Long-term effect of mowing on the restoration of Pannonian sand grassland to replace invasive black locust plantation

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Invasive species are among the main threats to grassland biodiversity, and nature conservation management seeks the best methods to eliminate them and to restore natural habitats. We studied the long-term effect of mowing on the restoration of Pannonian sand grassland after elimination of invasive black locust (Robinia pseudo-acacia) plantation in Hungary, Europe. Stands of R. pseudo-acacia at three sites were felled and stumps herbicide-treated in the winter of 1994–1995. Mowing with hay removal treatment was applied twice a year in 1995–2001 to assist grassland recovery. A block of 12 adjacent plots of 10 m by 10 m was assigned for the experiment at each site, with six control (unmowed) and six treatment (mowed) plots randomly selected. Vegetation was sampled in June and August yearly in 1995–1999 in all sites, plus in seminatural reference grasslands then re-sampled six times until 2017 in two sites. Herbicide application with repeated mowing successfully eliminated R. pseudo-acacia. In the unmowed plots, dense woody cover developed at all sites. Vegetation of mowed plots approached the reference grasslands in sites with better propagule availability of target species according to trajectory analyses. In these sites, higher cover of target species was found in mowed compared to unmowed plots, though still significantly lower than in the reference. Mowed plots were more prone to secondary invasion than the unmowed. The long-term monitoring revealed that initial mowing assisted the restoration of Pannonian sand grassland, but further management is needed to control secondary invasion and increase target species cover.

Key words: black locust, grassland restoration, invasive species, long-term monitoring, mowing, shrub encroachment

Implications for practice

- The eradication of re-sprouting and N-fixing invasive species, like Robinia pseudo-acacia is a challenge, but it is possible in dry grasslands by herbicide application combined with repeated mowing.
- Initial mowing with hay removal is suitable for controlling shrub expansion and assisting sand grassland recovery on previous R. pseudo-acacia plantation in the long term. However, initial mowing is insufficient to control other alien species and to entirely restore invaded grasslands, probably due to legacy effects of R. pseudo-acacia.
- Integrated management should be implemented to fully restore sand grassland on previous R. pseudo-acacia plantations to overcome legacy effects, including dispersal, biotic, and abiotic limitations.
- Long-term monitoring is essential in evaluating restoration success correctly, because short-term monitoring can lead to misleading results.

Introduction

Biological invasion is considered one of the major drivers leading to biodiversity loss (Sala et al. 2000) and also to be a threat to the recovery of natural vegetation (Sztár et al. 2008; Buisson et al. 2019; Török et al. 2019). Invasive species can alter community structure and function, and ecosystem processes, such as primary productivity, decomposition, hydrology, nutrient cycling, and disturbance regimes (Vitousek et al. 1997). These alterations can negatively affect the development of native vegetation, facilitate invasion by the driver itself or other species (Von Holle et al. 2013; Yelenik & D’Antonio 2013), and threaten ecosystem health and integrity (Maron & Jefferies 2001; Corbin & D’Antonio 2012). Apart from alteration of ecosystem functions, invasive species have a strong impact on ecosystem services and human well-being that should be taken into account.
account in decision making and conflict resolution in restoration (Pejchar & Mooney 2009).

Although grassland habitats in general are characterized by intermediate levels of invasion and low invasion risk in the temperate region (Török et al. 2018a), high-intensity land use like afforestation increases this risk (Török et al. 2003). Several widely used forestry trees are among the most dangerous invasive species, that is, 21 of them can be found on the list of “100 of the World’s Worst Invaders” (Brundu & Richardson 2016). Invasive tree plantations can alter the decomposition rate of organic matter, nutrient cycles, nutrient availability, and light availability to such an extent that they negatively impact native species and increase the likelihood that other alien species will invade these habitats (Von Holle et al. 2013; Csceserits et al. 2016; Medvecká et al. 2018). This leads to conflicting interest, because of the positive economic but negative environmental impacts, especially when invasive species escape from plantations and threaten native ecosystems (Brundu & Richardson 2016; Vitková et al. 2017).

Black locust (Robinia pseudo-acacia) is the most planted alien species used for forestry (Brus 2016) and possibly the second most abundant deciduous tree in the world (Rice et al. 2004). At the same time, it is among the 40 most invasive woody angiosperms globally and is considered the most problematic invasive forest tree in Europe (Vitková et al. 2015, 2017). However, only a few countries have policies to confront this problem (Sitzia et al. 2016). Its popularity is due to its timber quality, fast early growth, resistance to diseases, easy propagation, and ecological tolerance, but also because of the additional ecosystem services it provides, such as soil stabilization or honey production (Brundu & Richardson 2016; Vitková et al. 2017; Nicolescu et al. 2018). Besides the traditional uses, black locust has been planted for carbon sequestration and energy production (Brundu & Richardson 2016; Vitková et al. 2017; Nicolescu et al. 2018). Its future expansion is also expected with changing climate (Kleinbauer et al. 2010).

