

THE IMPACT OF TIME OF GAP ORIGIN ON MICROSITE CONDITIONS AND SEEDLING RECRUITMENT IN *MOLINIETUM CAERULEAE* MEADOWS

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Abstract

The effect of the timing of artificial gap formation on micro environmental conditions and seedling recruitment was studied in Molinietum caeruleae patches dominated by small meadow taxa (patch I), prevailed by tall-growing macro forbs and large-tussock grasses (patch II) and dominated by the willow-shrubs and bordered by trees (patch III). In all patches the micro environmental conditions (i.e. light availability at ground level, as well as soil moisture and temperature) differed significantly between spring, summer and autumn gaps. The number of species and seedlings observed in spring and summer gaps were quite similar and much greater than those observed in the autumn. In all patches, there were distinguished taxa occurring mostly in spring gaps, taxa colonizing chiefly summer gaps, taxa recruiting mostly in autumn gaps, as well as taxa appearing equally in all types of gaps. In light of the performed investigations, it might be concluded that gap creating during the entire growing season enables the recruitment of plants with different germination requirements and finally contributes to sustained diversity of species in Molinietum caeruleae meadows.

Keywords: Germination; Light availability; Soil moisture; Soil temperature; Spontaneous colonization

Introduction

The cessation of traditional low-intensity land use is a general trend affecting all semi-natural habitats of Europe nowadays. The gradual changes following land abandonment are recognized as a major threat to biodiversity [1]. Therefore, the disturbances created by non biotic (such as windstorms, floods, fires, landslides), as well as biotic factors (i.e. burrowing, wallowing, trampling, scraping by animals) contribute to the formation of openings in continuous plant cover and litter layer providing “safe place for seedling recruitment” [2]. So far, the spontaneous seedling recruitment in openings was observed in wide spectrum of habitats: from alluvial meadows, through grasslands and pastures, to headlands. The colonization process was investigated in relation to different gap origins [3-11], different sizes of openings [12-17], diverse timing of gap formation [18-20], as well as different shapes of the gaps [15].

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Recently, the number of research projects into spontaneous colonization of openings in *Molinietum caeruleae* meadows has increased. Many authors have focused on species richness and seedling abundance in gaps created as a result of disturbances in litter, plant canopy or top layer of soil [21-23]. In addition to that, the impacts of the opening origin mode on the appearance of clonal plants with various life-history traits [24], as well as the effects of opening size on recruitment of small- and large-seeded species [25] were investigated. Moreover, Fibich et al. [26] studied space-temporal dynamics of establishment of generative and vegetative offspring in artificial gaps. In spite of the growing interest in the processes of seedling recruitment in *Molinietum caeruleae* meadows, the process of colonization of openings still remain poorly explored. Therefore, further studies may substantially broaden the knowledge of the mechanisms and consequences of colonization processes and may have promising implications for the restoration of meadows designated for conservation by International Conventions [27, 28], and those included in the NATURA 2000 network [29].

To address the shortage of present knowledge, the effects of the timing of gap formation in overgrowing *Molinietum caeruleae* meadows on (1) the micro site conditions, (2) the total number of taxa and seedlings, as well as (3) the abundance of seedlings of particular taxa were studied.

Material and methods

Study area

The studies were carried out in the Kostrze district, on the western border of Krakow south of the Vistula River (southern Poland – Fig. 1).

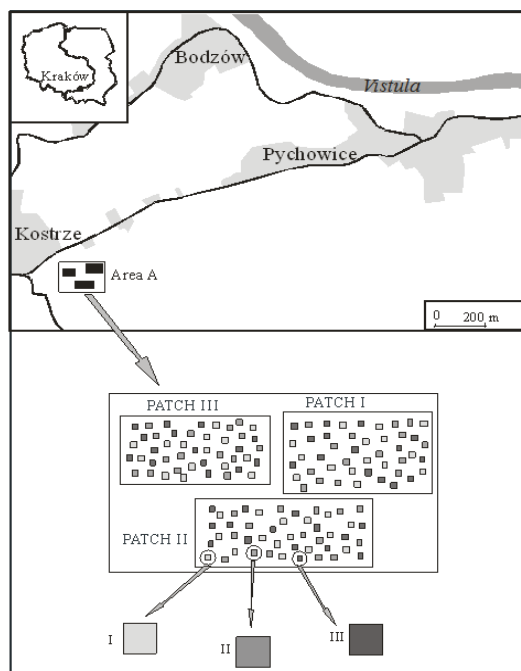


Fig. 1. The localization of patches of *Molinietum caeruleae* dominated by small meadow species (PATCH I), with large tussocks grasses and macroforbs (PATCH II), as well as overgrown by willows prevailing, and bordered by trees (PATCH III) and experimental design: I- spring gaps, II- summer gaps, III- autumn gaps.

