EFFECTS OF SITE PREPARATION TIME ON SEED GERMINATION AND SEEDLING SURVIVAL RATE IN MIXED SPECIES CONIFER FOREST STANDS IN GIRESUN, TURKEY

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ABSTRACT

This study was undertaken to investigate the effects of the year of site preparation relative to that of large-scale seed dispersal on seed germination and seedling survival success for natural regeneration of oriental spruce in a mixed forest of oriental spruce (*Picea orientalis*) and Caucasian fir (*Abies nordmanniana*) in the Paşakonagi region of Giresun province, Turkey. Two ecologically similar sites, both 120 years of age with an area of 12 ha (site 20) or 15 ha (site 21) were selected. Seed-tree cutting, site preparation and soil tillage activities were performed in "site 20" in 2014, which was a low seed year, and in "site 21" in 2015, which was an abundant seed year, respectively. Oriental spruce trees at both sites dispersed an average of 363 seeds/m^2 in the abundant seed year of 2015, and 92,840 and 143,704 oriental spruce seedlings per ha germinated from these seeds in 2016 in sites 20 and 21, respectively. Seedling number decreased significantly until May 2018 by 34.04% and 49.82% in "site 20" and 21, respectively ($P<0.05$). The greatest significant decrease took place during the first winter germination occurred. Taller and stockier seedlings occurred at "site 21" where site preparation and soil tillage was carried out in the same year as seed dispersal ($P<0.05$).

KEYWORDS:
Oriental spruce, Caucasian fir, seed dispersal, seedling, germination, survival

INTRODUCTION

Natural forests that are old, unstable and have lost canopy closure can be regenerated by sustainable management of the forests to produce wood for industrial use. Spruce forests are the most challenging in terms of natural regeneration studies in Turkey. A uniform shelterwood management method is generally used to regenerate such forests but the success rate remains low [1, 2]. High precipitation and humidity in the region are important factors for the low success rate of natural regeneration [3, 4]. In addition, dense ground vegetation (which can compete with tree seedlings) and the very slow growth of oriental spruce at the seedling stage are other important factors contributing to the low success rate of natural regeneration.

Oriental spruce (*Picea orientalis* (L.) Link) is a relict forest tree species native to northeast Turkey and the Caucasus. It forms mostly pure stands in mountains above 1,000 m with north-facing slopes. In its native range, the oriental spruce forms mixed stands with Caucasian fir (*Abies nordmanniana* Spach), oriental beech (*Fagus oritentalis* Lipsky) and Scots pine (*Pinus sylvestris* L.) [5, 6, 7]. Annual precipitation exceeds 1,500 mm in the native range of this species, and high humidity is characteristic of its growth environment [3].

The seeds of oriental spruce can mature over a short period of only 6 months. Seeds begin to detach from the cones in September and continue to disperse throughout the winter [8, 9]. The definitive determination of an abundant seed year takes place by July or August and results in delays in site preparation procedures and the cuts for seed-tree cutting which are required prior to seed dispersal. It also leads to underutilization of seeds produced in an abundant seed year. It is considered useful to carry out the scheduled cuts to prevent the interruption of regeneration procedures regardless of whether the season is an abundant seed year. However, there has been no study addressing the impact of cuts and site preparation procedures performed in a low seed year on the regeneration success.

In this study, the effects of site preparation time (conducted in 1 year with abundant and 1 year with less-abundant seed production) on seedling germination and seedling survival success were investigated in a mixed forest of oriental spruce and Caucasian fir, which was planned for regeneration under the uniform shelterwood management method.