Among the non-forested areas, natural habitats most at risk from R. pseudo-acacia invasion include thermophilous grasslands that are among the most species-rich and endangered habitats in Central Europe (Vitková et al. 2015, 2017). The greatest impact of R. pseudo-acacia is related to N-fixation that contributes to increased soil nitrogen content, increased nitrogen returns in litter fall, and elevated nitrogen mineralization rates, that in turn alter the pools and cycling rates of nitrogen and other elements (Rice et al. 2004; Liao et al. 2008; Malcolm et al. 2008; Corbin & D’Antonio 2012). These changes influence not only the aboveground vegetation resulting in homogenization and the dominance of early successional nitrophilous species, including archeophytes and invasive species (Von Holle et al. 2006; Vitková et al. 2017), but may substantially alter the soil microbial community, building long-term barriers to ecosystem restoration (Corbin & D’Antonio 2012; Nsikani et al. 2018).

Since 1992 the EU LIFE has financed 33 projects to eliminate R. pseudo-acacia mainly from invaded thermophilous habitats (Silva et al. 2014; Vitková et al. 2017). However, there is no efficient and broadly accepted method to overcome the barriers that hinder recovery and restoration after black locust eradication (Vitková et al. 2017; Nsikani et al. 2018). Chemical treatment or mechanical control (e.g. mowing or grazing) can be applied to prevent the re-sprouting, whereas fire suppression is needed to avoid the stimulation of seed germination (Csizsár & Korda 2015; Nsikani et al. 2018). Mowing and hay removal can decrease the amount of N-containing litter and accelerate leaching of nitrogen from the upper soil layers (Tilman & Isbell 2015), and thus enhance the competitive ability of target species adapted to nutrient-poor environments (Török et al. 2014). This often results in a reduction of nitrophilous and also invasive species (Eschen et al. 2007). The presence of well-dispersing invasive species and also the lack of native species at neighboring areas can compromise restoration efforts (Holl & Aide 2011).

Although much is known about black locust and also about its control, little is known about the long-term changes after black locust removal. We carried out a restoration experiment and evaluated the long-term effect of mowing to assist the recovery of the Pannonian sand steppe after the elimination of black locust at three sites located in different landscape matrices. We analyzed the impact of initial 7 years of mowing on vegetation recovery up to 22 years. Our questions were: (1) How does the vegetation composition change with time and due to mowing? (2) Does mowing accelerate the recovery of sand grassland after elimination of black locust compared to unmowed areas?

Methods

Study Area

The present study was carried out in the Kiskun LTER site (46°53’N, 19°24’E) (Kovács-Láng et al. 2008) central Hungary, Pannonian biogeographic region, Europe. The climate in the region is continental with a sub-Mediterranean influence (Csceserits et al. 2011), with an average temperature of 10–11°C and mean annual precipitation between 550 and 600 mm (Sztár et al. 2018). The characteristic soil type is Calcic Arenosol with more than 90% sand and less than 1% humus content (Lellei-Kovács et al. 2011). The natural vegetation is a mosaic of open forest steppe with sparse juniper–poplar woodland scattered in dry sand grassland matrix (Erdős et al. 2018). The open perennial sand grassland (which belongs to Natura 2000 priority habitat 6,260 Pannonic sand steppes) is the most widespread natural herbaceous community of the area. This grassland is dominated by perennial tussock grasses such as Festuca vagintana and Stipa borysthenica (nomenclature follows Király 2009) and the grassland canopy cover is around 40–70% with bare soil and cryptogams covering the remaining surface (Erdős et al. 2018). Over the past two centuries 92% of open sand grasslands of the region have been destroyed (Biró et al. 2013). Fifty percent of this area has been planted by mainly black locust stands (Biró et al. 2013). The present landscape of the Kiskunság region consists of mainly agricultural fields (57%), seminatural grasslands (19%), forest plantations (19%—mostly alien species), and settlements (6%) (Biró et al. 2013).
Sites
Experiments to investigate accelerating the succession of sand steppe on clear-cut black locust plantations started in 1994. Three plantations were chosen as study sites that were mature enough for cutting (approx. 35 years old) and were situated on dune tops with an herb layer indicating dry soil conditions, suggesting that these areas were previously sand grasslands. The three sites reflected differing landscape compositions (Fig. 1). Landscape matrices were defined based on the percentage of remnant seminatural grasslands along the borders of the experimental sites assuming that dispersal probability of target species is linked to the amount of grasslands adjacent to the sites. Bugac stand (1.1 ha, 46°39′53.1″N 19°36′11.7″E) was situated within a closed forest plantation (Robinia-Poplar plantation) with only a few remnant grassland patches within the forested area (no grassland cover at the borders) that basically means no propagule availability of target species. Fülop háza stand (1 ha, 46°52′31.0″N 19°24′37.6″E) was located in a grassland-forest plantation mosaic with 74% grassland along the borders. Izsák stand (0.4 ha, 46°45′23.2″N 19°19′53.3″E) was established in a predominantly open area surrounded by native seminatural grasslands (85% grassland along the borders).

