative stable states to be installed.

44

45   **Keywords:** Canada goldenrod; plant invasion; plant–pollinator interaction; fallow; set-aside;

46   *Apis*; Romania; Natura 2000; biodiversity conservation

47

48 **Introduction**

49

50 Over the last few decades, various social and economic changes have driven the abandonment

51 of extensive areas of formerly arable lands in several parts of the world. If these areas are left

52 alone and propagule sources are available, secondary succession takes place and diverse semi-

53 natural vegetation can be formed in short time (Stoate et al. 2009). Former arable lands

54 (hereafter old fields) may support farmland biodiversity and provide valuable ecosystem

55 services (e.g. pollination) at the beginning of secondary succession (Tscharntke, Batáry &

56 Dormann 2011) and their conservation value may increase in the later stages (Corbet 1995).

57 However, reaching a stage of diverse secondary vegetation is not free from challenges, as old

58 fields are highly susceptible to invasion by alien plant species (Catford et al. 2012). Non-

59 native invasive species are likely to colonize the freshly abandoned croplands because of

60 increased soil nutrient availability associated with previous fertilizer applications, and limited

61 competition for resources (Standish, Cramer & Hobbs 2008). Longer-lived alien species can

62 persist until the later stages of succession, therefore perturbing natural succession pathways

63 and altering the pattern of vegetation recovery (Meiners, Pickett & Cadenasso 2002). By

64 preventing the establishment of late-successional species, alien plant invasion may cause the

65 invaded habitats to remain in the early successional stages for a long time (Cramer, Hobbs &

66 Standish 2008).

67 The establishment of invasive plants may influence not only the native flora and the

68 trajectory of its secondary succession, but can also have an impact on the related invertebrate

69 communities, such as pollinator insects (van Hengstum et al. 2014). Former studies presented

70 highly controversial results, reporting either positive (Bartomeus, Vilà & Santamaría 2008) or

71 negative effects (de Groot, Kleijn & Jogan 2007; Moroń et al. 2009) on pollinator abundance

72 and diversity. Invasive plants are reported to influence the abundance and diversity of smaller,

73   mostly solitary wild bees (Apoidea) of limited dispersal ability (Gathmann & Tscharntke

74   2002), as well as hoverflies (Syrphidae) and bumblebees, which generally have larger

75   foraging ranges (Greenleaf et al. 2007). Therefore, invasive species might threaten

76   biodiversity not only at the local level, but at the landscape scale as well (Hejda, Pyšek &

77   Jarošík 2009). Moreover, invasive plant species can also act indirectly on ecological

78   communities by weakening the mutualistic links between native plants and their pollinators

79   (Aizen, Morales & Morales 2008) by luring pollinators away from native plants, either

80   partially (Baskett, Emery & Rudgers 2011) or completely (Bartomeus, Vilà & Santamaría

81   2008). This expansion of plant preference for pollination can result in fewer pollinator visits,

82   pervasive interspecific pollen transfer (Baskett, Emery & Rudgers 2011), and consequently,

83   decreased reproduction success and diversity of the native flora. Despite these possible

84   detrimental effects, the direct effect of invasive plant species on secondary succession of old

85   fields is relatively understudied (Flory and Clay 2010), while the effects of invasive species

86   on the pollinator communities along the successional trajectory has so far never been studied.

87   Here, we focus on a large-scale biological invasion phenomenon in southern

88   Transylvania, Romania. Large-scale abandonment of arable lands has occurred over the last

89   two decades due to a significant class of absentee landowners and the decreasing profitability

90   of traditional subsistence agriculture (Fischer, Hartel & Kuemmerle 2012). *Solidago*

91   *canadensis* L. (Canada goldenrod) is the most successful invasive colonizer of these old

92   fields, often becoming dominant even at the landscape scale. Its impact on the species

93   richness of vascular plants and pollinators has already been studied in other countries (de

94   Groot, Kleijn & Jogan 2007; Moroń et al. 2009). However, these studies analysed only the

95   final stage of invasion, when the invasive species had already established dominance in the

96   community, and compared this stage with non-invaded, semi-natural communities,

97   representing only the two contrasting extremes of the invasion gradient.

4

98   To fill these gaps in our knowledge regarding the impact of invasion on both plant and

99   pollinator communities over the course of secondary succession on old fields along a

100   continuous gradient of invasion, we initiated a large-scale field study. We aimed to answer the

101   following questions: (1) Does the invasion of *S. canadensis* cause shifts in the composition

102   and diversity of plant communities over the course of secondary succession of old fields? (2)

103   Does abundance and species richness of pollinators change due to the invasion of the old

104   fields? (3) To what extent does the presence of *S. canadensis* affect flower visitation of native

105   plant species by pollinators? To answer these questions, we studied the effect of *S. canadensis*

106   density (0–80% relative cover) in 36 old fields with various ages (1–20 years since the last

107   ploughing).

108

109   **Materials and methods**

110   *The invasive species*

111   *S. canadensis* is a rhizomatous, patch-forming perennial herb of the Asteraceae. It has become

112   an exceptionally successful invasive species since its introduction to Europe in the 18th

113   century and is now widespread throughout the continent (Weber 1998). In addition to its

114   prolific vegetative propagation (Meyer & Schmid 1999), *S. canadensis* releases chemicals

115   that inhibit the growth, germination and survival of native plants (Abhilasha et al. 2008), and

116   change the soil composition by diverting nutrients and minerals (Zhang et al. 2009).

117

118   *Location and study sites*

119   Our study area was located in the middle section of the Târnava Mare Valley in Southern

120   Transylvania, Romania (Fig. 1A). Climate is classified as moderate continental. Mean annual

121   rainfall is approximately 650 mm, while mean annual temperature is 8 °C (Drăgulescu 2003).

122   The region has been designated as Natura 2000 Site of Community Interest (Sighişoara-

123   Târnava Mare Natura 2000 site, ROSCI0227) because 18 habitats listed in the EU Habitats

124   Directive Annex I can be found in this region.

125   We examined the succession of vegetation and pollinator communities in this site

126   using a chrono-sequence of old fields representing different stages of succession (space for

127   time substitution, sensu Pickett 1989). We chose old fields with a known abandonment date in

128   the vicinity of four villages (Biertan, Laslea, Malâncrav, Richiş; Fig. 1A). Abandonment here

129   can be defined as the cessation of ploughing, though other uses and activities such as mowing

130   or grazing may be present in the fields. These old fields may also be infrequently burned by

131   locals. The 36 fields examined ranged in age from 1 to 20 years since abandonment. These

132   sites were also chosen to represent the whole range of *S. canadensis* abundance from highly to

133   less invaded areas (relative cover 0–80%). Most sites were quite small (mean area: 1.3 ha,

134   range: 0.08–2.7 ha). The year of last ploughing, and current land-use practices were

135   determined by interviewing landowners, and based on that information, we categorized the

136   old fields as mown, grazed or without management. To determine whether the sites had been

137   recently burned, or not, we looked for local signs of fire (burned trees and shrubs, incinerated

138   litter or grass tussocks) and asked local people.