TABLE 1
Number and volume of trees per hectare at each study site before and after seed-tree cutting

Species	Management plan data prior to seed-tree cutting		After seed-tree cutting			
			Site 21		Site 20	
	Count	Volume (m^3)	Count	Volume (m^3)	Count	Volume (m^3)
Caucasian fir	77	125,006	30	53,000	31	59,900
Oriental spruce	217	278,648	86	186,830	88	180,160
Total	294	403,654	116	239,830	119	240,060

MATERIALS AND METHODS

Description of the Study Area. The study site was located in the Pasakonagi region of Bulancak district of Giresun province in the Black Sea region of Turkey (41°30′22″ – 41°30′22″ N; 41°30′22″–41°30′22″ E; 1450 m, southerly aspect). The site had forest communities primarily composed of oriental spruce (69%) and Caucasian fir (31%). Sites 20 (12 ha) and 21 (15 ha) in the Pasakonagi Forest Management Office were reserved for regeneration and were selected as the study areas. The whole site had a density of 294 trees per ha, totaling 403,654 m^3 of wood per ha, according to the management plan data of the region (Table 1). On average, 217 trees per ha were oriental spruce (278,648 m^3 per ha) and 77 trees per ha (125,006 m^3 per ha) were Caucasian fir. Precipitation occurred throughout the year with mild winters and cool summers. The high precipitation meant that there was no water deficit or drought at either site.

Seed-Tree Cutting and Site Preparation. Seed-tree cutting, ground vegetation removal and soil tillage were performed on "site 20" in 2014, which was not an abundant seed year for oriental spruce at this site, while seed-tree cutting, ground vegetation removal and soil tillage were carried out prior to seed dispersal in "site 21" in 2015; this was an abundant seed year for oriental spruce at "site 21". Of the total tree stock in sites 20 and 21, 40% and 41%, respectively, were harvested during seed-tree cutting. The number and volume of trees remaining at the site after seed-tree cutting are shown in Table 1. Dense ground vegetation, such as *Rhododendron ponticum, Rubus* sp. *Pteridium* sp. *Ilex colchica* and some types of grasses were found at both sites, and branch and root debris remaining after cutting were removed from the sites by mechanized operations, using a bulldozer. Soil was tilled to a depth of 30-40 cm before seed dispersal in "site 20" in 2014 and in "site 21" in 2015.

Determination of Seed Quantity and Seed Characteristics. Nine rectangular seed traps with dimensions 0.5 m × 0.6 m were installed randomly in each site before seeds detached from the cones in the seed year of 2015. Traps were kept at the sites from September 2015 to March 2016. Seeds in the traps were collected periodically and used to determine the total number of seeds dispersed at the site. The empty/full ratio, 1,000-seed weight and number of seeds per kg of the collected oriental spruce seeds were determined [10]. The number of Caucasian fir seeds was recorded but no characteristic determination was performed since the number of Caucasian fir seeds collected was very low.

Seedling Counts. Nine plots, each 1.5 m × 1.5 m, were established randomly, using wooden sticks, in April 2016 (immediately before germination started) at each of the two study sites to determine germination percentage of oriental spruce seeds, seedling survival percentage and development conditions of seedlings.

Seedlings at both sites were counted in July 2016 and germination percentage was determined. Seedlings were recounted in September 2016, May and September 2017, and May 2018 and the percentage survival rates of seedlings over time were determined. For Caucasian fir, only the number of seedlings that germinated alongside oriental spruce seeds was reported but the data were not used in calculations or assessments.

Diameter and height of oriental spruce seedlings. The diameter and height of each of 100 randomly selected oriental spruce seedlings were measured, to the nearest mm, at each site in May 2018 to determine the effect of site preparation time on the growth of oriental spruce seedlings. The sturdiness index (SI) of each oriental spruce seedling at each site in May 2018 was also calculated [11]:

SI = *height (cm)/diameter (cm)*

Statistical analysis. Student's "t" test was used to analyze the change in the number of oriental spruce seedlings at the sites over time, the degree of decrease in the number of seedlings at the sites across time periods as well as the change in the diameter and height values of seedlings at each site and their sturdiness indices in 2018. The change in the decrease percentage of seedlings (death) across time periods was analyzed using analysis of variance (ANOVA), with a multiple comparison test being used to compare individual treatment means where the ANOVA had been shown to be significant. Comparison of two treatments was carried out using the *t*-test.