Experimental Design, Treatments, and Monitoring
Black locust stands were clear-cut in the winter of 1994–1995 followed by cut stump herbicide application (Garlon® 4E) to prevent re-sprouting. Chemical application was repeated where necessary in spring 1995. After cutting Robinia pseudo-acacia, a block of 12 adjacent plots of 10 m by 10 m was assigned for the experiment at each site, where six control (unmowed) plots and six treatment (mowed) plots were randomly selected. Mowing with hay removal was applied as treatment to control weed species and shrub establishment and to decrease nitrogen from the soil in order to assist grassland recovery. Treatments were applied twice a year during the growing season (early June and early September) from 1995 to 2001. Mowing was ceased in 2001 due to lack of financial support and at that time it was also affecting some target species development negatively (Török & Lohász 2004).
The vegetation was sampled by three 2 m by 2 m quadrats per plot (n = 18 per treatment and per site). Reference quadrats of the same size were selected in adjacent grasslands to each site to represent the target grassland community (n = 1 for 1995–1998 for all sites and n = 18 from 1999 only in Fülöpháza and Izsák), dominated by F. vaginata, S. borysthenica, and Fumana procumbens. Vegetation monitoring was carried out each year from 1995 to 1999, then the experiment was re-sampled six times (2002, 2003, 2005, 2007, 2009, 2017) in Fülöpháza and Izsák. Bugac was monitored from 1995 to 1999 yearly, after that, re-sampling was not possible due to Robinia encroachment in unmowed plots. The projected canopy cover of all vascular plants was estimated by eye using the Braun-Blanquet scale (Braun-Blanquet 1965) in June and August in each monitoring year. The data were transformed to the percentage scale (Zólyomi 1951) and the maximum cover between June and August of each species per plot per year was used for further analysis.

Data Analysis
Vegetation composition was analyzed for all sites together by principal co-ordinate analysis for cover data using Euclidean distance. The centroids of mowed, unmowed, and reference samples were calculated along the first and second axis in the ordination space for each year and site to draw the restoration trajectories depicting changes in vegetation composition for the study period. The analysis was performed using the package “vegan” (Oksanen et al. 2018).

We also categorized the species first according to their growth forms (herbaceous or woody) and second according to their role in restoration (target, indigenous, or invasive species). The list of woody species was based on the Flora of Hungary (Király 2009). Out of 176 species, 17 species were considered as woody, the rest herbaceous. Target species were selected according to Csecserits et al.’s (2011) classification of characteristic species of sand grasslands in the Kiskunság region. The group of invasive species was based on the list of invasive neophyte species in Hungary (Balogh et al. 2004). The rest of the species, being native, but not showing high fidelity to sand grasslands, were considered indigenous. Fifty-one species were considered as target (only herbaceous species) and 105 as invasive and 20 as invasive and 105 indigenous (for the complete list see Table S1). We selected three indicator groups, woody, target, and invasive species, for which we calculated the relative cover for all plots and treatments for each sampling year.

Linear mixed effects models (LME) were applied to investigate the differences in relative cover (%) of woody, target, and invasive species among the treatments for each site separately using “nlme” package (Pinheiro et al. 2017). Treatment and year
were treated as fixed categorical explanatory variables, while plots were treated as random effects in the models. Treatment included three levels (mowed, unmowed, reference) in case of target species and only two (mowed, unmowed) in case of woody and invasive species, since the cover of the latter groups was mostly zero in reference plots; therefore reference was excluded from the statistical analyses. Year included five levels (1995–1999) for Bugac and 11 levels (1995–1999, 2002, 2003, 2005, 2007, 2009, 2017) for Fülöpháza and Izsák. Cover of woody species were log transformed, cover of target species were arcsine transformed, and cover of invasive species were square root-transformed to meet assumptions of normality and homoscedasticity. In cases where the variance of residuals within analyzed groups (treatment and year) were high (Bugac: target species and invasive species; Fülöpháza: target species and Izsák: woody, target and invasive species) we used varIdent variance structure. The significance of fixed factors was based on Type II Wald chi-squared tests.