The research area is at ca. 210m above sea level, on the low flood terrace of the Vistula, with the height of 3.0-6.0m. The water table is 0.2m below the ground surface. The soils in the

Vistula River valley consist mostly of black earth and light clay and are covered by *Molinietum caeruleae* patches [30]. The abandonment of traditional land use for at least the past dozen years has favored the encroachment of *Phragmites* swamps and willow brushwood, leading to the fragmentation of meadows [31, 32]. In the vicinity of meadows, numerous plant communities have formed, from the deciduous forests covering the slopes of the Vistula valley, through the exothermic calcareous grasslands occurring on limestone and chalk hill slopes, to the ruderal communities appearing near buildings and along the edges of roads.

Vegetation studies

The studies were carried out at study area consisting of three adjacent abandoned patches of *Molinietum caeruleae* with various species compositions. The patch I was dominated by meadow species creating delicate, procumbent stems or small-tussocks. Such plants capture a very small amount of light, contributing to the strong insolation of the ground surface and accelerating the evaporation of water from the soil area. The patch II was dominated by tall-growing macro forbs and large-tussock grasses. The high, closely packed shoots partly shade the soil surface and prevent it from rapid desiccation. Moreover, it should be added that in described site the groundwater table is usually elevated from late autumn to mid-spring. The patch III was dominated by willow-shrubs and bordered by trees. The wide leaf canopy of willows strongly reduces the amount of solar radiation reaching the soil area. Furthermore, the prolonged elevation of the water level in local depressions until late spring or even early summer ensures considerable hydration of the soil. In 2012, the average height of vascular plants was evaluated on the basis of measurements of 30 randomly chosen stems of different species, performed using a folding tape measure. Site conditions in individual patches are described in Table 1.

Table 1. The site conditions in patches of *Molinietum caeruleae* dominated by small meadow species (Patch I), with large tussocks grasses and macro forbs (Patch II), as well as overgrown by willows prevailing, and bordered by trees (Patch III) at area A in the year 2012.

Patch	I	II	III
The patch area (m²)			
The number of species in patch	61	53	44
The dominants (species, with cover exceeding 20%)	<i>Lathyrus pratensis</i> <i>Lotus corniculatus</i>	<i>Molinia caerulea</i> , <i>Deschampsia caespitosa</i> , <i>Lythrum salicaria</i>	<i>Salix repens</i> ssp. <i>rosmarinifolia</i>
The subdominants (species, with cover level ranging 5-20%)	<i>Lychnis flos-cuculi</i> , <i>Succisa pratensis</i>	<i>Serratula tinctoria</i> , <i>Filipendula ulmaria</i>	<i>Betula pendula</i> , <i>Salix aurita</i>
The mean vascular plant height (cm) ¹	46.7	104.3	117.0

The value of statistical significance of differences (the H Kruskal-Wallis test, $df=2$) among patches in vascular plant height achieved 42.95 ($p<0.001$).

Gap creation

In each patch, 3 series consisting of 15 square-shaped openings dimensioned 40 cm x 40 cm were arranged. The first series was set up on April 15, 2012 (spring gaps), the second one on July 1 (summer gaps), whilst the third one on September 15 (autumn gaps). In each opening, the aboveground plant biomass and litter layer was removed because according to Kostrakiewicz-Gieralt [24] such treatment is considered optimal for seedling establishment in the *Molinietum caeruleae* meadows. Moreover, it should be added that gaps were situated *ca* 2.0m from the border of patch to avoid edge effect, as well as they were separated from each other by at least 3.0m wide belts of intact vegetation to ensure sample independency.

Studies of microsite conditions

Site conditions in the central part of each opening were monitored once a week from April 19 till June 30 (spring gaps), from July 4 till September 14 (summer gaps) and from September 19 till November 30 (autumn gaps). All measurements were taken between 10.00 and 12.00 a.m. The light intensity at the soil level was examined with a digital light meter Voltcraft MS-1300 (accuracy +/- 5% + 10 digits; measuring range 0.01-50 000 lx). The humidity at the ground level was measured using a handheld digital soil moisture sensor OMEGA HSM50 (accuracy $\pm 5\%$ + 5 digits; measuring range 0% to 50% moisture content on soil). The temperature of soil at the depth of 5 cm was measured using an electronic temperature sensor HANNA (accuracy +/- 0.1°C; measuring range -10°C - +60°C).

