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Development and anatomical traits of black pine on an abandoned agricultural land compared to forested areas

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Abstract Global acreage of forested lands has increased in some countries. At least some of this increase is due to the natural conversion of abandoned agricultural lands into forests. However, little is known about how these new stands develop on abandoned agricultural lands in comparison with natural regeneration of existing forests. Specifically, knowledge of how black pine (*Pinus nigra* Arnold) naturally establishes and develops on abandoned agricultural lands is limited. In this study, we examined the density and growth of black pine saplings as well as some morphological and anatomical characteristics on an abandoned agricultural land (AAS). These data were compared with those observed in a naturally regenerated stand (NRS), and in a forest opening (FOS). The greatest sapling density was observed in the NRS site, while sapling growth and stem biomass were higher in AAS followed by NRS and FOS. Moreover, each study site exhibited site-specific morphological and anatomical traits in their saplings. Our findings showed that site treatments and overstory openness would both play crucial role for establishment and development of black pine.

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Introduction

Forest degradation has been a serious issue worldwide throughout the history of humankind. Total global acreage of forested lands has decreased due to the exploitation of forested areas for agriculture, energy, transportation, and habitation (Ahmed et al., 2015; Lim et al., 2019; Nathaniel & Bekun, 2019). However, clearance of forested lands for agriculture has at times been followed by agricultural abandonment, and consequently by forest regrowth (Flinn & Vellend, 2005). In addition, reforestation activities have also occurred in many European countries since the nineteenth century (Altunel et al., 2020; Meyfroidt & Lambin, 2011) resulting in increasing acreage of forested lands (Popovic & Cirkovic-Mitrovic, 2016; FAO, 2016; Bebi et al., 2017). Aside from the afforestation practices, socio-economic reasons have also significantly contributed to the increasing forested lands across Europe (Alcantara et al., 2012; Atmış & Günşen, 2016; Kuemmerle et al., 2011); the abandoned agricultural lands have been naturally reverted to forests (Atmış, 2020).

Black pine (*Pinus nigra* Arnold) is one of the most widespread and economically important native tree species across Europe and Asia Minor (Kara &

Topacoglu, 2018). Within its natural distribution areas, land expansion of the species has been occasionally observed both following the afforestation practices as well as the abandonment of agricultural lands in several countries (Bulut, 2018; OGM, 2019; Piermattei et al., 2013). It should be noted that this study does not intend to determine the reasons for the increased forested acreage of black pine within its natural distribution area, because the main question remains as to how these new stands develop, and what the potential is of abandoned agricultural lands to grow forest trees. Instead, the study aims to observe whether development of forest trees, black pine in particular, on abandoned agricultural lands are comparable to the forested lands.

During forest establishment, foresters are usually concerned with controlling density, successful growth, and ensuring development of saplings (Kara et al., 2017). Morphological and cambial activity traits both show environmental adaptability and growth performance of saplings, and are also closely related to growing site conditions (de Luis et al., 2011; Fonti et al., 2013). Site conditions can affect morphological characteristics such as thickness of leaves and bark, stem height and diameter, and aboveground biomass (Auty et al., 2012; Rocha et al., 2019; Will et al., 2010). Furthermore, cambial activity of wood and anatomical traits of tree ring play crucial roles in determining tree performance and adaptations to growing site conditions (de Luis et al., 2011).

Knowledge of how black pine naturally establishes and develops, and how its anatomical characteristics vary on abandoned agricultural lands, is limited. To our knowledge, there has not been any research to examine this within the natural range of black pine. Therefore, in this study, the main objective was to examine and quantify the density and growth of black pine saplings as well as some morphological and anatomical characteristics on an abandoned agricultural land (AAS). Moreover, the study also aimed to compare these data with those observed in a naturally regenerated stand (NRS), and in a forest opening (FOS). We hypothesized that black pine saplings would present higher density and growth on abandoned agricultural land than the forested lands. Morphological and anatomical characteristics may provide better insights to the understanding of growing site effects on tree growth and also help to improve success of forest management. The findings in this study would create a basis for our knowledge on the natural conversion of abandoned agricultural lands into forests.