139

140   ***Vegetation survey***

141   Percent cover of vascular plant species with a resolution of 1% was visually estimated within

142   three 4×4 m plots per site (Fig. 1B). We assigned 0.5% to species with a cover smaller than

143   1% (species represented by one small individual or by seedlings only). The level of invasion

144   was characterized in each plot by the relative cover of *S. canadensis* and averaged over the

145   three plots at site level. The surveys were conducted once per site in May–June 2012.

146   In order to accurately depict compositional changes during the course of succession,

147   we calculated the following variables for each site:

6

148   1. *Native vascular plant species richness and diversity*. Species richness was calculated by

149   averaging the number of species in the three plots per site. The Shannon diversity index of

150   each site was calculated based on the proportional cover of each species. *S. canadensis* was

151   not included in the calculation.

152   2. *Naturalness*. All plant species were assigned to one of the three naturalness groups

153   according to Sanda et al. (1983): (i) species of natural and semi-natural habitats, hereafter

154   called "grassland species", (ii) species common to both natural and ruderal communities,

155   hereafter "generalist species", and (iii) species of ruderal communities, hereafter "ruderal

156   species". The proportion of each category was calculated for each site and used in subsequent

157   analyses.

158   3. *Functional guilds.* The proportion of the main functional groups was taken into account by

159   distinguishing (i) graminoid species belonging to the Poaceae, Cyperaceae or Juncaceae, (ii)

160   legume species of the Fabaceae and (iii) forbs, i.e. herbaceous plants belonging to other

161   families. As the presence of tree and shrub species never exceeded 1% average cover at site

162   level, we excluded those species from the calculation of this variable.

163

164   ***Pollinator sampling***

165   We sampled pollinator insects in 22 sites (out of 36 sites with botanical surveys). We selected

166   these sites to cover the whole range of successional age (1 to 20 years) and low to high

167   densities of *S. canadensis* cover. The sites were at least 250 m from each other. Pollinators

168   were sampled along two 100 m transects per study site; each transect assigned was at least 15

169   m from the field edge and 15 m apart from each other (Fig. 1B). We surveyed the pollinators

170   twice: first during the peak flowering season of indigenous vegetation (21–25 July 2012) and

171   second during the peak flowering of *S. canadensis* (19–23 August 2012). Transect counts

172   were performed between 9 a.m. and 6 p.m. under favourable weather conditions with little

173   wind and few clouds at most. A pair of observers walked along each transect for 20 min and

174   recorded all insects actively pollinating, flying or perching on the vegetation. In the case of

175   active pollination, the visited plant species was also recorded. Pollinators were hand-netted

176   and preserved in 96% ethanol for later identification at the species level.

177   We distinguished four pollinator groups: honey bee (*Apis mellifera* L.), bumblebees

178   (*Bombus* spp.), other wild bees (Apoidea: Collettidae, Melittidae, Halictidae, Megachilidae,

179   Andrenidae, Apidae except *Bombus* spp. and honey bee), and hoverflies (Syrphidae).

180   Bumblebees and other wild bees were analysed separately because these two groups have

181   different biological traits in terms of floral requirements, flying abilities and sociality

182   (Gathmann & Tscharntke 2002; Greenleaf et al. 2007; Michener 2007), and therefore

183   different responses to landscape and local scale environmental conditions were expected.

184   Although there are some semi-social species and/or genera among the collected bees (e.g.

185   some *Halictus* spp.), we use 'solitary bees' in the current paper for wild bees other than

186   bumblebees.

187   As the presence of pollinators depends on the pollen and nectar supply, we gathered

188   additional botanical information: flowering plant species and the number of flowers at species

189   level were recorded at ten 1×1 m quadrates placed equidistantly along the same two transects

190   per site (Fig. 1B). We counted the number of heads in the case of Asteraceae species and

191   simple umbels for Apiaceae, but refer to both as flowers for the sake of simplicity.

192   We calculated several variables to detect potential changes in the pollinator

193   communities during succession:

194   1. *Abundance of each pollinator group* (bumblebees, solitary bees, honey bee, hoverflies) per

195   transect was calculated as the number of individuals per group per transect;

196   2. *Species richness of bumblebees, solitary bees and hoverflies*; species data from the two

197   transects per site were pooled because of the relatively low species numbers;

8

198   3. *Number of visited flowers other than S. canadensis* was calculated as the total number of

199   flowers visited (actively pollinated) by each pollinator group per transect.

200

201   ***Statistical analyses***

202   All statistical analyses were carried out in the R statistical environment version 3.0.1 (R Core

203   Team 2013). We built separate linear mixed-effects models (LMMs) for each vegetation and

204   pollinator variable using the *nlme* package (Pinheiro et al. 2013).

205   For vegetation data, the dependent variables were plant species richness, diversity,

206   naturalness and functional guilds, all calculated from the data collected in the three 4×4 m

207   plots per site. We included old field age, the proportional cover of *S. canadensis* and field

208   area (in ha) as continuous fixed variables, land-use type (mown, grazed, without

209   management), anthropogenic fire (burned, unburned) as fixed factors, and village (4 levels) as

210   a random term. In addition, the pairwise interaction between fixed variables and proportional

211   cover of *S. canadensis* was also included the models.

212   For pollinator variables (abundance and species richness of pollinator groups and the

213   number of visited flowers except *S. canadensis*) land-use, fire, sampling period (July *vs*.

214   August) were included as fixed factors. The proportional cover of *S. canadensis*, field area,

215   old field age, species richness of flowering plants, the number of flowers of native species and

216   the number of *S. canadensis* flowers (collected from the ten 1×1 m quadrates placed along

217   the pollinator transects) were built in as fixed covariates. The two-way interactions between *S.*

218   *canadensis* cover and the other five explanatory variables (except flower variables) were also

219   included in the models. We included study site nested within village as a random effect for

220   abundance and number of visited flowers; we used only village as a random effect for species

221   richness of pollinators because data from the two transects per sites were pooled due to low

222   number of available species at the transect level.

9

223   In each case, we started with saturated models (Appendix A) and removed non-

224   significant variables using backward stepwise selection, except those that were part of a

225   significant interaction. We extracted the contrasts for retained significant terms as follows: the

226   contrasts for covariates or interactions between covariate and factors, based on differences of

227   least square means, were extracted using the *lsmeans* function of the *lsmeans* package (Lenth

228   2013). For land-use types, Tukey contrasts were performed using the *glht* function of

229   *multcomp* package (Hothorn, Bretz & Westfall 2008). Contrasts for interactions between

230   covariates cannot be computed, hence we drew regression surface plots with the dependent

231   variable and the interacting covariates. Normality of the residuals was tested using the

232   Shapiro-Wilk test and homoscedasticity of the residuals verified by plot diagnosis.

233   Abundance variables were log-transformed prior to analyses to reach normal residual

234   distribution. We calculated effect sizes (partial r) and their 95% confidence intervals

235   (Nakagawa & Cuthill 2007) to present the biologically meaningful magnitude by which the

236   significant explanatory variables influenced plant and pollinator abundance and diversity.