Finally, in the case of significant interactions between fixed factors, Wald test was applied as post hoc pairwise test to detect significant differences between the treatments by using the “contrast” package (Kuhn et al. 2016). In case of nonsignificant interactions, fixed factors were analyzed separately by Tukey’s HSD test, using the “multcomp” package (Hothorn et al. 2008). Statistical analyses were performed using R v 3.5.1 (R Core Team 2018). Means and SE reported in figures and in the text are based on untransformed data.

Results

Trajectory for Vegetation Development

In the PCoA for cover data, 16% of variability is explained by the first axis and 11% by the second axis (Fig. 2). Reference plots were aggregated in the ordination space for all three sites (Fig. 2A). Mowed samples were closer to the reference grassland than unmowed along the first axis for all sites (Fig. 2A). In Bugac, the site with limited propagule availability of target species, trajectory of mowed plots moved parallel to the unmowed along the first axis and both moved away from the reference along axis 2 (Fig. 2B). The trajectory of unmowed plots moved away from reference and mowed plots along the first axis in the ordination space for Fülöpháza (Fig. 2C). In Izsák, where availability of grassland were higher in the landscape, mowed plots trajectory moved towards the reference along the second axis in the ordination space, and it almost reached reference, whereas unmowed plots trajectory moved away from reference along axis 2 in the later phase of the experiment (Fig. 2D).

Cover of Woody Species

Significant interaction between Treatment and Year was found for all sites when analyzing woody species (Bugac: $\chi^2 = 51.8777, df = 4, p < .001$; Fülöpháza: $\chi^2 = 235.338, df = 10, p < .001$; Izsák: $\chi^2 = 134.806, df = 10, p < .001$; Table S2). A woody cover developed with time in unmowed areas that became significantly different from the mowed parcels with time in all sites (Fig. 3). In Bugac woody cover reached ca. 20% by 1999 (Fig. 3A) and in that site the woody species with the greatest cover in unmowed plots was Robinia pseudo-acacia (15%) (Fig. S1). We have experienced the highest shrub encroachment in unmowed plots since the beginning of the experiment in Fülöpháza, reaching 61% cover by 2017, 56% of which was native hawthorn (Crataegus monogyna) (Fig. 3B). In Izsák woody cover rarely exceeded 20% (Fig. 3C) and it was mainly due to native Berberis vulgaris and C. monogyna (11 and 10% in 2017, respectively) (for the complete statistical results see Table S3). In Fülöpháza and in Izsák R. pseudo-acacia did not recover.

Figure 3. Relative cover of woody species in mowed and unmowed plots for (A) Bugac (1995–1999); (B) Fülöpháza (1995–2017); (C) Izsák (1995–2017). Within-year significant difference (p < .05) between mowed and unmowed plots based on Wald test is indicated by asterisk. Means and ± SE are reported in figures based on untransformed data.
Cover of Target Species

There was significant interaction between Treatment and Year for Fülöpháza and Izsák and nonsignificant interaction for Bugac when analyzing target species (Bugac: $\chi^2 = 7.3548, df = 8, p = .4989$; Fülöpháza: $\chi^2 = 154.42, df = 20, p < .001$; Izsák: $\chi^2 = 87.460, df = 20, p < .001$; Fig. 4 and Table S2).

There was an overall increase in the cover of target species with Year from 1998 independent of Treatment in Bugac (Fig. 4A; Table S4). As for the Treatment, the cover of target species did not differ significantly between mowed and unmowed plots that had significantly lower cover of target species (<10%) compared to the reference (ca. 50%) (Fig. 4A; Table S5). In the other two sites with higher propagule availability, a higher cover of target species developed with time in mowed plots compared to unmowed. The difference between mowed and unmowed plots became significant after the cessation of mowing as follows: in Fülöpháza from 2002 ($t = -3.08, p = .0022$), and in Izsák from 2003 ($t = -2.38, p = .0176$) (Fig. 4B & C). In 2005 the cover of target species decreased in both sites, and mowed plots became similar to unmowed in Fülöpháza for that year ($t = -1.83, p = .068$). Although the cover of target species reached up to 70–80% in the mowed plots in some years, it was still significantly lower than in the reference (ca. 100%), even 22 years after clear-cutting (for the complete statistical results see Table S6).

Cover of Invasive Species

Significant interaction between Treatment and Year was observed for invasive cover in all sites (Bugac: $\chi^2 = 37.6180$, Table S5).

Figure 4. Relative cover of target species in mowed, unmowed, and reference plots for (A) Bugac (1995–1999); (B) Fülöpháza (1995–2017); (C) Izsák (1995–2017). Between-year significant difference ($p < .05$) is indicated by capital letters based on Tukey’s HSD for Bugac. Within-year significant difference ($p < .05$) between control, treatment, and reference samples based on Wald test is indicated by lowercase letters for Fülöpháza and Izsák. Means and ± SE are reported in figures based on untransformed data.