Materials and methods

Study sites

This study was carried out within the Karadere Forestry Directorate in Kastamonu, northern Turkey (Fig. 1). The study area is located within the Euro-Siberian phytogeographic region and is in the natural range of black pine. Kastamonu region shows the typical characteristics of a continental climate with cold winters and rainy summers. The mean monthly temperature ranges from − 1 °C in January to 20.3 °C in July, with a mean annual temperature of 9.8 °C. The mean total annual precipitation is approximately 490 mm, with the maximum monthly precipitation in May (68.8 mm) and a minimum in February (28.5 mm). Soil is moderately deep (50–90 cm) and primarily brown forest soil in the study area. Topography varies from gently sloping to steep slopes, and elevation ranges from 810 to 1705 m above sea level. The main trees are black pine, Scots pine (*Pinus sylvestris* L.), Oriental beech (*Fagus orientalis* Lipsky.), Trojan fir (*Abies nordmanniana* subsp. *equi-trojani*), hornbeam (*Carpinus* spp.), and oaks (*Quercus* spp.), while common hazel (*Coryllus avellana* L.), raspberry (*Rubus* spp.), and common juniper (*Juniperus communis* var. *saxatalis* Pall.) are the common understory species.

Three study sites were selected and named as AAS, NRS, and FOS for abandoned agricultural land, natural-regenerated site, and forest opening site, respectively. It should be noted that the environmental conditions for each site were similar due to their close proximity. Moreover, similar topography and elevation of the study sites were also crucial to eliminate the effects of topographic and site conditions on growth and aboveground biomass. AAS with an area of about 2 ha was located within Çaltepe Forest Planning Unit (41° 16′ 3″ N, 34° 1′ 48″ E), adjacent to compartment 177 that is occupied with mature black pine trees, at an elevation of 1390 m. Although the region is mostly mountainous, the topography of AAS is nearly flat. AAS was abandoned for more than 10 years, and the site was likely seeded by the