237   Before building LMMs, we tested all predictor variables for multicollinearity by

238   calculating the variance inflation factor (VIF) using *vif* function of the *fmsb* package in R. VIF

239   values were <2 in all cases, so no multicollinearity was detected in any of the performed

240   models (Rogerson 2001). In addition, all dependent variables were tested for spatial

241   autocorrelation (see Appendix A).

242

243   **Results**

244   *Species richness, diversity and composition of plant community*

245   The old field age, the proportional cover of *S. canadensis*, and their interactions were

246   important sources of variation for all vegetation variables (Table 1). Plant species richness

247   and diversity increased with field age. The density of *S. canandensis* was negatively

248 associated with the number of plant species irrespective of old field age and with the diversity

249 of older fields (Table 1, Fig. 2A and B).

250 The proportion of grassland species increased, while that of generalist and ruderal

251 species decreased with old field age. The effect of *S. canadensis* cover was non-significant in

252 all three naturalness groups. However, *S. canadensis* did significantly interact with old field

253 age, as the proportion of grassland species decreased, and that of generalist species increased

254 in the older fields, while the abundance of generalist species declined in younger fields with

255 higher *S. canadensis* cover (Table 1, Fig. 2C, D and E).

256 In the case of the plant functional guilds, the proportion of graminoids and legumes

257 increased, while that of forbs decreased with old field age (Table 1). Despite their temporal

258 decline, forbs were the dominant group throughout succession. High *S. canadensis* cover was

259 associated with increased proportion of legumes in younger sites, and that of graminoid

260 species in older fields, while the proportion of forbs decreased irrespective of old field age

261 (Fig. 2F, G and H). Land-use type predicted only the proportion of legumes, which was

262 smaller in mown and grazed sites than in sites without management (Table 1).

263

264 ***Abundance and species richness of pollinators***

265 The abundance of solitary bees was significantly decreased by *S. canadensis* cover in July (F

266 = 6.33, p = 0.022), but no such effect was found in August (Table 2, Fig. 3A). The abundance

267 of solitary bees was influenced by land-use, sampling time and species richness of actually

268 flowering plants (Table 2). The abundance of bumblebees was decreased by *S. canadensis*

269 cover in July (F = 6.78, p = 0.018, Fig. 3A) and marginally significantly increased in August

270 (F = 3.03, p = 0.099, Fig. 3B). Bumblebees were present in higher abundance in July than in

271 August, and enhanced by both native and *S. canadensis* flower abundance. The abundance of

272 honeybees decreased with *S. canadensis* cover and old field age. The abundance of hoverflies

273   showed the opposite pattern with higher values in August than in July. Hoverfly abundance

274   was enhanced by the field area and by the abundance of *S. canadensis* flowers (Table 2).

275   The species richness of solitary bees was higher in July than in August and increased

276   with the number of flowering plant species. Similarly, more bumblebee species were found in

277   July than in August. The species richness of hoverflies increased with field area and the

278   flower abundance of both native species and *S. canadensis* (Table 2).

279

280   *Flower visitation*

281   In general, the number of flowers (excluding *S. canadensis*) visited by bumblebees and

282   solitary bees was higher in July than in August (Table 3). The flower visitation of indigenous

283   plant species by solitary bees and honey bees was positively influenced by the number of

284   flowering species. The abundance of flowers (excluding *S. canadensis*) had a positive effect

285   on the number of bumblebee-visited flowers (Table 3). Further, the increasing cover of *S.*

286   *canadensis* had a significant negative effect on the flower visitation of native species by

287   solitary bees, honey bees and hoverflies (Table 3, Fig. 4). In addition, we found a significant

288   interaction between sampling time and *S. canadensis* cover: the number flowers (excluding *S.*

289   *canadensis*) visited by bumblebees was slightly decreased by *S. canadensis* in July (Fig. 4A)

290   but marginally significantly increased in August (Fig. 4B).

291

292   **Discussion**

293

294   *Effects of S. canadensis invasion on different-aged old field plant communities*

295   Although it was already known that plant communities invaded by *S. canadensis* might suffer

296   shifts in diversity and composition (de Groot, Kleijn & Jogan 2007; Moroń et al. 2009), we

297   are the first to report reduced species richness and altered naturalness and functional

298   composition in the context of secondary succession.

299   The early years of vegetation succession are usually characterized by a wide array of

300   disturbance-tolerant ruderal species and fast colonizing generalist species (Ruprecht 2006).

301   *Solidago canadensis* displaced the native generalist species in young successional fields,

302   which might slow down the secondary succession, as generalist species would lead the

303   community to the next stage of succession by outcompeting disturbance-tolerant ruderal

304   species (Catford et al. 2012). *Solidago canadensis* can monopolize resources and hinder the

305   colonization of other species due to its quick capacity for vegetative expansion and a dynamic

306   foraging strategy (Bartha et al. 2014). Only native legumes were able to increase their

307   importance during the early years of the succession, probably due to their nitrogen-fixing

308   ability.

309   In older successional fields, the species turnover decreases, and biotic filters can be

310   particularly important (Pickett et al. 2001). If a monopolist species becomes dominant (fast-

311   growing clonal species tending to eliminate other species, sensu Faliňska 1991), it may

312   outcompete the subordinate species and may have a strong negative impact on local diversity

313   (Bartha et al. 2014). *S. canadensis* seems to be a monopolist species as it negatively

314   influenced the species richness of forbs and the diversity of the older fields. It outcompeted

315   species belonging to the same functional group (perennial non-nitrogen-fixing forbs), but

316   seemed to facilitate or have no effect on graminoid species. Fargione, Brown and Tilman

317   (2003) proved that the strongest inhibitory effects of resident plants are on introduced plants

318   of the same functional guild. Our results indicate that this relationship may work inversely as

319   well: the invasive species might out-compete the natives belonging to the same functional

320   group.

13

321   Most of the grassland specialists are expected to enter the community at mid-

322   successional stages, because they are adapted to colonize the few available microsites under a

323   closed canopy (Bartha et al. 2014). However, the increased presence of disturbance-tolerant

324   generalists and the decreased proportion of the grassland specialists in the invaded old fields

325   underlines that if this invasive species is abundant, secondary succession slows down, or even

326   halts at a prolonged earlier successional state (Cramer, Hobbs & Standish 2008) instead of

327   recovering towards semi-natural grassland vegetation. Similar results were found in mid-

328   successional old fields invaded by *S. gigantea* as well, having very strong negative impact on

329   succession (Bartha et al. 2014).

330

331   ***Effect of S. canadensis on the abundance and species richness of pollinators***

332   The decreased plant species richness and especially the decline of forbs led to food resources

333   becoming more limited for pollinators in the old fields we studied (see also de Groot et al.