Studies of gap colonization

The process of gap colonization was monitored twice a week from April 19 till June 30 (spring gaps), from July 4 till September 14 (summer gaps) and from September 19 till November 30 (autumn gaps). The seedlings and saplings were removed and examined according to Csapodý [33] and Muller [34] with the support of an own comparative collection. The nomenclature of taxa follows Mirek et al. [35]. After identification, the genets of rare and protected plants were replanted and marked with plastic rings and sticks.

Statistical analysis

The normal distribution of the untransformed data was studied by means of the Kolmogorov-Smirnov one-sample test at the significance level of $p < 0.05$. Subsequently, the variance homogeneity was tested with the Brown-Forsythe test at the significance level of $p < 0.05$. As the distribution of characteristics in some groups of data was not consistent with the normal distribution, and the variance was not homogeneous, the statistical analysis was based on the nonparametric tests. The H Kruskal-Wallis test was applied to establish if there had been significant differences in:

- a. the height of vascular plants at every study site,
- b. the light intensity at the soil surface in spring, summer and autumn gaps at every study site,
- c. the ground moisture in spring, summer and autumn gaps at every study site,
- d. the soil temperature in spring, summer and autumn gaps at every study site,
- e. the total number of taxa/seedlings appearing in openings originated in various time at every study site,
- f. the abundance of seedlings of particular taxa appearing in openings originated in various time at every study site.

After a numerous tests, post-hoc comparisons were made. All analyses were computed using *STATISTICA* software (version 10).

Results and Discussions

Irrespective of patch character, the greatest light intensity was found in spring gaps, lower light availability was recorded in summer openings, while the lowest light intensity was recorded in autumn openings (Table 2).

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Table 2. The mean light intensity level, soil moisture and soil temperature in spring (SP), summer (SU) and autumn (AU) gaps in patches of *Molinietum caeruleae* dominated by small meadow species (Patch I), with large tussocks grasses and macro forbs (Patch II), as well as overgrown by willows prevailing, and bordered by trees (Patch III) at area A in the year 2012.

Environmental parameter	Patch	Spring gaps	Summer gaps	Autumn gaps	The statistical significance level (the H Kruskal-Wallis test; $df = 2$)	The post hoc comparison
Light Intensity	I	47 385 N=150	44 349 N=150	41 899 N=150	174.93 ($P < 0.001$)	a***, b***, c*
	II	42 846 N=150	41 139 N=150	37 485 N=150	90.17 ($P < 0.001$)	a***, b***, c***
	III	38 028 N=150	37 295 N=150	33 482 N=150	34.35 ($P < 0.001$)	a ^{ns} , b***, c***
Soil Moisture	I	21.7 N=150	15.2 N=150	18.0 N=150	50.46 ($P < 0.001$)	a***, b**, c**
	II	29.3 N=150	19.8 N=150	23.2 N=150	35.47 ($P < 0.001$)	a*, b***, c**
	III	31.5 N=150	22.2 N=150	26.3 N=150	173.35 ($P < 0.001$)	a***, b***, c***
Soil temperature	I	11.8 N=150	17.9 N=150	8.3 N=150	200.0 ($P < 0.001$)	a***, b***, c***
	II	10.8 N=150	16.8 N=150	7.6 N=150	207.3 ($P < 0.001$)	a***, b***, c***
	III	10.1 N=150	15.5 N=150	6.9 N=150	203.3 ($P < 0.001$)	a***, b***, c***

The symbols mean the statistical significance of differences between spring and summer gaps (a), spring and autumn gaps (b) summer and autumn gaps (c) * $P \leq 0.05$; ** $P \leq 0.01$; *** $P \leq 0.00$, ns-not significant

Such phenomenon might be due to growing shading as a result of gradual increase of dimensions of adjacent plants. These results correspond to the findings of Kotowski and van Digglen [36], who argued that in fens the amount of light, which reaches to lower canopy layers, diminishes along with the gradient of standing vegetation height. Moreover, in all study sites, the greatest water content in soil was observed in spring openings, in summer gaps the ground humidity was much lower, while in autumn openings it raised moderately (Table 2). The seasonal changes of soil moisture in gaps reflect the natural fluctuations of groundwater levels in *Molinietum caeruleae* meadows, where high water table recorded in autumn, winter and spring, decreases dramatically in summer [37-39]. Performed observations showed that the moderate temperature noted in spring gaps increased in summer gaps, and finally dropped in autumn openings (Table 2). The changes of mean temperatures noted in gaps which originated at different times might result from seasonal fluctuations of air temperature. The influence of air temperature on soil thermal processes was repeatedly observed in a wide range of natural stands [40-42], as well as in laboratory conditions [43].