Figure 5. Relative cover of invasive species in mowed and unmowed plots for (A) Bugac (1995–1999); (B) Fülöpháza (1995–2017); (C) Izsák (1995–2017). Within-year significant difference ($p < .05$) between mowed and unmowed plots based on Wald test is indicated by asterisk. Means and ± SE are reported in figures based on untransformed data.
Mowed plots were more prone to invasion compared to unmowed principally in sites with lower amount of grasslands in the surroundings (Fig. 5). Cover of invasive species in mowed plots reached 49% in Bugac by 1999 (Fig. 5A) and 30% in Fülöpháza by 2017 (Fig. 5B). Conyza canadensis contributed to 33.5% of cover in mowed plots and R. pseudo-acacia to 15% in unmowed plots in Bugac by 1999 (Fig. S1). Asclepias syriaca and C. canadensis were the two invasive species that reached the highest cover (16 and 14%, respectively) in mowed plots and C. canadensis was generally the most abundant in unmowed plots in Fülöpháza. Invasion was also detected in Izsák (Fig. 5C), but it was more intense in the initial years of restoration (mainly due to C. canadensis), and the cover of invasive species diminished in the last three samplings, resulting in less than 10% of cover by 2017. Significant difference between mowed and unmowed plots was found only in 1996 (t = 2.12, p = 0.0344) and 1999 (t = −3.64, p = 0.0003) for this site (Fig. 5C).

Reference plots were barely affected by invasion, apart from the year of 2005, when we found 1.6% cover in Izsák and 13% in Fülöpháza, mainly due to C. canadensis. In the same year, a high invasion level (over 40% cover) was observed in mowed plots in Fülöpháza (Fig. 5B) (for the complete statistical results see Table S7).

**Discussion**

Long-term monitoring revealed that initial mowing accelerates the recovery of seminatural sand grasslands after elimination of black locust plantations at sites with no propagules limitation of target species; however, the reference composition was not fully achieved according to the trajectory analysis. It was also observed that mowing assisted the establishment of target species resulting in a significant difference between mowed and unmowed plots after 7 years of mowing, and it controlled shrub encroachment. At the same time mowed plots were less resistant to alien species invasion than unmowed plots in sites with lower amount of grasslands in the surroundings.

In our study the differences between unmowed and mowed trajectories can be explained primarily by the presence of woody species which developed a dense shrub cover without mowing. The eradication of Robinia pseudo-acacia and restoration of native vegetation is usually considered to be expensive and very risky, because of the high re-sprouting ability of R. pseudo-acacia (Sádlo et al. 2017; Vítková et al. 2017). In our case the application of herbicide followed by 7 years of mowing proved to be successful in the control of mature R. pseudo-acacia stands on sand dune tops (previous grassland habitats). Re-sprouting was the strongest in the site surrounded by forest plantations (Bugac) where R. pseudo-acacia was responsible for woody encroachment. However, regular mowing prevented its recovery. In the other two sites R. pseudo-acacia was successfully controlled by the herbicide treatments. Native shrubs (primarily C. monogyna and B. vulgaris) that were already present in the R. pseudo-acacia plantations prior to the experiment expanded in control plots, but were prevented by mowing. In dry habitats, like in the Pannonian lowland, native xerophilous shrubs can survive under the canopy cover of plantations and spread during the spontaneous succession of these stands (Sádlo et al. 2017). Shrub encroachment is often a threat to seminatural grasslands, but surprisingly, there was no shrub encroachment in mowed plots even 15 years after the mowing had stopped.

Seven years of mowing resulted in higher cover of the target species compared to unmowed plots in sites with better availability of target propagules (Izsák and Fülöpháza), but contrary to our expectations, mowed plots were more susceptible to invasion. Restoration management causes disturbance, e.g. mowing has created establishment windows and invasive species may be the first to colonize after disturbance if present in the landscape (Holl & Aide 2011), an existing constraint in our case (Csecserits et al. 2016). Additionally, some legacies of R. pseudo-acacia might have remained even after its removal. Although total N content of the soil (0–10 cm) decreased in the shorter term (1996–1999) in all sites equally for mowed and control plots (Halassy 2004), in 2019 we have found considerable differences in soil available N between treatments (20.79–52.58 mg/kg for control and 12.17–34.31 mg/kg for treatment (unpublished data).