334   2007). As a more diverse and flower rich plant community can harbour a more diverse and

335   stable pollinator assemblage over time (Ebeling et al. 2008), it is understandable why the

336   abundance of solitary bees and bumblebees declined in July, the pre-blooming period of the

337   invader (see also de Groot, Kleijn & Jogan 2007; Moroń et al. 2009; Lenda, Skórka & Moroń

338   2010). However, in August, the myriad of flowers produced by *S. canadensis* had a positive

339   effect on the abundance of bumblebees and on the abundance and species richness of the

340   mostly generalist hoverflies. Therefore, this invasive species might serve as an alternative

341   food resource for certain pollinators during its flowering period, especially for bumblebees

342   that have still persistent colonies until late summer/early autumn, and for late-emerging

343   hoverflies (see also de Groot et al. 2007). From this point of view, *S. canadensis* may

344   indirectly act as facilitator by maintaining larger pollinator populations throughout the

345   flowering season (Bjerknes et al. 2007).

346   *S. canadensis* is considered one of the most important plants yielding unifloral honey

347   sources in Central Europe (Farkas and Zajácz 2007). Therefore, a highly interesting finding is

348   that the increasing cover of *S. canadensis* significantly decreased the abundance of honey

349   bees, even during mass flowering. Although honey bees are important pollinators of *S.*

350   *canadensis*, they are likely to begin visiting goldenrod flowers only when the abundance of

351   other flowering plant species has declined (Gross and Werner 1983). As the traditionally

352   managed landscape we studied is a diverse mosaic of croplands, old fields and managed

353   grasslands (pastures or hayfields), pollinators can easily find other pollen sources in the

354   vicinity of the invaded old fields.

355   Although plant species richness and composition went through considerable alteration

356   along the 20 years of succession, neither solitary bees nor hoverflies showed any difference

357   among old fields of different age. Therefore, the bimodal species richness distribution of

358   pollinators along secondary succession presented by Steffan-Dewenter and Tscharntke (2001)

359   could not be replicated for bees or for hoverflies in our study. This is because neither the

360   number of flowering species, nor the number of flowers depended on the age of old fields

361   (data not shown). One exception was the honey bee, which seemed to prefer plant species

362   typical of the beginning of succession in younger old fields.

363

364   ***Indirect effects of invasion: flower visitation of native species***

365   Plants and their pollinators are tightly intertwined components of ecological communities,

366   therefore the adverse effect of *S. canadensis* might have further indirect repercussions as well.

367   Alien plant species can have a strong effect on the pollination success of native species, often

368   competing with them by causing either reduced pollinator visitation rates or increased

369   heterospecific pollination of native flowers. However, in some cases, both at local and

15

370　landscape scales, these often mass-flowering invasives may facilitate native plant pollination

371　by increasing pollinator densities (Bjerknes et al. 2007).

372　　　　We found a negative correlation between the cover of *S. canadensis* and flower

373　visitation of native plants (hereafter flowers) by bees and hoverflies. On the one hand this

374　result might be the consequence of an indirect competition for pollination between the

375　superior invasive and suppressed native species, competing possibly for light, space, soil

376　nutrients. On the other hand, the positive effect of *S. canadensis* on bumblebees and

377　hoverflies suggests also direct competition for pollination between invasive and native species

378　in August due to the large amount of pollen and nectar provided by *S. canadensis*. The lower

379　frequency of flower visitation induced by invasives can possibly result in a pollination deficit

380　in native species (Chittka & Schürkens 2001). The reduction in the pollinator pool by alien

381　plants might thus corrupt the reproductive success of insect-pollinated native plants (Stout &

382　Morales 2009). Although flowers of *S. canadensis* are self-incompatible and dependent on the

383　presence of pollinators for seed production (Gross & Werner 1983), it can also vigorously

384　spread locally by rhizomes, thus its reproductive gain is less affected by a decreased

385　pollinator pool. Through this two-way competition by direct and indirect routes, *S. canadensis*

386　can further propagate its dominance and, therefore, contribute to the development of an

387　alternative, self-perpetuating stable state in which it dominates.

388

Conclusions and recommendations

390　Without management interventions, degraded, alternative states driven by *S. canadensis*,

391　instead of valuable secondary grasslands, might dominate the landscape for decades (this

392　invasive species can persist in the fields for up to 50–75 years; Hartnett and Bazzaz 1983),

393　and could arrest the natural trajectory of secondary succession in an earlier, less diverse

394　phase. Therefore, we cannot allow spontaneous processes to revegetate the old fields invaded

395   by *S. canadensis*. Instead, active management actions are needed to guide vegetation

396   recovery.

397   Long-term experiments testing the effects of different land-use techniques on

398   controlling *S. canadensis* are already available from Central-Eastern Europe (Horváth 2012).

399   According to these experiments, a combination of prescribed disturbances, such as regular

400   mowing and extensive autumnal grazing with cattle or sheep, could reduce the target invasive

401   species' abundance and enhance the recovery of native vegetation.

402

411

412   Appendix A. Supplementary data

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

414   XXXXX."

415

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23

525 **Table 1.** The effects of *Solidago canadensis* cover, age of the old fields and land-use (M – mown, W – without management) on vegetation

526 characteristics according to the linear mixed-effects models. Positive effects are indicated by "+" and negative effects by "–". Effect sizes (Partial

527 r) and their 95% confidence interval are also shown. P-values of significant effects are in bold. Significant interactions between the covariates *S.*

528 *canadensis* cover and old field age have no contrasts but are plotted on Fig. 2.

	df	F	p	Contrasts	Effect size	95% C.I.
Richness and diversity						
Plant species richness						
S. canadensis cover	1, 29	4.43	**0.044**	–	0.09	-0.24; 0.43
Old field age	1, 29	32.88	**<0.001**	+	0.72	0.38; 1.06
SCC × OFA	1, 29	3.09	0.088			
Diversity						
S. canadensis cover	1, 29	20.43	**<0.001**	–	0.24	-0.09; 0.58
Old field age	1, 29	6.07	**0.019**	+	0.61	0.27; 0.95
SCC × OFA	1, 29	15.19	**<0.001**		-0.58	-0.92; -0.24
Naturalness						
Grassland species						
S. canadensis cover	1, 29	0.01	0.956			
Old field age	1, 29	53.51	**<0.001**	+	0.80	0.46; 1.14
SCC × OFA	1, 29	5.30	**0.028**		-0.39	-0.73; -0.05
Generalist species						
S. canadensis cover	1, 29	0.94	0.339			
Old field age	1, 29	21.43	**<0.001**	–	-0.72	-1.06; -0.38
SCC × OFA	1, 29	10.69	**0.002**		0.51	0.17; 0.86
Ruderal species						
Old field age	1, 31	45.09	**<0.001**	–	-0.76	-1.11; -0.42
Functional guilds						
Graminoids						

	df	F	p		Estimate	CI
S. canadensis cover	1, 29	1.56	0.221		0.21	-0.12; 0.55
Old field age	1, 29	9.53	**0.004**	+	0.40	0.06; 0.74
SCC × OFA	1, 29	5.78	**0.022**			
Legumes						
S. canadensis cover	1, 27	5.42	**0.027**	+	0.52	0.18; 0.86
Old field age	1, 27	8.10	**<0.008**	+	0.68	0.34; 1.02
SCC × OFA	1, 27	8.39	**0.007**		-0.48	-0.82; -0.14
Land-use	1, 27	4.47	**0.021**	W>M	-0.47	-0.81; -0.13
Forbs						
S. canadensis cover	1, 30	12.30	**0.001**	−	-0.50	-0.84; -0.16
Old field age	1, 30	38.31	**<0.001**	−	-0.74	-1.09; -0.40