The mean abundance rates of taxa and seedlings observed in spring and summer openings were quite similar. The number of species and seedlings noted in autumn openings was significantly lower (Table 3). The greatest recruitment rates observed in spring gaps might be the result of high light availability and sufficient soil moisture. The lower number of taxa and seedlings in summer gaps might be due to shading caused by growing size of the surrounding plants. The lower recruitment rates might also result from the occurrence of soil desiccation cracks appearing in clay soil as a consequence of summer drought. The desiccation cracks act as seed traps when they fall too deep to initiate germination process. Similar phenomenon was observed in alluvial meadows [44]. The lowest recruitment rates in autumn gaps might be due to the shading by adjacent plants and litter accumulation increasing in October and November. The negative interactive role of plant canopy and litter was documented by Xiong et al. [45]. Additionally, it should be stressed that litter by itself might

hamper the seed germination by light interception, retention of water after rain events, creation a barrier to water vapor diffusion, fostering pathogens and attracting predators [46].

Table 3. The average number of species (\pm SD) and seedlings (\pm SD) appeared in spring, summer and autumn gaps in patches of *Molinietum caeruleae* dominated by small meadow species (Patch I), with large tussocks grasses and macro forbs (PatchII), as well as overgrown by willows prevailing, and bordered by trees (Patch III) in the year 2012.

	Patch	Spring gaps	Summer gaps	Autumn gaps	The statistical significance level (the H Kruskal-Wallis test)	The post-hoc comparison
Species	I	25.7 (\pm 3.6)	24.6 (\pm 1.9)	13.6 (\pm 3.2)	30.26 ($P < 0.001$)	a ^{ns} , b ^{***} ,c ^{***}
	II	18.7 (\pm 3.9)	17.7 (\pm 4.2)	11.8 (\pm 3.6)	18.34 ($P < 0.001$)	a ^{ns} , b ^{***} ,c ^{**}
	III	16.3 (\pm 1.9)	14.9 (\pm 3.5)	7.8 (\pm 2.8)	27.64 ($P < 0.001$)	a ^{ns} , b ^{***} ,c ^{**}
Seedlings	I	44.4 (\pm 25.3)	111.6 (\pm 28.4)	57.8 (\pm 13.6)	32.10 ($P < 0.001$)	a ^{ns} , b ^{***} ,c ^{***}
	II	90.1 (\pm 30.7)	76.9 (\pm 23.8)	46.7 (\pm 13.1)	30.10 ($P < 0.001$)	a ^{ns} , b ^{***} ,c ^{***}
	III	52.1 (\pm 14.3)	46.5 (\pm 14.7)	26.7 (\pm 11.0)	28.73 ($P < 0.001$)	a ^{ns} , b ^{***} ,c ^{***}

The asterisks mean the statistical significance of differences between spring and summer gaps (a), spring and autumn gaps (b) summer and autumn gaps (c) * $P \leq 0.05$; ** $P \leq 0.01$; *** $P \leq 0.00$, ns-not significant

Taking into account all patches, it might be assumed that different patterns of seedling recruitment were observed. In all, 28 taxa recruited mostly in spring gaps, 8 taxa appeared chiefly in summer openings, 2 species recruited mostly in autumn gaps, while 11 taxa colonized spring, summer and autumn openings equally (Table 4).

A review of the data published showed that to colonizers of spring gaps belong chiefly taxa which produce long-term viable seeds released in late summer or autumn. The endogenous primary dormancy of diaspores might be broken by cold stratification and mechanical scarification of seed coat, which might occur in winter season (Appendix A). At the same time it is worth to be mentioned that the results achieved in the presented studies support observations of the spring peak of seedling recruitment of numerous species such as *Galium boreale* [47], *Geum rivale* [48], *Ranunculus acer* [49], *Selinum carvifolia* [47], *Serratula tinctoria* [50-52], *Solidago canadensis* [53], as well as *Viola* species [47]. Moreover, it should be added that spring gap colonizers might also appear in summer and autumn openings. Such phenomenon is presumably linked with the ability of seeds for germination in a wide spectrum of temperatures after or without any pretreatment (Appendix B).

Taxa colonizing summer openings exhibit long-term seed viability and ability for germination after and without the pretreatment, but the majority of them demand high temperatures for seedling recruitment (Appendices A, B). Such requirement might be seen as the most common strategies for the avoidance of harmful autumn and winter germination. The obtained results support observations of Stammel et al. [54], who discovered the greatest recruitment of *Succisa pratensis* seedlings in summer mown meadows.