Grasslands recovery was also dependent on propagule availability of target species in the surrounding landscape. According to Holl and Aide (2011), the surrounding vegetation can influence the succession process of degraded areas, because it serves as an important source of propagules as well as potential disturbances. Where target species are still present in the landscape, it is possible to rely on natural regeneration to restore grasslands (e.g. Prach et al. 2015; Csecserits et al. 2011; Albert et al. 2014). But if propagule limitation exists (e.g. Török et al. 2018b; Halassy et al. 2019), introduction of desired species is essential to accelerate the re-establishment of grassland vegetation (Kiehl et al. 2010; Kövendi-Jakó et al. 2019). In our case, the seed banks were dominated by ruderal species, only scarce occurrence of visibly viable seeds of sand grassland specialists was found (Halassy 2004). Dispersal limitation was evident only at one site surrounded by forest plantations. In the other cases insufficient recovery can be due to the establishment of invasive species that may hinder native species establishment (e.g. Adams & Galatowitsch 2008; Yelenik & D’Antonio 2013), and their control is essential to assist grassland restoration. At the same time, the establishment and impact of invasive species was also linked to the landscape context (Csecserits et al. 2016).

Our results draw attention to the importance of long-term monitoring in restoration management and suggest that short analyses can lead to misinterpretation of the data. Herrick et al. (2006) presented long-term (up to 75 years) studies from the western United States where short-term monitoring of plant community composition predicted restoration failure, yet it was ultimately successful, but the opposite can occur when the predicted success had ultimately failed. Most of our results (differences in target and invasive cover between mowed and unmowed) were significant only 10 years after initiating the experiment and shorter-term monitoring (3–5 years) would have incorrectly predicted the failure of mowing. On the other hand, mowing has favored the establishment of perennial grasses in
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the early stages (2–3 years) of our experiment, but has caused the dieback of target species (*Festuca vaginata*) afterwards (Török & Lóránt 2004). Long-term studies can result in new insight related to invasion, for example, to reveal that the effect of invasive alien species on the ecosystems shifts over time (Yelenik & D’Antonio 2013) or that the community becomes more resistant to invasion with time (Richardson & Pyšek 2006). The latter can explain the decrease in the cover of one invasive species (*C. canadensis*) at our site (Izsák), where we found the highest cover of target species and a similar vegetation composition to reference grasslands developed after 12 years. *C. canadensis* was outcompeted by perennial sand grasses in experimental rainfall manipulations (Mohácsy et al. 2020, manuscript) and this may be the reason for its observed decline in the field.

We conclude that mowing is effective in the first years of Pannonian sand steppe restoration after eliminating black locust plantation, but for complete recovery, other measurements are needed. Multiple treatments, e.g. seedling, invasive control and potentially manipulation of the soil microbiota are required to overcome dispersal, biotic (considering especially the invasive species) and abiotic limitations (Halassy et al. 2016, 2019). Landscape configuration and constraints should be taken into account when selecting the restoration measures. We also argue that long-term monitoring is essential in understanding community assembly processes in restoration, evaluating restoration success and adapting management to achieve restoration objectives.

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LITERATURE CITED

Adams CR, Galatowitsch SM (2008) The transition from invasive species control to native species promotion and its dependence on seed density thresholds. Applied Vegetation Science 11:131–138

Albert ÁJ, Kelemen A, Valkó O, Miglécz T, Cseserits A, Rédei T, Deák B, Tóthmérsék B, Török P (2014) Secondary succession in sandy old-fields: a promising example of spontaneous grassland recovery. Applied Vegetation Science 17:214–224

Balogh L, Dancza I, Király G (2004) A magyarországi neofitokonok időszerű jegyzéke és besorolásuk inváziós szempontból (actual list of neophytes in Hungary and their classification according to invasion success). Pages 61–92. In: Mihály B, Botta-Dukát Z (eds) Összövénymek – Invasive plants. Természetvédelmi Alapítvány Kiadó, Budapest, Hungary

Bíró M, Szi tér K, Horváth F, Bagi I, Molnár Z (2013) Detection of long-term landscape changes and trajectories in a Pannonian sand region: comparing land-cover and habitat-based approaches at two spatial scales. Community Ecology 14:219–230

Braun-Blanquet J (1965) Plant-sociology: the study of plant communities. Hafner, London

Brundu G, Richardson DM (2016) Planted forests and invasive alien trees in Europe: a code for managing existing and future plantations to mitigate the risk of negative impacts from invasions. NeoBiota 30:5–7

Brus R (2016) Current occurrence of non-native tree species in European forest. Presentation to the Joint WG Meeting of COST Action Non-native tree species for European forests—experiences, risks and opportunities (NNEXT), Lisbon, 4 October 2016

Buisson E, Le Stradic S, Silveira FA, Durigan G, Overbeck GE, Fidelis A, et al. (2019) Resilience and restoration of tropical and subtropical grasslands, savannas, and grassy woodlands. Biological Reviews 94:590–609

Corbin JD, D’Antonio CM (2012) Gone but not forgotten? Invasive plants’ legacies on community and ecosystem properties. Invasive Plant Science and Management 5:117–124