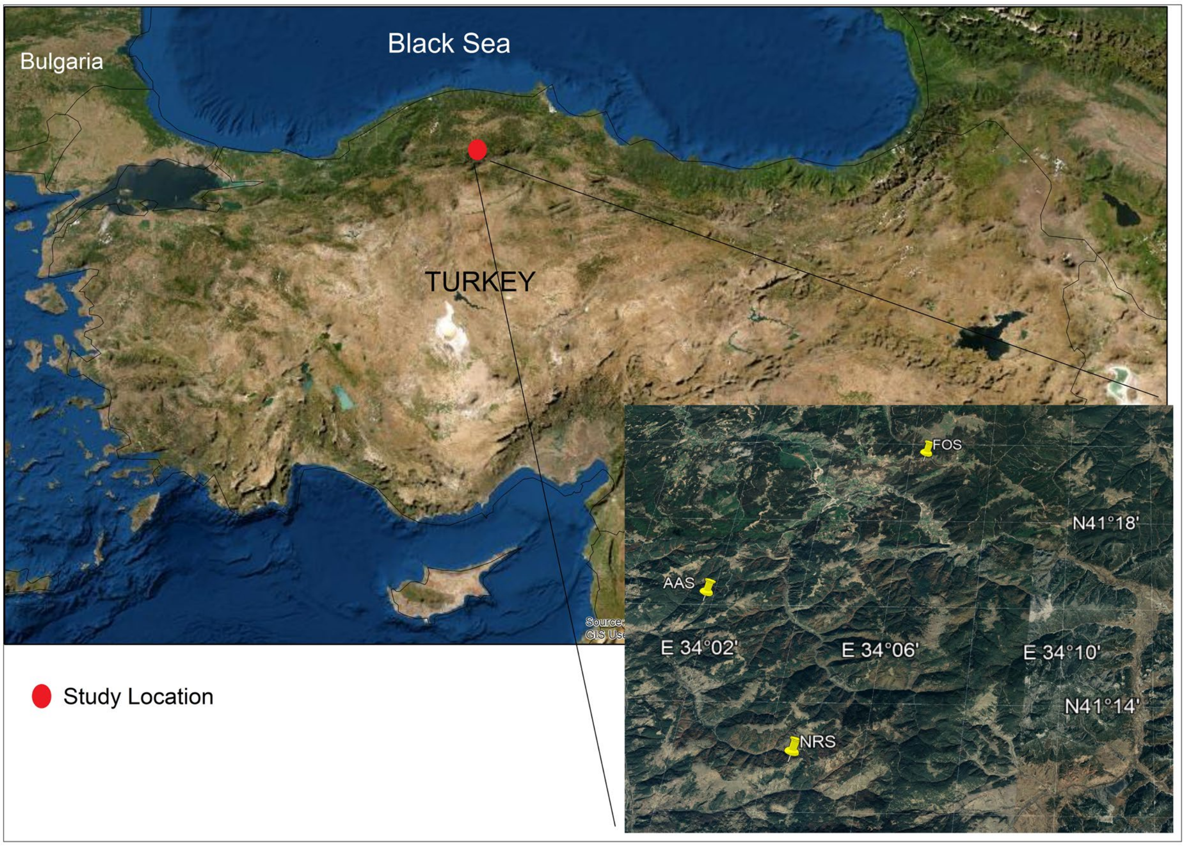


Fig. 1 Location of the study sites in Kastamonu, northern Turkey

neighboring black pine stands and consequently naturally occupied with black pine saplings. Wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.), and rye (*Secale cereale* L.) were the main crops cultivated on AAS before abandonment. In addition, based on communication with locals, the site had been fertilized with manure before abandonment. NRS with an area of about 10 ha was located within Tepe Harman Forest Planning Unit (41° 12′ 52″ N, 34° 4′ 29″ E) in compartments 189 and 190 at an elevation of 1430 m. The topography of NRS is also nearly flat. The site has been managed using a traditional shelter-wood system for decades resulting in an even-aged forest structure. The latest regeneration activity on NRS started with a preparation cut in 2009. Next, mechanical site preparation was conducted the following year using a mini-excavator, and the regeneration was completed with a final removal conducted in 2019. FOS with an area of about 2 ha was located within Akkaya Forest Planning Unit (41° 19′ 32″ N, 34° 6′ 18″ E) in compartment 24 at an elevation of 1350 m. The mean slope of the site is less than 10%. Some mature and old black pine trees were sporadically distributed across the site, approximately 5–6 trees per ha. There has not been any site preparation or any other treatment within FOS for decades.

Density, growth, and aboveground biomass measurements

It should be noted that most of the saplings in the study sites are at the age of ten which was determined by counting tree rings from the harvested saplings that is explained in detail below. This suggests that dominant saplings are from the good seed crop year of 2010 on the study sites. In order to calculate the sapling density of the sites, sixty 25 m^2 square plots were randomly installed across each site in July and August of 2020. Next, saplings were counted within the plots, and converted to the number of saplings per ha on each study site. Root-collar diameters (RCD) (mm) of the saplings were measured to the nearest millimeter using a digital caliber to obtain the RCD of the saplings on each site after 10 years. In addition, the height of the saplings (cm) was measured to the nearest 0.1 cm using a measurement rod to determine the height of the saplings on each site after 10 years.

For aboveground biomass measurements, fifteen saplings were randomly chosen and harvested on each site in July and August of 2020. Since study sites are owned by the government, only a limited number of saplings were allowed to be harvested by the managers. The aboveground biomass measurements followed the methodology outlined by Daryaei et al. (2019). First, three components (i.e., stem, branch, and needle) of all harvested saplings were separated, and the components were measured as fresh on each site. Next, a representative fresh sample of each component was taken from the harvested saplings; they were weighted on the site, and brought to a laboratory. These representative samples were oven-dried at 80 °C for 2 days, and weighted again. Subsequently, the dry biomass of each component after 10 years was calculated using the formula below (Daryaei et al., 2019).

$$\mathrm{TDW} = \mathrm{TFW} + \frac{\mathrm{DWS}}{\mathrm{FWS}}$$

where TDW is the total dry weight of the component (i.e., stem, branch, or needle), TFW is the total fresh weight of the component, DWS is the dry weight of the representative sample of the component, and FWS is the fresh weight of the representative sample of the component.

Morphological, anatomical, and wood density measurements

The stem wood samples from the harvested saplings were used to determine morphological, anatomical (cambial traits and ring widths), and basic wood density (g cm $-^{3}$) traits from each site. Stem diameter (mm) was obtained from outside of the bark using a digital caliper and measured in both the plane and perpendicular to the plane of the stem. Then, the diameter of the pith (mm) was measured using a digital caliper. The bark was removed and the thickness of the bark (mm) was measured. Next, the xylem (woody part) proportion (%) was determined.