Similarly to my findings, also Hölzel and Otte [47] (2004) documented late germination of *Inula salicina* diaspores. The aforementioned authors argued that such phenomenon might be linked with greater soil moisture attributable to autumn precipitations. Additionally Eckstein and Donath [55] demonstrated greater recruitment of seedlings on humid ground than in dry soil. The greatest number of *Plantago lanceolata* off springs in autumn openings might be due to warm stratification occurring in summer and then seedling recruitment. On the other hand, Poons and van der Toorn [56] documented no seedling emergence in summer and autumn.

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Table 4. The average number of seedlings observed in spring (SP), summer (SU) and autumn (AU) gaps in patches of *Molinietum caeruleae* dominated by small meadow species (Patch I), with large tussocks grasses and macro forbs (Patch II), as well as overgrown by willows prevailing, and bordered by trees (Patch III) in the year 2012. Statistical significance level was tested by H Kruskal-Wallis test ($df = 2$).

Group of species	Species	Patch I			Statistical significance level	Patch II			Statistical significance level	Patch III			Statistical significance level
		SP	SU	AU		SP	SU	AU		SP	SU	AU	
A	<i>Filipendula ulmaria</i>	9.0	7.1	1.6	P< 0.001	4.8	3.1	1.2	P< 0.001	3.1	2.2	0.5	P< 0.01
	<i>Galium boreale</i>	8.1	2.8	1.7	P< 0.001	5.3	3.8	2.8	ns	3.9	1.5	0.0	P< 0.001
	<i>Solidago canadensis</i>	8.0	3.9	0.0	P< 0.001	3.1	1.1	0.7	P≤0.05	2.1	1.8	0.0	P< 0.01
	<i>Serratula tinctoria</i>	7.2	7.1	2.8	P≤0.05	2.9	1.5	1.2	ns	3.3	2.9	1.9	ns
	<i>Sanguisorba officinalis</i>	7.2	5.7	3.5	P≤0.05	6.2	4.2	2.7	P≤0.05	3.5	3.3	2.1	ns
	<i>Achillea millefolium</i>	6.9	4.7	0.0	P< 0.001	3.1	2.6	0.0	P< 0.01	2.6	2.0	0.0	P< 0.001
	<i>Galium verum</i>	6.9	3.7	0.0	P< 0.001	3.1	1.5	0.8	P< 0.01	1.5	1.2	0.7	ns
	<i>Cruciata glabra</i>	5.4	4.6	4.4	P≤0.05	4.9	4.1	4.4	ns	2.4	2.3	1.4	ns
	<i>Potentilla erecta</i>	5.5	4.5	0.9	P< 0.001	2.0	1.9	3.0	ns	1.7	0.7	1.1	P≤0.05
	<i>Geranium palustre</i>	4.3	3.7	2.0	P< 0.001	2.3	2.1	1.6	ns	2.1	2.0	1.5	ns
	<i>Selinum carvifolia</i>	4.3	3.8	1.8	P≤0.05	4.9	4.3	3.3	ns	2.0	1.6	0.9	ns
	<i>Geranium pratense</i>	4.1	2.3	1.9	P≤0.05	2.1	1.3	1.2	ns	-	-	-	-
	<i>Mentha longifolia</i>	4.0	1.9	0.7	P< 0.001	3.0	1.8	1.1	P≤0.05	1.7	0.9	0.4	ns
	<i>Leucanthemum vulgare</i>	3.4	2.0	1.2	P< 0.001	2.1	1.3	0.8	P≤0.05	1.7	1.3	0.7	P< 0.01
	<i>Lysimachia nummularia</i>	3.3	2.2	0.0	P< 0.001	2.5	1.7	0.5	ns	2.5	1.8	0.0	P< 0.001
	<i>Valeriana officinalis</i>	3.0	1.8	0.5	P< 0.001	1.9	1.8	0.0	P< 0.01	1.5	0.9	0.3	P< 0.01
	<i>Geum rivale</i>	3.0	0.7	0.0	P< 0.001	1.7	1.1	1.1	ns	1.1	0.8	0.3	ns
	<i>Lotus corniculatus</i>	2.9	1.6	0.0	P< 0.001	1.9	0.9	0.0	P< 0.001	1.1	0.3	0.1	P≤0.05
	<i>Iris sibirica</i>	2.0	0.0	0.0	P< 0.01	0.9	0.9	0.0	P≤0.05	0.4	0.3	0.0	ns