Core Team R (2018) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Retrieved from http://www.R-project.org/

Cseserits A, Botta-Dukát Z, Köröl-Dulay G, Lhotsky B, Öndö G, Rédei T, Szi tér K, Halassy M (2016) Tree plantations are hot-spots of plant invasion in a landscape with heterogeneous land-use. Agriculture, Ecosystems & Environment 226:88–98

Cseserits A, Czaicz B, Halassy M, Köröl-Dulay G, Rédei T, Szabó R, Szi tér K, Török K (2011) Regeneration of sandy old-fields in the forest steppe region of Hungary. Plant Biosystems 145:715–729

Csizsár Á, Korda M (2015) Inváziós növényfajok visszavonulásával kapcsolatos kezelési kísérletek összefoglalása (Summary of treatment experiments related to the control of invasive plant species). Pages 201–235. In: Csizsár Á, Korda M (eds) Összövénymek visszavonulásának gyakorlati tapasztalatai (Practical experiences in the control of invasive plants). Rosalia kézikönyvek, 3, Duna–Ipoly Nemzeti Park Igazgatóság, Budapest, Hungary

Erdős L, Köröl-Dulay G, Bátori Z, Kovács B, Németh C, Kiss PJ, Tölyesi C (2018) Habitat heterogeneity as a key to high conservation value in forest-grassland mosaics. Biological Conservation 226:72–80

Eschen R, Mortimer SR, Lawson CS, Edwards AR, Brook AJ, Igual JM, Hedlund K, Schaffner U (2007) Carbon addition alters vegetation composition on ex-arable fields. Journal of Applied Ecology 44:95–104

Halassy M (2004) A nyílt homokpusztagyep regenerációjának és restaurációjának lehetőségei degradált területeken (the regeneration and restoration possibilities of open sand grassland in degraded areas). PhD dissertation. ELTE-TEK, Hungary

Halassy M, Botta-Dukát Z, Cseserits A, Szi tér K, Török K (2019) Trait-based approach confirms the importance of propagule limitation and assembly rules in old-field restoration. Restoration Ecology 27:840–849

Halassy M, Singh AN, Szabó R, Szili-Kovács T, Szi tér K, Török K (2016) The application of a filter-based assembly model to develop best practices for Pannonian sand grassland restoration. Journal of Applied Ecology 53:765–773

Herrick JE, Schuman GE, Rango A (2006) Monitoring ecological processes for restoration projects. Journal for Nature Conservation 14:161–171

Holl KD, Aide TM (2011) When and where to actively restore ecosystems? Forest Ecology and Management 261:1558–1563

Hothorn T, Bretz F, Westfall P (2008) Simultaneous inference in general parametric models. Biometrical Journal 50:346–363

Kiehl K, Kirmmer A, Donath TW, Rasran I, Holzé N (2010) Species introduction in restoration projects – evaluation of different techniques for the establishment of semi-natural grasslands in central and Northwestern Europe. Basic and Applied Ecology 11:285–299

Király G (ed) (2009) Új Magyar Fűveszkövnyv. Magyarország hatáslas növényei (New Herbal Guide to the Hungarian Flora). Ágteleki Nemzeti Park Igazgatóság, Jósvafő, Hungary

Kleinhofer I, Dullinger S, Peterseil J, Essl F (2010) Climate change might drive the invasive tree Robinia pseudoacacia into nature reserve and endangered habitats. Biological Conservation 143:382–390

Kovács-Láng E, Molnár E, Köröl-Dulay G, Barabás S (2008) The KISKUN LTER: long-term ecological research in the Kiskunság, Hungary. Institute of Ecology and Botany of HAS, Vác, Hungary