Tree ring width (mm) measurements were conducted on the transverse section using Leica LAS EZ Image Analysis Software (Leica Microsystems Ltd., Switzerland). The wood samples were cut into small pieces (approximately 10 × 10 mm in size) transversely to investigate anatomical cell characteristics. The samples were softened in boiling water, and then kept in a mixture of water, glycerol, and ethanol (equal amount) (Yaltirik, 1971). After that, the softened wood segments were cut in cross and tangential sections of the thickness of 20–25 µm using a sliding microtome. To measure tracheid cell characteristics (tracheid length and width), the small wood pieces were cut into 1 × 10 mm strips and macerated in1:1 (v:v, mixture of equal amounts) hydrogen peroxide and concentrated glacial acetic acid solution per Franklin's method (Franklin, 1945). The macerated samples were stained with safranin and placed on glass slides (Bond et al., 2008). Micrographs were obtained using a Leica DM750 photomicroscope (Leica Microsystems Ltd., Switzerland). Tracheid length (TL), tracheid width (TW), tracheid lumen width (TLW), tracheid wall thickness (TWT), ray height (RH), and ray width (RW) were identified using the Leica LAS EZ Image Analysis Software. For each cell anatomical characterization, twenty-five measurements were conducted per sample (i.e., twenty-five tracheid lengths were determined in one sample, then the mean was used) (IAWA, 1989; Yaman, 2007).

The basic wood density of a total of forty-five samples (i.e., 15 for each site) was determined using Archimede's principle. The fresh stem segments were first cut into small pieces, and each sample was kept in water using an airtight container until all samples were fully hydrated. Each hydrated piece was then submerged in water using a needle in a beaker standing on an electronic weighing balance that gave the mass of water displacement. To obtain a constant mass of wood samples, the samples were oven-dried at 103 °C. Finally, the basic wood density was calculated by dividing the oven-dried mass to volume (Barnett & Jeronimidis, 2003).

Analysis

Differences between the study sites in terms of height and RCD, sapling density per ha, and aboveground biomass (i.e., dry stem, branch, and needle biomass) after 10 years, morphological traits (pith%, bark%, and xylem%), anatomical traits (tree ring width, tracheid length and width, tracheid lumen width, tracheid wall thickness, ray height, and ray width), and basic wood density were tested using the analysis of variance (ANOVA) with the mixed-effect statistical model (α-level = 0.05). For sapling density and growth analysis, plots were treated as a random effect nested within the sites, while the harvested saplings were treated as a random effect nested within the sites for aboveground biomass and anatomical-morphological analyses. Multiple comparisons of means of the sites were performed using Tukey's test. The relationship between morphological, anatomical, and basic wood density properties of saplings in three sites was also quantified using linear regression. All statistical analyses were conducted using the "aov," "lm," and "TukeyHSD" functions in the R-Statistical software (Development Core Team, 2010). Normality and homogeneity of variance were also tested through the residual analysis.

Results

Density and growth of saplings

The sampling density, RCD, and heights of the saplings after 10 years were significantly different between the sites ($p < 0.001$). Moreover, significant differences between all pairs of the study sites for sapling density ($p = 0.03$ for AAS-NRS, $p = 0.004$ for AAS-FOS, and $p < 0.001$ for NRS-FOS pairs) were attained (Fig. 2a, b, and c). The differences between AAS and NRS sites were not statistically significant ($p = 0.44$), while FOS was significantly different than AAS ($p < 0.001$) and NRS ($p < 0.001$) with regard to RCD growth after 10 years (Fig. 2b). Differences between each pair for heights of the saplings were also statistically significant ($p = 0.04$ for AAS-NRS, $p < 0.001$ for AAS-FOS and NRS-FOS pairs) (Fig. 2c). The mean sapling height was greater in AAS (Fig. 2c).