RESULTS AND DISCUSSION

Number of Dispersed Seeds and Seed Characteristics. There was an average of 363 oriental spruce seeds/m^2 (361,6667 seeds/ha), 17 Caucasian fir seeds/m^2 (166667/ha) and total of 380 seeds/m^2 (378,3334 seeds/ha) collected by the seed traps installed in sites 20 and 21 in the abundant seed year of 2015 (Table 2).

TABLE 2
Number of seeds (seeds/m^2) collected in 2015

No	Oriental Spruce	Caucasian Fir	Total
1	120	17	137
2	190	13	203
3	440	13	453
4	100	30	130
5	413	40	453
6	363	17	380
7	913	10	923
8	553	13	567
9	163	10	173
Mean	363	17	380

Based on the assessments conducted on the oriental spruce seeds dispersed in 2015, the fullness ratio of seeds was 70.08%, weight per 1,000 seeds was 4.09 g and there were 226,808 seeds per kg. Seed trees that were present in the stand during seed-tree cutting dispersed 15.94 kg seeds/ha.

Seedling germination and survival percentage. Based on data from the survey conducted in May 2016, 92,840 oriental spruce seedlings/ha germinated in “site 20”, where site preparation and seed-tree cutting had been carried out in 2014, compared with 143,704 oriental spruce seedlings/ha which germinated in “site 21”, which underwent seed-tree cutting and site preparation in 2015. The seedlings that could not tolerate the environmental stresses (biotic or abiotic) at the site died over time. Oriental spruce seedling mortality over the 2 years from the first survey conducted in May 2016 to May 2018 was 34.04% and 49.82% in sites 20 and 21, respectively (Fig. 1).

The percentage mortality of oriental spruce seedlings between survey dates are given in Table 4. The greatest significant seedling mortality after the germination in 2016 was observed between September 2016 and May 2017 at both sites, where 22.62% and 38.98% of the seedlings in sites 20 and 21, respectively, did not survive the first winter after germination.

The difference percentage decrease in seedling number between “site 20” and 21 was also significant between September 2016 and May 2017. The percentage decreases during the other intervals between the sites were not significantly different (Table 3).

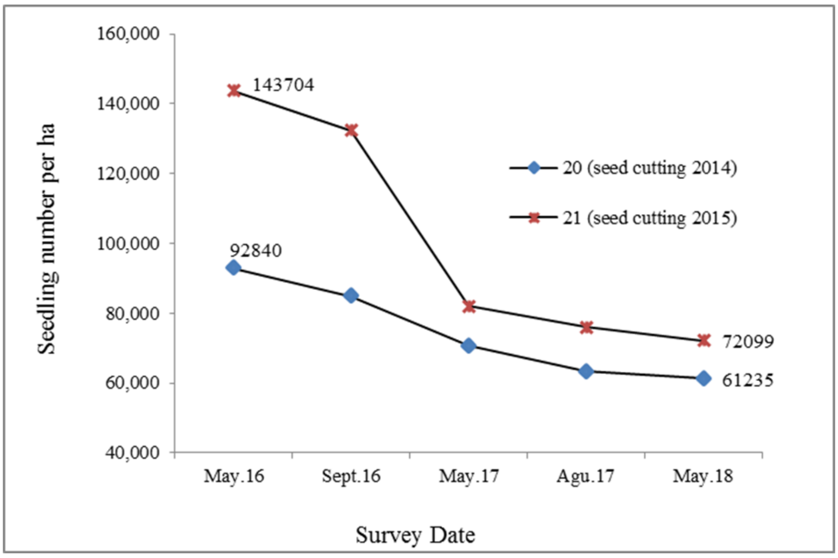

FIGURE 1
Change in the number of oriental spruce seedlings per hectar in two sites through time