	<i>Ranunculus acer</i>	1.2	0.4	0.5	ns	0.6	0.0	0.0	P< 0.01	2.3	1.1	0.9	ns
	<i>Filipendula vulgaris</i>	3.6	2.7	2.0	P≤0.05	1.3	1.1	1.0	ns	-	-	-	-
<i>Rumex acetosa</i>	2.1	1.4	0.7	P≤0.05	1.9	1.7	1.7	ns	-	-	-	-	
<i>Poaceae</i>	1.5	1.4	0.7	ns	4.9	0.0	0.0	P≤0.05	-	-	-	-	
<i>Caltha palustris</i>	1.0	0.9	0.6	ns	2.4	1.6	0.0	P< 0.001	-	-	-	-	
<i>Polygonum bistorta</i>	3.5	1.6	2.1	P≤0.05	-	-	-	-	-	-	-	-	
<i>Myosotis palustris</i>	0.9	0.3	0.42	P≤0.05	-	-	-	-	-	-	-	-	
<i>Lathyrus pratensis</i>	0.8	0.1	0.2	P≤0.05	-	-	-	-	-	-	-	-	
<i>Viola sp.</i>	0.3	0.1	0.2	ns	-	-	-	-	-	-	-	-	
B	<i>Betonica officinalis</i>	8.1	8.4	3.5	P≤0.05	3.3	3.8	2.5	ns	1.1	1.2	0.8	ns
	<i>Succisa pratensis</i>	6.1	8.9	7.1	ns	5.7	6.0	0.8	P< 0.001	2.3	2.5	1.7	ns
	<i>Cirsium rivulare</i>	2.4	3.8	4.3	ns	2.1	4.1	3.2	P≤0.05	1.7	3.7	2.9	ns
	<i>Cirsium arvense</i>	3.2	3.3	1.8	ns	2.4	4.0	1.3	ns	1.2	2.9	0.0	P< 0.001
	<i>Lychnis flos-cuculi</i>	0.8	2.1	0.9	P≤0.05	0.7	1.2	0.5	ns	0.5	1.2	0.4	ns
	<i>Dianthus superbus</i>	0.5	1.1	0.4	ns	1.0	1.4	0.0	P≤0.05	0.4	0.7	0.2	ns
	<i>Centaurea jacea</i>	1.1	1.7	0.7	P≤0.05	-	-	-	-	-	-	-	-
<i>Sonchus arvensis</i>	0.3	1.3	0.1	P≤0.05	-	-	-	-	-	-	-	-	
C	<i>Inula salicina</i>	1.9	3.1	3.8	ns	3.0	6.4	6.7	P≤0.05	1.8	3.2	4.5	P≤0.05
	<i>Plantago lanceolata</i>	1.2	0.6	1.3	ns	0.4	0.9	1.9	P≤0.05	0.8	0.7	2.1	P< 0.01
D	<i>Lysimachia vulgaris</i>	2.4	2.1	1.9	ns	1.7	1.6	0.7	ns	1.8	1.5	1.3	ns
	<i>Betula pendula</i>	0.8	1.0	0.9	ns	0.0	2.1	0.0	ns	-	-	-	-
	<i>Hypericum sp.</i>	0.8	0.2	0.1	ns	-	-	-	-	-	-	-	-
	<i>Tussilago farfara</i>	0.7	0.1	0.1	ns	-	-	-	-	-	-	-	-
	<i>Taraxacum sp.</i>	0.4	0.2	0.1	ns	-	-	-	-	-	-	-	-
	<i>Chenopodium album</i>	0.2	0.2	0.1	ns	-	-	-	-	-	-	-	-
	<i>Urtica dioica</i>	0.1	0.3	0.1	ns	-	-	-	-	-	-	-	-
	<i>Lythrum salicaria</i>	-	-	-	-	0.4	0.3	0.3	ns	-	-	-	-
	<i>Salix sp.</i>	-	-	-	-	-	-	-	-	0.6	0.5	0.0	ns
	<i>Epilobium sp.</i>	-	-	-	-	-	-	-	-	0.4	0.3	0.3	ns
	<i>Populus sp.</i>	-	-	-	-	-	-	-	-	0.4	0.3	0.2	ns

The group of species: A- taxa recruit mostly in spring gaps, B- taxa recruit mostly in summer gaps, C- taxa recruit mostly in autumn gaps D- taxa recruit similarly in spring, summer and autumn gaps.

The group of taxa recruited similarly in all types of gaps requires exposure to light (Appendix A). In consequence, seeds might initiate germination process after removal of plant canopy and litter layer regardless of timing of disturbance. Moreover, the ability of above-mentioned group of taxa to germination after chilling or seed coat scarification, as well as without any pretreatment in broad range of temperatures, is likely to augment the possibility of colonization of spring, summer, as well as autumn gaps. On the other hand, contrary to the presented findings, previous studies proved that recruitment of seedlings of *Betula pendula* [57, 58], *Chenopodium album* [59], *Lysimachia vulgaris* [60] and *Tussilago farfara* [61] occurred mostly during spring.