Aboveground biomass of saplings

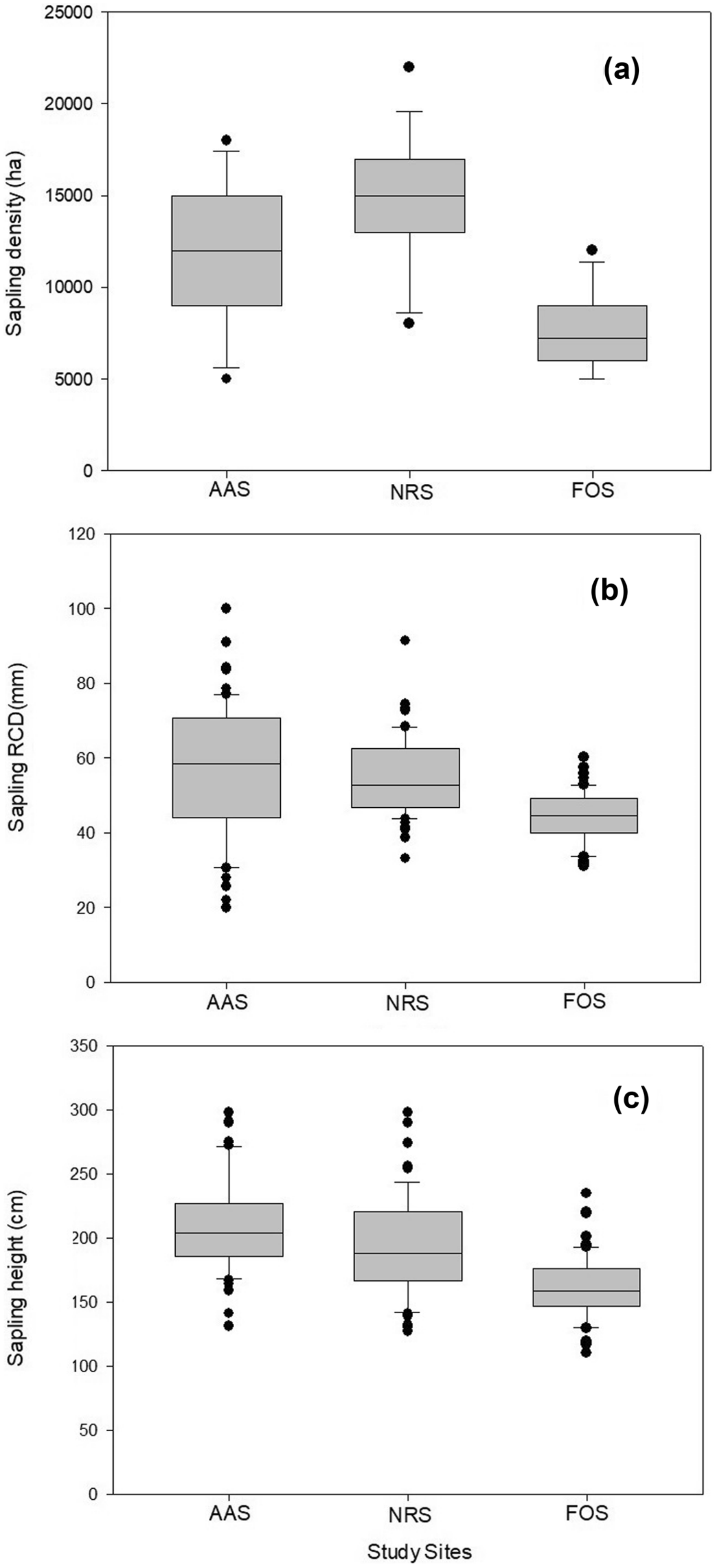
The mean total biomass (i.e., sum of stem, branch, and needle biomass), the mean stem biomass, the mean branch biomass, and the mean needle biomass of harvested saplings were significantly different across the study sites ($p < 0.001$). The differences between AAS and NRS regarding total, stem, and branch biomass were found to be statistically insignificant ($p = 0.274$, $p = 0.16$, and $p = 0.091$ respectively), while FOS was significantly different than AAS ($p < 0.01$) and NRS ($p < 0.001$) (Fig. 3a, b, and c). Moreover, Tukey's test indicated statistically significant differences between all pairs of the study sites for needle biomass ($p = 0.019$ for AAS-NRS, $p < 0.003$ for AAS-FOS, and $p < 0.001$ for NRS-FOS pairs) (Fig. 3d).

Most of the aboveground biomass components were strongly correlated (Table 1). Total biomass had a very strong degree of correlation with stem biomass followed by branch and needle biomass in all study sites. The strongest correlation was between the total biomass and stem biomass in the FOS site. Overall, RCD and height were strongly correlated with total biomass and stem biomass (Table 1). Height was strongly correlated with stem biomass in all study sites. Moreover, RCD and height were also correlated with each other. The correlation between height and aboveground biomass components was stronger than the correlation between RCD and the components (Table 1).