TABLE 3
Percentage seedling mortality within survey periods at each site

Period	N	Site 20	Site 21	Significant LSD (P<0.05))
May 2016- September 2016	9	13,72^{ab1}	8,73^{a1}	0,39
September 2016- May 2017	9	22,62^{b1}	38,98^{b2}	0,02
May 2017–September 2017	9	13,05^{ab1}	7,99^{a1}	0,32
September 2017- May 2018	9	3,27^{a1}	4,02^{a1}	0,77
Significant LSD (P value)		0,01	0,00	

TABLE 4
Characteristics of oriental spruce seedlings at the two sites (May 2018)

Characteristic	Site	N	Mean	Significance (P)
Height (cm)	20	100	6.09^{a}	0.00
	21	100	6.79^{b}	
Diameter (mm)	20	100	4.42^{a}	0.18
	21	100	4.54^{a}	
SI (eight/diameter)	20	100	1.38^{a}	0.00
	21	100	1.50^{b}	

Any two dates within a site with a common letter superscript were not significantly different (P>0.05). If two sites at the same date have a common number superscript, the % mortality was not significantly different (P>0.05). The analysis was carried out using ANOVA. The multiple pairwise comparison test used was Fisher's Protected Least Significant Difference (LSD).

Seedling Growth. The oriental spruce seedlings in "site 21" were significantly taller and of higher quality (i.e., stockier) than the ones in "site 20" according to the measurements and assessments conducted in May 2018 (P<0.05). The difference in the seedling diameter between the sites was not significant (Table 4).

In this study, the 86–88 oriental spruce trees remaining per hectare (Table 1), following seed-tree cutting in a mixed stand of oriental spruce and Caucasian fir, produced an average of 363 seeds/m^2 in the abundant seed year of 2015. Atasoy [12] reported that 1,170 seeds/m^2 were collected in an oriental spruce site with a density of 203 trees/ha in the Meryemana forest of Trabzon in the abundant seed year of 1985. The number of seeds collected per tree in that study was greater than the one in the current study when the number of trees at the two sites was taken into account. This difference could be attributed to the differences in the number of trees and in growth conditions. Hofgaard [13] found that Norway spruce with a density of 215 trees/ha produced 475 seeds/m^2 in a study conducted in the Scandes mountains of Sweden in the abundant seed year of 1984. When the number of trees at the sites was taken into account, seed production by Norway spruce [13] seemed to be lower than the seed production of oriental spruce according to the results from both the current study and the ones reported by Atasoy [12]. The lower seed production of Norway spruce might be the result of a more northerly range and colder climate. In a study conducted in the black pine forests of the Dursunbey-Alaçam region of Turkey, Boydak et al. [14] determined that an average of 162 seeds/m^2 were collected in the abundant seed year of 1972 and that 30% of these were empty. In another study conducted to assess seed production in Scots pine, Boydak et al. [15] found that 253 seeds/m^2 and 246 seeds/m^2 were produced in 1971 and 1975, respectively and 26%–28% of these were empty. The emptiness ratio of 29.2% determined in our study was similar to the emptiness ratio values reported for black and Scots pines [15].

The weight of seeds dispersed in the abundant seed year of 2015 at the Paşakonağı sites was 15.94 kg/ha. Boydak [16] reported that 20-30 kg/ha pure seeds would be sufficient for regeneration by seed-tree cutting in Lebanese cedar stands on karstic sites that were regenerated via the uniform shelterwood method. The weight per 1,000 seeds dispersed at the site in 2015 was 4.09 g in the current study. Atasoy [12] measured 1,000-seed weights for oriental spruce seeds collected at different elevations and reported that it was 8.2 g in Meryemana located at 1,600 m, 7.5 g in Bicik at 1,550 m, 9.4 g in Ardanuç at 1,500 m, 9.1 g in Maçka at 1,100 m and 10.4 g in Hamsiköy at 1,450 m. The 1,000-seed weight in the present study was lower than those reported in these other sites.