Conclusions

The presented studies show that creating gaps is very effective means of conservation of *Molinietum caeruleae* meadows. It might be concluded that promising results bring creating of gaps in patches dominated by small meadow taxa, dominated by large-tussock grasses and macro forbs, as well as overgrown by shrub-willows and trees. Although the greatest recruitment rates were observed in spring gaps, the best results are brought by removal of plant canopy and litter layer during entire growing season. Such treatment enables recruitment of taxa with various germination requirements and finally contributes to the maintenance of species diversity.

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Appendix A. The period of seed shedding [62], seed longevity [63] and type pretreatment required to initiate process of seed germination of species found in seedling pool*.

A group of species	Species	Period of seed shedding months	Seed in	Seed longevity in years	Pretreatment			
					Stratification		Scarification	Light exposure
					cold	warm		
A	<i>Filipendula ulmaria</i>	VIII-X	-		1, 2			
	<i>Galium boreale</i>	IX-X	< 1		2			
	<i>Solidago canadensis</i>	IX-XII	< 1		12			
	<i>Serratula tinctoria</i>	IX-X	-		11		3	
	<i>Sanguisorba officinalis</i>	IX-III	-		2			
	<i>Achillea millefolium</i>	VI-XII	< 15		1			
	<i>Galium verum</i>	VIII-XI	-					
	<i>Cruciata glabra</i>	IX-XII	-					
	<i>Potentilla erecta</i>	VI-X	< 3		2			
	<i>Geranium palustre</i>	VIII-X	< 1					
	<i>Selinum carvifolia</i>	VI-X	< 15		2, 3			
	<i>Geranium pratense</i>	-	< 1		2			
	<i>Mentha longifolia</i>	IX-XII	-					
	<i>Leucanthemum vulgare</i>	VI-X	< 19		13,			
	<i>Lysimachia nummularia</i>	IX-X	1-5					
	<i>Valeriana officinalis</i>	VIII-IX	-		2, 3			
	<i>Geum rivale</i>	VIII-X	< 1		2			
	<i>Lotus corniculatus</i>	VI-XI	< 25		5, 6		3, 6, 14	
	<i>Iris sibirica</i>	X-X	-		2			
	<i>Ranunculus acer</i>	VI-VII	< 22		3		3	
	<i>Filipendula vulgaris</i>	VIII-X	< 11		1, 2			
	<i>Rumex acetosa</i>	V-XI	< 13					
	<i>Caltha palustris</i>	VI-IX	< 2		2, 3			
<i>Polygonum bistorta</i>	VII-IX	-		2				
<i>Myosotis palustris</i>	VIII-X	< 1						
<i>Lathyrus pratensis</i>	VII-X	< 15				3		
B	<i>Betonica officinalis</i>	VI-XII	< 20		3		3	
	<i>Succisa pratensis</i>	VI-XII	-		1, 3, 11			
	<i>Cirsium rivulare</i>	-	< 1					
	<i>Cirsium arvense</i>	VII-XII	< 12		4, 10		1	
	<i>Lychnis flos-cuculi</i>	-	< 27		8			
	<i>Dianthus superbus</i>	IX-XII	< 19		7			
	<i>Centaurea jacea</i>	-	< 21					
	<i>Sonchus arvensis</i>	VI-XII	< 2		1		1	
C	<i>Imula salicina</i>	X-XII	< 1				3	
	<i>Plantago lanceolata</i>	VI-X	< 20			3		
D	<i>Lysimachia vulgaris</i>	IX-III	< 13		1		1	
	<i>Betula pendula</i>	I-XII	< 3		1, 15		1	
	<i>Tussilago farfara</i>	III-VII	-				1, 16	
	<i>Chenopodium album</i>	VI-XI	< 17		1, 3, 9		1, 17	
	<i>Urtica dioica</i>	VI-IX	< 2		3		1	
	<i>Lythrum salicaria</i>	VIII-XI	< 20		2, 3			

*The numbers refer to following sources given in References: 1- [64]; 2-[65]; 3-[63]; 4- [66]; 5- [67]; 6-[68]; 7-[69]; 8- [70]; 9- [59]; 10- [71]; 11-[54]; 12-[72]; 13- [73]; 14- [74]; 15-[75]; 16- [61], 17- [76]

Appendix B. The temperatures of germination of seeds of species found in seedling pool*, **