Wood morphological, anatomical, and wood density characteristics

Each study site showed different patterns in their stem woods (Table 2). The study sites significantly affected the mean pith% of the saplings ($p < 0.0001$). The bark% significantly varied across three sites ($p < 0.0001$) with greater bark% in FOS. The mean xylem% was significantly affected by the study sites ($p < 0.001$) with the greatest mean xylem% in NRS (Table 2). AAS and NRS showed almost 2 times greater mean ring widths than FOS ($p = 0.000$) (Table 2). The mean tracheid length (TL) did not differ significantly between the three study sites ($p = 0.538$). The mean tracheid width (TW) values also did not show significant differences between the three

Fig. 2 The means of **a** mean density, **b** mean RCD, and **c** mean height of saplings within the study sites after 10 years. The bars indicate the standard deviations



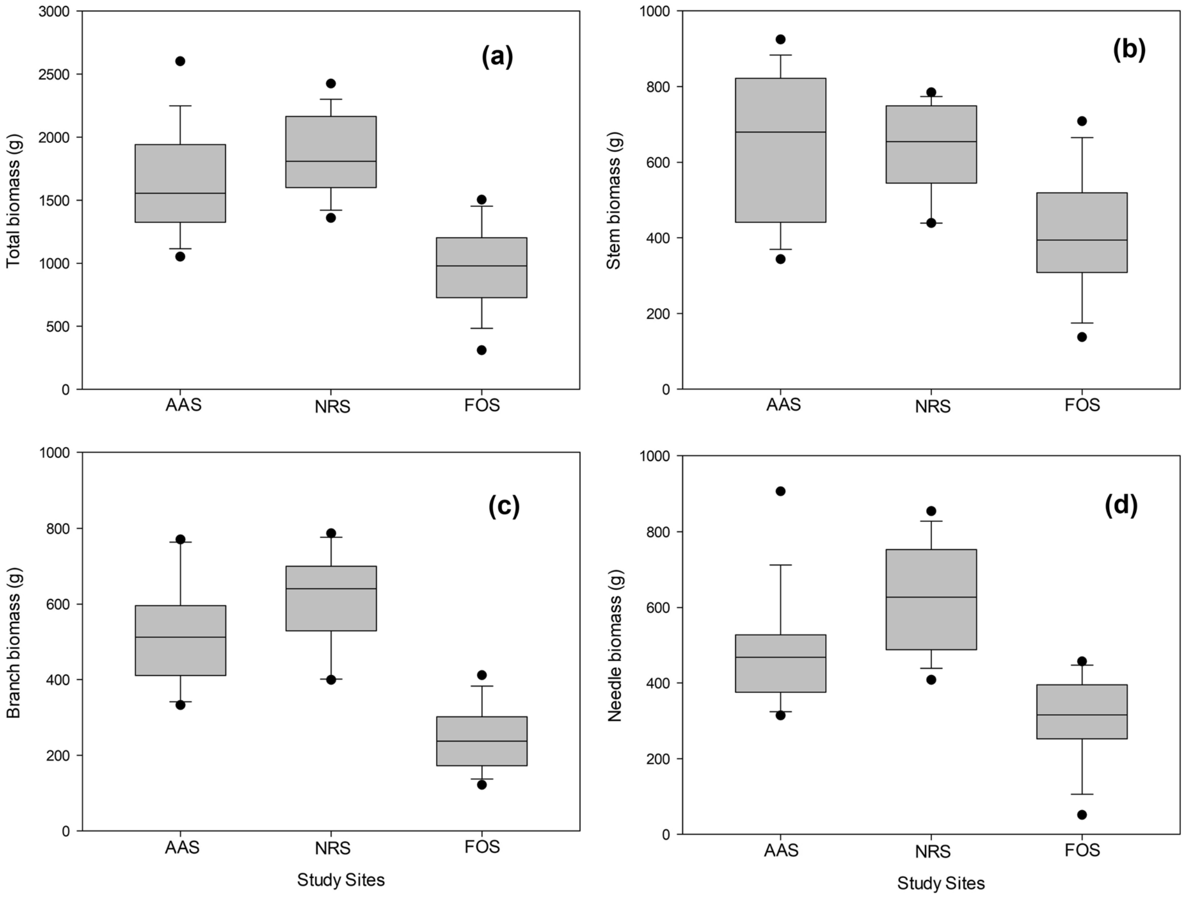


Fig. 3 The means of **a** mean total, **b** mean stem, **c** mean branch, and **d** mean needle biomass of saplings within the study sites after 10 years. The bars indicate the standard deviations

sites ($p = 0.135$) (Table 2). The mean tracheid lumen width (TLW) was greatest in AAS followed by FOS and NRS ($p < 0.0001$). The mean tracheid wall thickness (TWT) of saplings also varied significantly in three study sites ($p = 0.004$). The study site had a significant effect on mean ray height (RH) ($p < 0.0001$); NRS showed greater mean RH than AAS and FOS. The mean RH of saplings was significantly different between each site ($p < 0.0001$). An insignificant difference was obtained in mean ray width (RW) in three study sites ($p = 0.060$). Moreover, no significant difference was found in mean values of wood density between the three study sites ($p = 0.315$).