Three percent of the dispersed seeds germinated in "site 20", which underwent seed-tree cutting before the seed dispersal year, in comparison with 4% of the dispersed seeds germinating in ""site 21"", which underwent seed-tree cutting in the seed year. The remainder of the seeds at both sites either decayed or were consumed by birds, mammals or insects or carried away by the wind. Only 1% of the Norway spruce seeds dispersed germinated in a study carried out by Hofgaard [13], while Grombone-Guaratini and Rodrigues [17] reported that 16% of the seeds collected in a mixed deciduous forest germinated. Similarly, Du et al. [18] determined that only 11% of the collected seeds germinated in *Castanopsis fargesii* stands. The differences between our results (low but not the lowest germination) and other published studies could be attributed to the growth environment conditions, species differences and seed quantity and quality.

The seedlings which germinated from the dispersed seeds following seed-tree cutting were lost to biotic and abiotic stresses (competition with ground vegetation, predation by insects, mammals and birds, and inclement weather) over time. Of the seeds which germinated for the first time in 2016, 34.04% and 49.82% were lost after 2 years in sites 20 and 21, respectively. Erkuloğlu et al. [19] and Ata [1] reported that 70%–80% of the seedlings that germinated in the first year were lost within 5 years in oriental spruce forests, while 53% of the cedar of Lebanon seedlings died within 5 years after germination in naturally regenerated forests. In a regeneration study of *Picea abies*, Hanssen [20] found that only 123 (32%) of 382 seedlings/m^2 that germinated 1 year after the abundant seed year of 1995 survived after five years, as of September 2000. We expect the seedling loss percentage after five years from germination at our site to be around 70%–80% as reported by Erkuloğlu et al. [19] and Ata [1]. Üçler et al. [21] found that there were an average of 16,920 oriental spruce trees/ha in a 14–20-year-old oriental spruce stand that had reached the thicket stage. We predict that the number of seedlings at our site to be similar to this value when the stand reaches that age.

This study proved the importance of timing of site preparation and soil tillage prior to seed-tree cutting on the number of seedlings germinating, seedling survival percentage and the growth of the seedlings. Seedlings which germinated at the site where soil was tilled in the abundant seed year developed better than the ones which germinated at the site which was tilled 1 year before the abundant seed year. Different soil structures and germination conditions have important effects on germination of seeds [22, 23, 24]. For example, phenolic compounds obtained from two different forest soils in Aspromonte (southern Italy) were reported to have significant effects on germination of *Pinus laricio* seeds [25].

However, tillage changes the structure of the soil. Iijima and Shibuya [26], reported higher seedling germination in tilled areas compared to untilled areas in *Picea jezoensis* stands. In a regeneration study conducted in black pine forests, seedling loss was lower in sites where soil was tilled using ripper, hoe and plough compared to reference sites [27]. Likewise, Celma et al. [28] showed that soil tillage and the timing of soil tillage, as in our study, had a significant and positive effect on the development of seedling roots and stems. However, Eşen and Zedaker [29] and Yıldız et al. [30] pointed out that losses of soil carbon and nutrient occurred as a result of soil tillage, in particular during the removal of thick ground vegetation cover like rhododendrons, even though these procedures improved seedling germination and survival success.

CONCLUSIONS

Oriental spruce trees at the study site dispersed an average of 363 seeds/m^2 in the abundant seed year of 2015. From these seeds, 92,840 seedlings/ha germinated in “site 20”, where seed-tree cutting, site preparation and soil tillage were performed in 2014 while 143,704 seedlings/ha germinated in “site 21”, where seed-tree cutting, site preparation and soil tillage were carried out in the abundant seed year of 2015. Of these seedlings, 34.04% in “site 20” and 49.82% in “site 21” had died 2 years after the germination that occurred in 2016. The greatest number of deaths occurred in the winter immediately after germination. Of the seedlings that survived 2 years, those in “site 21” developed better than the ones in “site 20”.

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Sinan Guner and Zuhre Kutlu conceived and designed the experiments; and analysed the data, and Sinan Guner wrote the paper.

The authors declare no conflicts of interest.

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