The group of species	Species	Temperature of germination (°C)*, **				
		After stratification		After scarification	After light exposure	Without pretreatment
		cold	warm			
A	<i>Filipendula ulmaria</i>	¹ 5-10				² 25/10, 23/9
	<i>Galium boreale</i>	¹ 20				
	<i>Solidago canadensis</i>					¹ 20; ² 25/10, 25/15; ¹² 25-30
	<i>Serratula tinctoria</i>	⁸ 22/12			² 26	¹ 20
	<i>Sanguisorba officinalis</i>	¹ 20				² 21, 26, 23/9,
	<i>Achillea millefolium</i>					¹ 10; ² 10, 15, 16, 20, 25, 26; ¹⁶ 10, 15, 25, 15/25;
	<i>Galium verum</i>					¹ 20; ² 15, 20, 25, 26,
	<i>Cruciata glabra</i>					¹ 20
	<i>Potentilla erecta</i>	¹ 20				² 33/19
	<i>Geranium palustre</i>					
	<i>Selinum carvifolia</i>	¹ 5-12; ² 21/11, 23/9,				² 23/9
	<i>Geranium pratense</i>	¹ 5-12				² 26,
	<i>Mentha longifolia</i>					¹ 20; ² 15, 20,
	<i>Leucanthemum vulgare</i>					¹ 20; ² 16, 21, 26; ¹³ 20/18, 5-10;
	<i>Lysimachia nummularia</i>					
	<i>Valeriana officinalis</i>	¹ 20; ² 20,				² 23/9,
	<i>Geum rivale</i>	¹ 20;				² 15, 20, 25, 25/15,
	<i>Lotus corniculatus</i>	³ 10, 12/10, 15/6			² 15, 20, 21, 25, 26/16;	¹ 20; ³ 10, 15/6; ⁷ 15-30; ¹⁷ 15-30;
	<i>Iris sibirica</i>	¹ 5-12;				
	<i>Ranunculus acer</i>	² 5,			² 5,	
	<i>Filipendula vulgaris</i>	¹ 20;				² 16;
	<i>Rumex acetosa</i>					² 16, 21/11;
	<i>Caltha palustris</i>	¹ 20; ² 23/9;				² 21,
<i>Polygonum bistorta</i>	¹ 5-12;					
<i>Myosotis palustris</i>					¹ 20; ² 11;	
<i>Lathyrus pratensis</i>				² 11, 16, 21;		
B	<i>Betonica officinalis</i>	² 21;		² 21;		² 20, 21, 25/10;
	<i>Succisa pratensis</i>	² 15, 16; ⁸ 22/12				² 20; ⁹ 22/15;
	<i>Cirsium rivulare</i>					
	<i>Cirsium arvense</i>	⁶ 20;				² 23/9, 33/19; ⁶ 30;
	<i>Lychnis flos-cuculi</i>	¹ 20; ⁵ 25/15			² 21, 26, 21/11, 23/9;	
	<i>Dianthus superbus</i>	⁴ 20;				¹ 20; ² 26;
	<i>Centaurea jacea</i>					¹ 20; ² 15, 21, 25/10;
	<i>Sonchus arvensis</i>					² 33/19; ¹⁵ 25-30;
C	<i>Inula salicina</i>			² 15;		
	<i>Plantago lanceolata</i>		² 15, 20, 25			¹ 20; ² 15, 16, 20, 21, 5/10; ¹⁸ 30/20, 20/10;
D	<i>Lysimachia vulgaris</i>					² 21/11,
	<i>Betula pendula</i>					² 26, 25, 30;
	<i>Tussilago farfara</i>				¹⁹ 15-30	² 11, ¹⁹ 15-30
	<i>Chenopodium album</i>	² 16, 26/11, 35/20;				² 15, 16, 20, 26, 31, 21/11, 23/9, 25/10, 31/11, 35/20; ¹⁰ 12-32; ¹¹ 15-35;
	<i>Urtica dioica</i>	² 25/10, 33/19;				
<i>Lythrum salicaria</i>	¹ 20; ² 21/11, 20/10;				² 20, 25/10, 35/20; ¹⁴ 15-20;	

*Information on germination temperatures is in the following format: 21°C- germination at stable temperature 21°C with a 12 hour photoperiod, 21/11°C- germination in fluctuating temperatures (in this case seeds were kept in the light at 21°C for 12 hours , and in the dark at 11°C for 12 hours), 5-12°C- germination between 5°C and 12°C.

**The numbers refer to following sources given in References: 1- [65]; 2- [63], 3-[67]; 4-[69]; 5-[70]; 6-[71]; 7- [68]; 8- [54]; 9-[77]; 10- [78]; 11- [79]; 12- [80]; 13- [73]; 14- [81]; 15-[82]; 16- [83]; 17- [74]; 18- [84]; 19- [85].