Discussion

Density and growth of saplings

Our findings indicated that sapling density was the highest in NRS. For a shade-intolerant tree species, it stands to reason that decreased density and the subsequent increase of light entering the stand would positively affect seedling density (Noémie et al., 2011). However, black pine has been observed to survive under parent trees in juvenile stages of development (Odabaşı et al., 2004). Similarly, Köseoğlu and Kara (2019) pointed out that the shelter of overstory trees increased the number of seedlings in black pine

Table 1 Pearson correlation matrix for the AAS, NRS, and FOS study sites regarding total biomass (TB), stem biomass (SB), branch biomass (BB), needle biomass (NB), root-collar diameter (RCD), and height (H) of black pine saplings

Variables	SB	BB	NB	RCD	H
TB					
AAS	0.93***	0.91***	0.78***	0.56*	0.89***
NRS	0.81***	0.85***	0.88***	0.59*	0.56*
FOS	0.97***	0.77***	0.88***	0.57*	0.68**
SB					
AAS	-	0.85***	0.53*	0.58*	0.88***
NRS		0.54*	0.55*	0.63*	0.68**
FOS		0.69**	0.83***	0.50 ns	0.71**
BB					
AAS	-	-	0.55*	0.57*	0.75**
NRS			0.65**	0.49 ns	0.53*
FOS			0.45 ns	0.46 ns	0.50 ns
NB					
AAS	-	-	-	0.32 ns	0.71**
NRS				0.39 ns	0.56*
FOS				0.56*	0.59*
RCD					
AAS	-	-	-	-	0.62*
NRS					0.81***
FOS					0.63*

ns, *,**,*** are $p > 0.05$, $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively

stands in northern Turkey, as was observed in NRS, where overstory trees were present for about ten years before final removal. Moreover, Tiscar and Linares (2014) observed the influence of overstory cover on black pine seedling density, and concluded that black pine regeneration is facilitated by tree shelter. In another study conducted in Spain, Granda et al. (2012) found that the establishment of black pine was higher under a canopy than open-site conditions. This can likely be associated with decreasing soil surface moisture typical of open-grown conditions found in AAS and under low-tree-density conditions in FOS, since deficiencies in soil surface water negatively affect the growth of black pine (Topaçoğlu et al., 2016). High irradiance under open-grown (i.e., AAS) and low-tree-density (i.e., FOS) conditions may reduce water availability and increase evaporative demand, and consequently hinder seedling establishment (Valladares et al., 2005). Moreover, AAS was seeded from the neighboring stands, which may likely be considered as another reason for the lower sapling density in AAS than NRS.

Black pine seeds germinate better on mineral soil (Odabaşı et al., 2004). Thus, the higher sapling density can also be associated with the intensive site preparations conducted in NRS before the seed dispersion. In addition to exposing the mineral soil, site preparation also aims to control the competition of seedlings with other vegetation. Karadağ (1999) examined the impacts of site preparation on black pine seedling density in northwestern Turkey, and found that site preparation significantly increased seedling density. In another study conducted in northwest France, Dassot and Collet (2020) found that mechanical site preparation resulted in better root development of black pine seedlings. Moreover, del Cerro Barja et al. (2009) revealed higher seed germination and survival

Table 2 The mean values of morpho-anatomical traits and basic wood density of saplings in three study sites (AAS, NRS, and FOS)