529

530

Table 2. The effects of *S. canadensis* cover, old field age, land-use (G – grazed, W – without management), sampling time (July, August) and several vegetation parameters on the abundance and species richness of different pollinator groups according to the linear mixed effect models. Flower abundance refers to all flowers except those of *S. canadensis*. Positive effects are indicated by "+", negative effects by "–". Effect sizes (Partial r) and their 95% confidence interval are also shown. P-values of significant effects are in bold.

	df	F	p	Contrasts	Effect size	95% C.I.
Abundance						
Solitary bees						
S. *canadensis* cover	1, 15	4.19	0.059	(–)	-0.64	-1.08; -0.19
Land-use	2, 15	3.80	**0.046**	W > G	-0.58	-1.03; -0.13
Sampling time	1, 63	38.87	**<0.001**	Jul > Aug	0.48	0.03; 0.93
Flowering plant species richness	1, 63	21.81	**<0.001**	+	0.32	-0.13; 0.77
SCC × ST	1, 63	7.29	**0.009**	July: –		
Bumblebees						
S. *canadensis* cover	1, 17	1.14	0.301		-0.60	-1.05; -0.15
Sampling time	1, 62	31.48	**<0.001**	Jul > Aug	0.28	-0.17; 0.73
Flower abundance	1, 62	9.18	**0.004**	+	0.05	-0.40; 0.50
S. *canadensis* flower abundance	1, 62	4.53	**0.037**	+	0.41	-0.04; 0.86
SCC × ST	1, 62	12.21	**0.001**	Jul –; Aug (+)		
Honey bee						
S. *canadensis* cover	1, 16	13.06	**0.002**	–	-0.68	-1.13; -0.23
Old field age	1, 16	8.11	**0.012**	–	-0.58	-1.03; -0.13
Hoverflies						
Area	1, 17	4.94	**0.040**	+	0.54	0.09; 0.99
Sampling time	1, 64	7.25	**0.009**	Jul < Aug	-0.04	-0.49; 0.41
S. *canadensis* flower abundance	1, 64	35.03	**<0.001**	+	0.59	0.15; 1.04

26

Species richness

Solitary bees						
Sampling time	1, 38	19.02	**<0.001**	Jul > Aug	-0.52	-0.97; -0.07
Flowering plant species richness	1, 38	21.30	**<0.001**	+	0.60	0.15; 1.05
Bumblebees						
Sampling time	1, 39	4.25	**0.046**	Jul > Aug	-0.31	-0.76; 0.14
Hoverflies						
Area	1, 37	12.22	**0.001**	+	0.50	0.05; 0.95
Flower abundance	1, 37	14.35	**0.001**	+	0.58	0.13; 1.03
S. canadensis flower abundance	1, 37	6.07	**0.019**	+	0.38	-0.07; 0.83

535

536

537 **Table 3.** The effects of *S. canadensis* cover, old field age, sampling time and several vegetation parameters on the number of flowers (excluding

538 *S. canadensis*) visited by different pollinator groups according to linear mixed effect models. Flower abundance refers to all flowers except those

539 of *S. canadensis*. Positive effects are indicated by "+", negative effects by "−". Effect sizes (Partial r) and their 95% confidence interval are also

540 shown. P-values of significant effects are in bold

	df	F	p	Contrasts	Effect size	95% C.I.
Number of visited flowers (excluding *S. canadensis*)						
Solitary bees						
S. canadensis cover	1, 17	11.47	**0.004**	–	-0.47	-0.92, -0.02
Sampling time	1, 64	81.71	**<0.001**	Jul > Aug	-0.72	-1.17; -0.27
Flowering plant species richness	1, 64	13.76	**<0.001**	+	0.42	-0.03; 0.87
Bumblebees						
S. canadensis cover	1, 17	1.08	0.313			
Sampling time	1, 63	36.64	**<0.001**	Jul > Aug	-0.54	-0.99; -0.09
Flower abundance	1, 63	8.37	**0.005**	+	0.29	-0.16; 0.74
SCC × ST	1, 63	6.54	**0.013**	Jul (−); Aug: (+)	0.31	-0.14; 0.76
Honey bee						
S. canadensis cover	1, 17	15.74	**0.001**	–	-0.63	-1.08; -0.18
Flowering plant species richness	1, 65	3.70	0.059	(+)	0.23	-0.22; 0.68
Hoverflies						
S. canadensis cover	1, 17	4.31	0.053	(−)	-0.45	-0.90; 0.01

541

542

543 **Figure legends**

544

545 **Fig. 1.** Map of the study area with the Natura 2000 site delineated. The four villages, where