Kovendy-Jákó A, Halassy M, Cseserits A, Hülber K, Szi tér K, Wrbka T, Török K (2019) Three years of vegetation development worth 30 years of secondary
succession in urban-industrial grassland restoration. Applied Vegetation Science 22:138–149
Kuhn M, Weston S, Wing J, Forester J (2016) The Contrast Package. http://cran.ma.ic.ac.uk/web/packages/contrast/vignettes/contrast.pdf
Lellei-Kovács E, Kovács-Láng E, Botta-Dukát Z, Kalapos T, Emmett B, Beier C (2011) Thresholds and interactive effects of soil moisture on the temperature response of soil respiration. European Journal of Soil Biology 47: 247–255
Liao CZ, Peng R, Luo Y, Zhou X, Wu X, Fang C, Chen J, Li B (2008) Altered ecosystem carbon and nitrogen cycles by plant invasion: a meta-analysis. The New Phytologist 177:706–714
Malcolm GM, Bush DS, Rice SK (2008) Soil nitrogen conditions approach pre-invasion levels following restoration of nitrogen-fixing black locust (Robinia pseudoacacia) stands in a pine-oak ecosystem. Restoration Ecology 16: 70–78
Maron JL, Jefferies RL (2001) Restoring enriched grasslands: effect of mowing on species richness, productivity, and nitrogen retention. Ecological Applications 11:1088–1100
Medveckj I, Jarolímek I, Hegedűsőv K, Škodavá I, Bazlavová D, Botková K, Šibiková M (2018) Forest habitat invasions—who with whom, where and why. Forest Ecology and Management 409:468–478
Mojzes A, Öndö G, Lhotský B, Kalapos T, Körö-Dulay G (2020) Experimental drought enhances the growth and reproductive performance of an invasive annual weed (manuscript sent in Oecologia)
Nicolevcs VN, Hernea C, Bákvti B, Kereser Z, Antal B, Réde K (2018) Black locust (Robinia pseudoacacia L.) as a multi-purpose tree species in Hungary and Romania: a review. Journal of Forestry Research 29: 1449–1463
Niskani MM, van Wilgen BW, Gaettner M (2018) Barriers to ecosystem restoration presented by soil legacy effects of invasive alien N2-fixing woody species: implications for ecological restoration. Restoration Ecology 26: 235–244
Oksanen J, Blanchet FG, Friendly M, Kindt R, Legendre P, McGlinn D, Minchin PR, O’hara RB, Simpson GL, Solymos P, Stevens MHH, Szoecs E, Wagner H (2016) Vegan: Community Ecology Package. R package version 2.5-1. Retrieved from https://CRAN.R-project.org/package=vegan
Pejchar L, Mooney HA (2009) Invasive species, ecosystem services and human well-being. Trends in Ecology & Evolution 24:497–504
Pinheiro J, Bates D, DebRoy S, Sarkar D, van Willigen B, Kuhn M, Weston S, Wing J, Forester J (2016) The Contrast Package. http://cran.ma.ic.ac.uk/web/packages/contrast/vignettes/contrast.pdf
Scholz M, Dzialowski M, Zabry R, Tóth M, Brabec J, Vítková M, Tonika J, Müllerová J (2015) Black locust—successful invader of a wide range of soil conditions. Science of the Total Environment 505: 315–328
Vitousek PM, Díaz S, Rejmanek M, Westbrooks R (1997) Introduced species: a significant component of human-caused global change. New Zealand Journal of Ecology 21:1–16
Von Holle B, Joseph KA, Largay EF, Lohnes RG (2006) Facilitations between the introduced nitrogen-fixing tree, Robinia pseudoacacia, and nonnative plant species in the glacial outwash upland ecosystem of Cape Cod, MA. Biodiversity Conservation 15:2197–2215
Von Holle B, Neill C, Largay EF, Budreski KA, Ozimec B, Clark SA, Lee K (2013) Ecosystem legacy of the introduced N2-fixing tree Robinia pseudoacacia in a coastal forest. Oecologia 172:915–924
Yelenik SG, D’Antonio CM (2013) Self-reinforcing impacts of plant invasions change over time. Nature 503:517–520
Zólyomi B (1951) Növényföldrajzi-térképezési tanfolyam jegyzete. Soó R, Zólyomi B (eds) Növényföldrajzi-térképezési tanfolyam jegyzete. Vácrátót, Hungary

Supporting Information
The following information may be found in the online version of this article:

Table S1. Species found in the experimental sites with categorization to plant growth form and restoration species groups.

Figure S1. Relative cover of Robinia pseudo-acacia in mowed and unmowed plots for Bugac (1995-1999).

Table S2. Results of Type II Wald chi-square ($\chi^2$) test of fixed effects (Treatment and Year) from linear mixed effects models (LME) for the three different sites (Bugac, Fülföpáza and Izsák) according to different indicator groups (woody, target and invasive species cover).
Table S3. Results of Wald test applied as post hoc pairwise test to detect within year significant differences between Treatments (mowed and unmowed) in three different sites (Bugac, Fülöpháza and Izsák) for woody species cover. Table S4. Results of Tukey HSD applied as post hoc pairwise test to detect significant differences between years (1995-1999) in Bugac for target species cover.

Table S5. Results of Tukey HSD applied as post hoc pairwise test to detect significant differences between Treatments (mowed, unmowed and reference) in Bugac for target species cover.

Table S6. Results of Wald test applied as post hoc pairwise test to detect within year significant differences between Treatments (mowed, unmowed and reference) in Fülöpháza and Izsák for target species cover.

Table S7. Results of Wald test applied as post hoc pairwise test to detect within year significant differences between Treatments (mowed and unmowed) in three different sites (Bugac, Fülöpháza and Izsák) for invasive species cover.