546 the sampling took place are shown in black (A). Schematic map showing sampling design
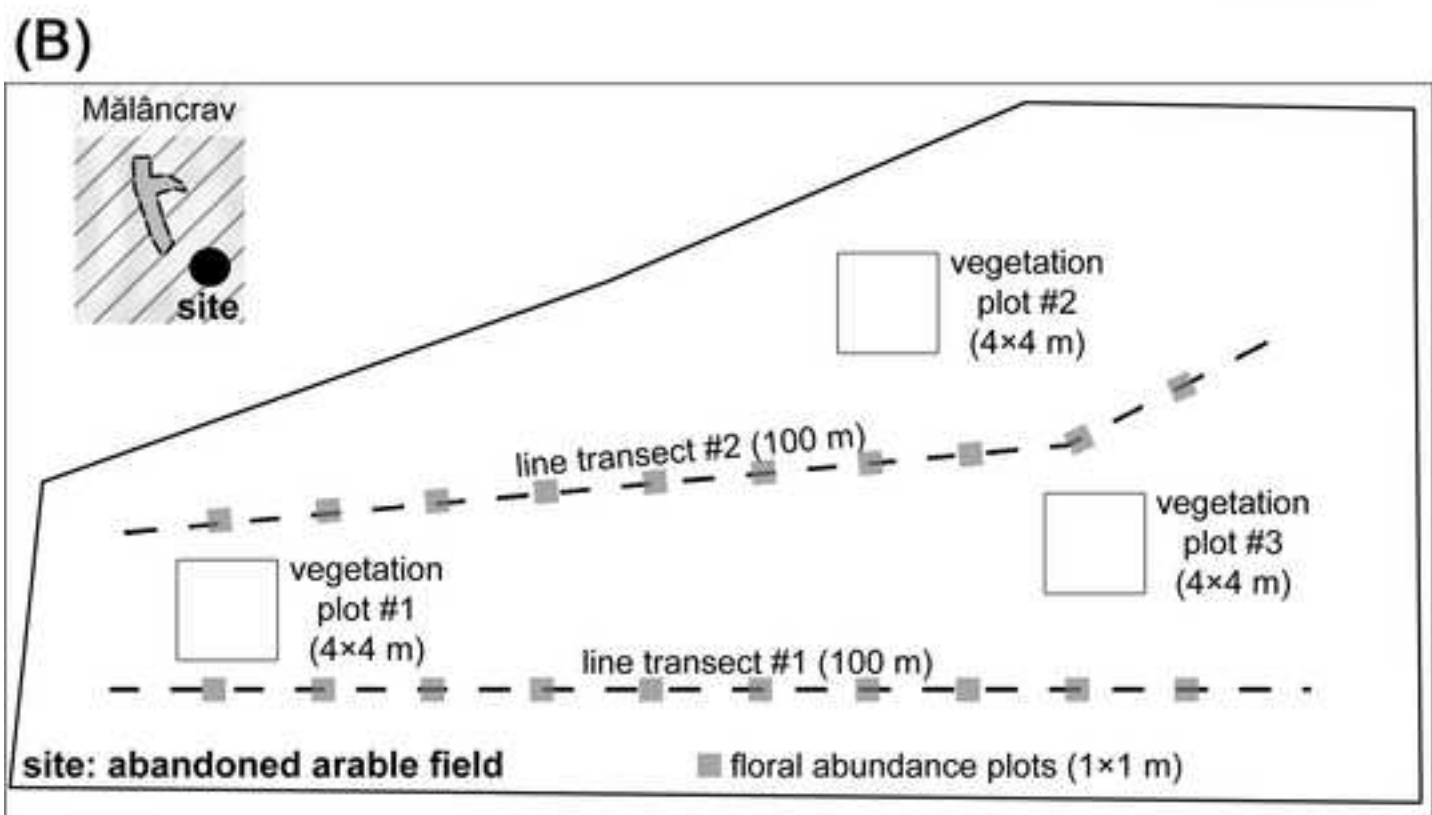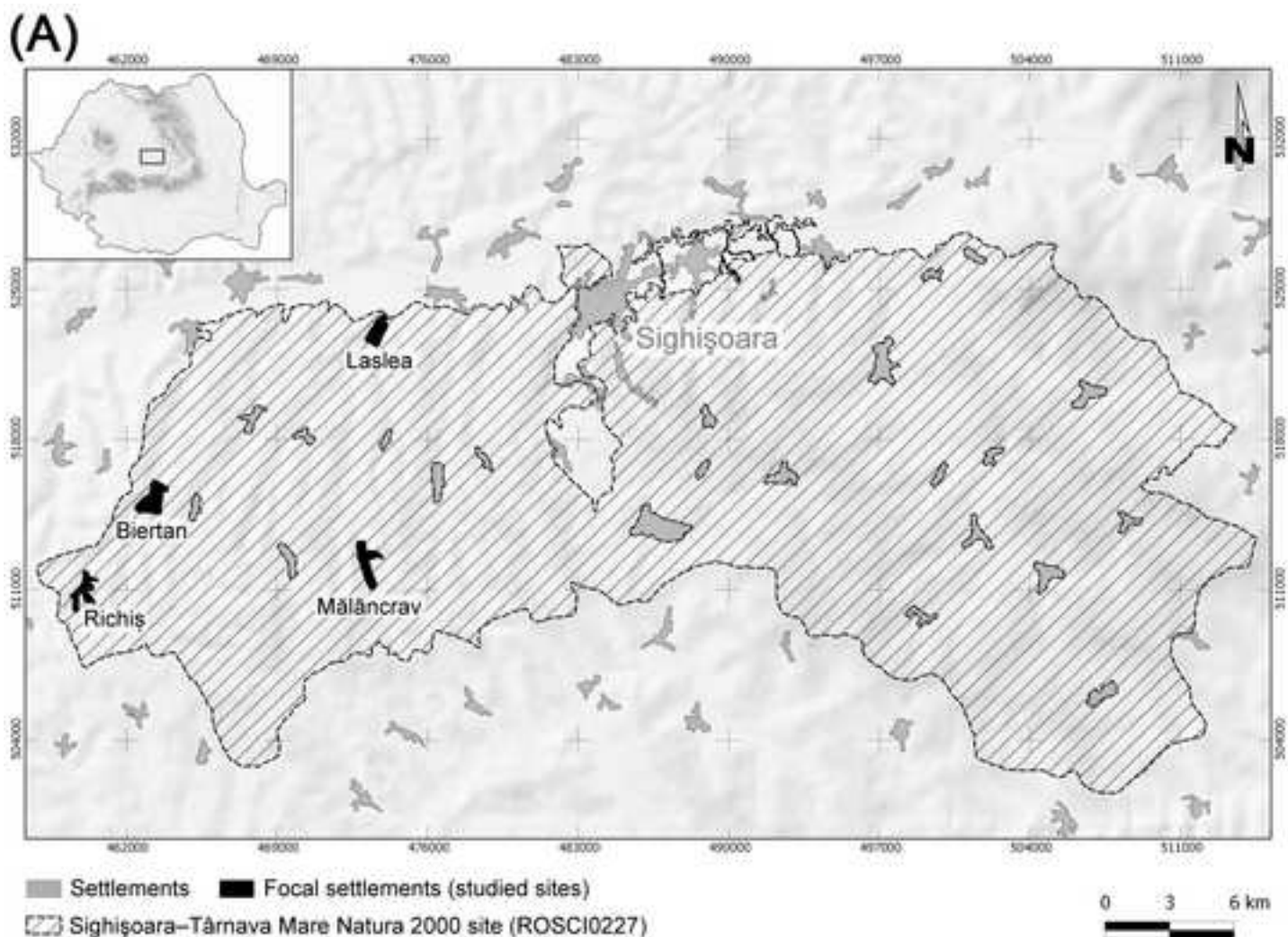
547 (B).

548

549 **Fig. 2.** Plant species richness (A), plant diversity (B), the proportion of three plant naturalness

550 groups (grassland species (C), generalist species (D) and ruderal species (E)), and the

551 proportion of three functional guilds (graminoids (F), forbs (G) and legumes (H)) plotted

552 against the increasing proportional cover of *Solidago canadensis* and old field age.
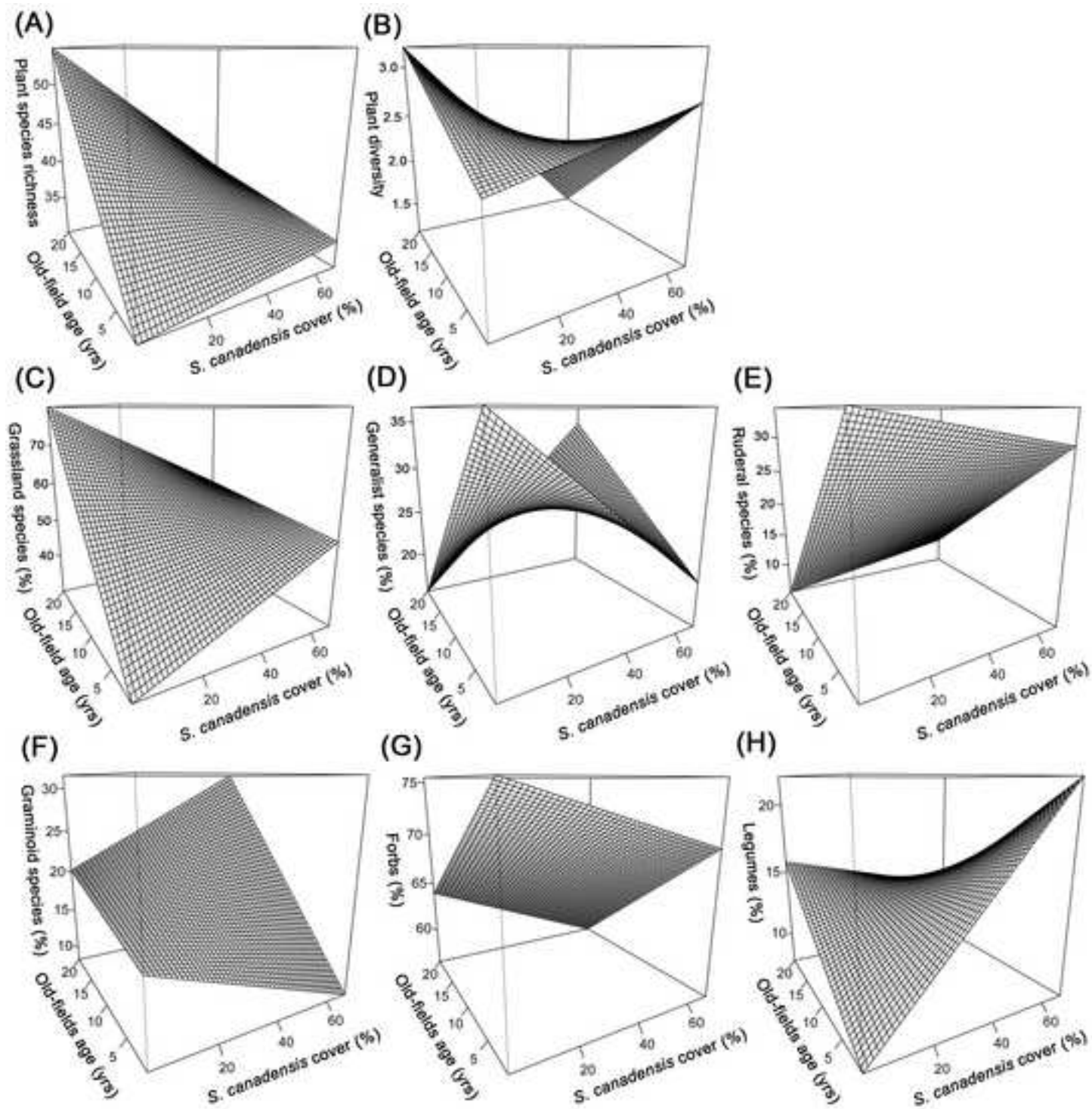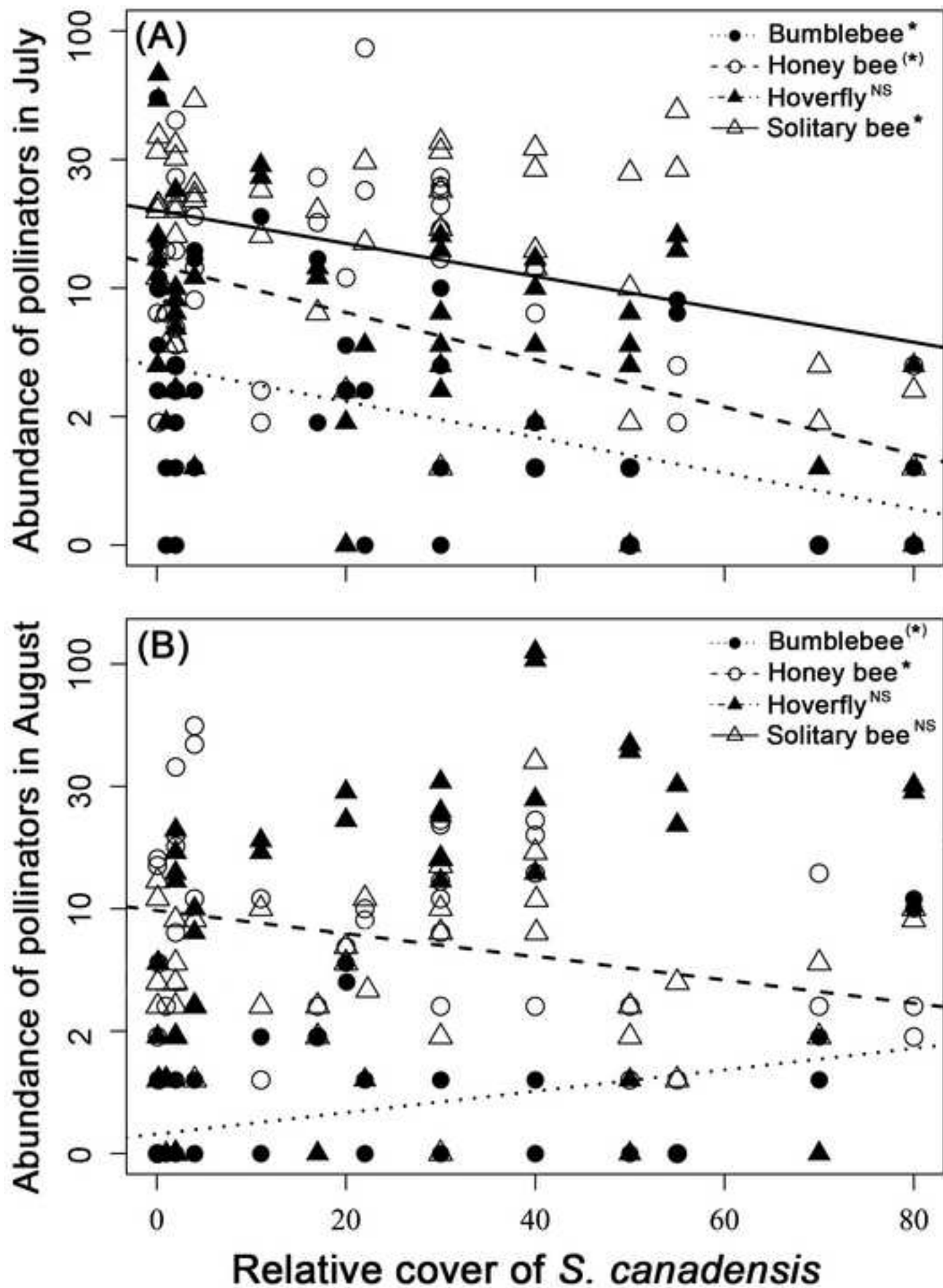
553

554 **Fig. 3.** Abundance of four pollinator groups (honey bee, bumblebees, solitary bees,

555 hoverflies) plotted against the increasing proportional cover of *S. canadensis* before its

556 flowering in July (A) and during its mass blooming in August (B). Note that y-axis has log-

557 scale. Statistically significant and marginally significant results are marked with * and [(*)] and

558 with regression lines; NS – non-significant regressions.
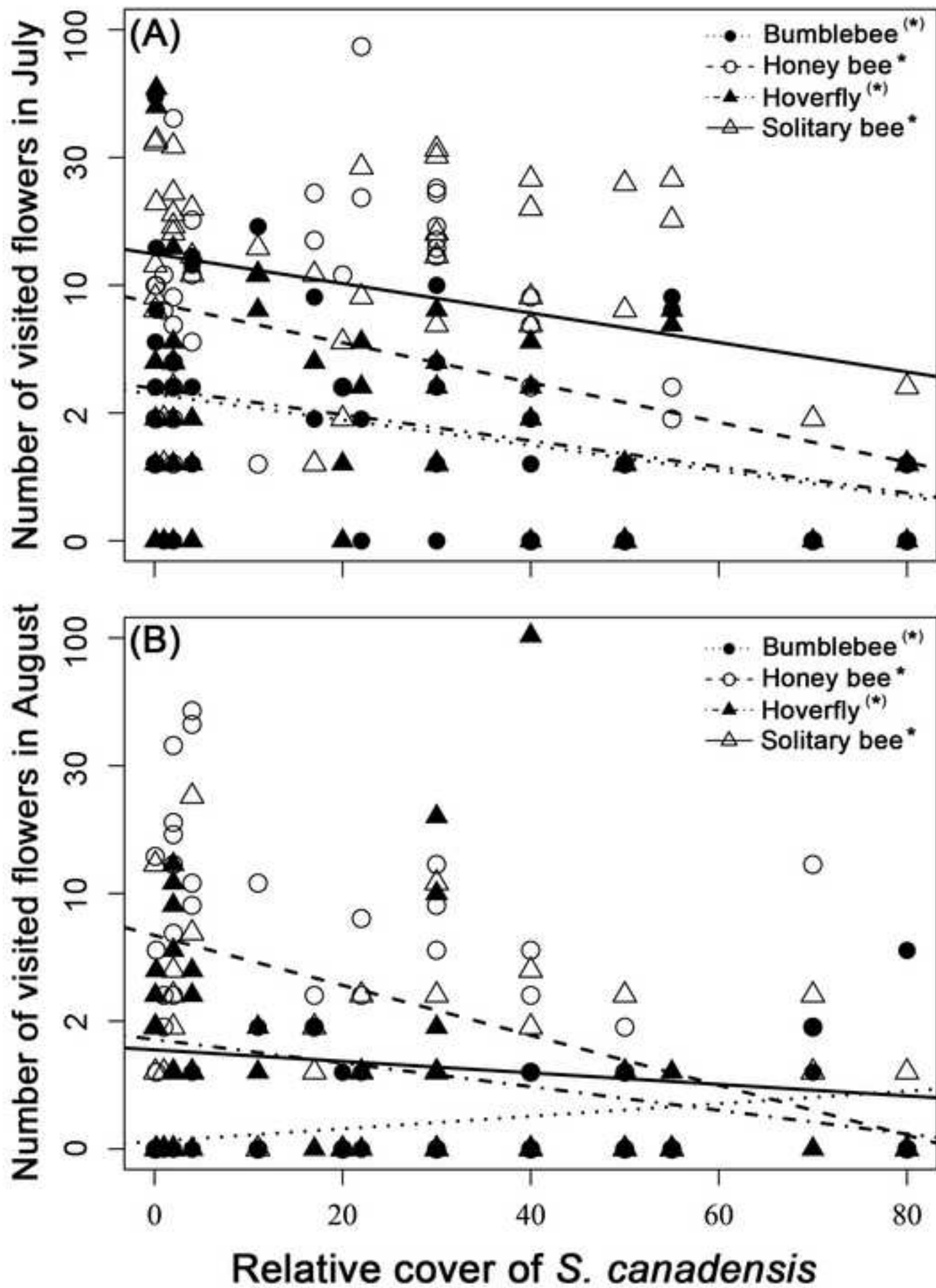
559

560 **Fig. 4.** Number of visited flowers except *Solidago canadensis* plotted against the increasing

561 proportional cover of *S. canadensis* in July (A) and in August (B). Note that y-axis has log-

562 scale. Statistically significant and marginally significant results are marked with * and [(*)] and

563 with regression lines; NS – non-significant regressions.

(A)

Settlements | Focal settlements (studied sites)

Sighişoara–Târnava Mare Natura 2000 site (ROSCI0227)

0 3 6 km

(B)

Mălâncrav

site

vegetation plot #2 (4×4 m)

line transect #2 (100 m)

vegetation plot #3 (4×4 m)

vegetation plot #1 (4×4 m)

line transect #1 (100 m)

site: abandoned arable field   floral abundance plots (1×1 m)

Relative cover of *S. canadensis*

Zusammenfassung

Die sekundäre Sukzession auf ehemaligen Feldern könnte durch die Besiedelung mit invasiven fremden Arten verändert werden, die möglicherweise die ganze Gemeinschaft beeinflussen, indem sie die Entwicklung hin zu artenreichen Gemeinschaften erschweren. Indessen wurden die Einflüsse von invasiven Arten auf lokale Gemeinschaften selten in Hinblick auf die sekundäre Sukzession betrachtet. Wir untersuchten deshalb den Einfluss der hoch-invasiven Kanadischen Goldrute (*Solidago canadensis*) auf Pflanzen- und Bestäubergemeinschaften entlang eines Gradienten unterschiedlich starker Invasion auf alten Feldern unterschiedlichen Alters (1 bis 20 Jahre seit dem letzten Pflügen) in Süd-Transsilvanien (Rumänien). Wir fragten, ob Invasion durch die Goldrute Veränderungen in der Zusammensetzung und Diversität der Pflanzen- und Bestäubergemeinschaften entlang des Sukzessionsgradienten hervorruft. Desweiteren fragten wir, inwieweit die Anwesenheit der Goldrute den Blütenbesuch an einheimischen Pflanzen beeinflusst. Die Invasion reduzierte den Artenreichtum der einheimischen Pflanzen über die gesamte Sukzession hinweg, aber der stärkste negative Effekt auf die Pflanzendiversität und die Natürlichkeit der Vegetation erfolgte in den älteren Sukzessionsgemeinschaften. Die Goldruten-Invasion hatte unabhängig vom Alter der Felder einen negativen Effekt auf die Abundanz der Bienen, aber es gab keinen gleichartigen Effekt auf die Schwebfliegen. Einheimische Pflanzen erfuhren einen reduzierten Blütenbesuch durch Wildbienen, Honigbienen und Schwebfliegen, der der vermehrten Anwesenheit der Goldrute geschuldet war. Deshalb ändert die Invasion durch diese ausdauernde

Pflanzenart den Verlauf der Vegetationssukzession, ändert die mutualistischen Verbindungen zwischen den einheimischen Arten der ehemaligen Felder und verursacht die Etablierung eines unterwünschten, alternativen stabilen Zustandes.