Morphological, anatomical, and basic wood density traits	AAS	NRS	FOS
Pith%	8.3 ± 0.91***	5.2 ± 0.67	3.07 ± 0.45
Bark%	9.1 ± 0.39	6.8 ± 0.59	10.9 ± 0.56***
Xylem%	82.5 ± 1.03	87.8 ± 1.05***	85.9 ± 0.65
Ring width	2.3 ± 0.16	2.6 ± 0.16***	1.2 ± 0.11
Tracheid length	1025.3 ± 84.6	1238.3 ± 80.8 ^{ns}	1180.9 ± 94.8
Tracheid width	31.2 ± 2.46	32.5 ± 1.97	37.7 ± 2.29 ^{ns}
Tracheid lumen width	15.6 ± 0.93***	10.6 ± 0.42	12.3 ± 0.38
Tracheid wall thickness	3 ± 0.15	3.4 ± 0.14**	2.7 ± 0.09
Ray height	180.4 ± 13.6	246.3 ± 15.9***	148.8 ± 14.5
Ray width	31.3 ± 0.76*	27.9 ± 1.02	27.4 ± 1.44
Basic wood density	0.39 ± 0.011	0.38 ± 0.016	0.43 ± 0.013 ^{ns}

Values indicate the mean ± standard error (SE); $n = 15$ for each study site for morphological and basic wood density parameters. The significance is indicated as *$p < 0.05$, **$p < 0.01$, and ***$p < 0.001$ and ns (i.e., not significant) of black pine following mechanical soil treatment in Spain. Thus, for the same reason, AAS which was subject to cultivation for decades before abandonment may have had greater sapling density than FOS where no soil treatment had been utilized for a prolonged time. Despite some old trees, which would produce seeds, sapling density was lower in FOS due to the absence of site preparation and low tree density. Thus, our findings regarding sapling density coincide with previous research.

Shelter of parent trees positively influences seedling density in black pine stands; however, negative effects of the overstory on black pine seedlings can be observed after 3 years following germination (Odabaşı et al., 2004). Previous research indicated that light availability is one of the most important drivers for black pine seedling growth and biomass (Köseoğlu & Kara, 2019). This may explain the higher (or comparable) RCD and height of saplings as well as aboveground biomass in AAS compared to the NRS site. Eler et al. (1989) monitored the impacts of the timing of overstory removal on seedling height growth in black pine forests in southwestern Turkey, and found that seedling height at age six was 49.9 and 14.8 cm when overstory removal was conducted in the second and fourth years following germination, respectively. In addition, Lucas-Borja et al. (2012) observed the impacts of overstory on the seeding height of black pine, and found that initial seedling growth was negatively affected by the overstory. It should be noted that the FOS site had a relatively higher density of competing vegetation, which likely affected the growth and aboveground biomass of saplings along with the absence of site preparation and sporadic overstory trees. Lucas-Borja et al. (2016) examined the effects of vegetation treatment on the recruitment of black pine in central-eastern Spain, and concluded that competition control enhanced seedling emergence in pure stands.

The higher branch and needle biomass in NRS than AAS and FOS can be associated with the lower light availability in understory prior to overstory removal in NRS. Light demanding species such as black pine can invest more biomass in branches and leaves to raise the leaf surface area for higher light interception under a canopy (Saldaña-Acosta et al., 2009). FOS, which had a relatively higher density of competitive vegetation, had significantly lower biomass. In order to attain a high amount of resources for competing with the competitive vegetation, saplings in FOS may have likely used most of their energy to develop their root system rather than their aboveground biomass. Kara and Topaçoğlu (2018) found that black pine seedlings generate higher root biomass than aboveground biomass while competing and under stress. Forest stands established on abandoned agricultural lands usually continue to reflect the agricultural history for a long time after abandonment (Dupouey et al., 2002). These stands often vary in soil chemical properties from the stands that have been perpetually forested for several centuries (Flinn & Vellend, 2005; Zhang et al., 2010). Therefore, the previously fertilized soil in the AAS site may have likely contributed to the RCD, height, and aboveground biomass of black pine saplings.

Wood morphological, anatomical and wood density characteristics

We found a high variety of different morphological and anatomical properties in saplings between three study sites. Each study site had almost similar cambial age in their saplings. However, individual saplings showed site-specific variables in their morphology and anatomy. Saplings growing at AAS and NRS mostly exhibited greater mean values of pith%, xylem%, tree ring width, tracheid lumen width, and tracheid wall thickness than in FOS. A previous study by Brown et al. (1995) has shown that the increase of pith proportion positively affects tree height growth. The results of this study are in agreement with the findings of Brown et al. (1995). In this study, we found greater height growth and pith proportion in AAS and NRS sites than in FOS. Therefore, we can assume that a greater proportion of pith in AAS and NRS sites can provide an advantage for the height growth of black pine saplings.

Bark thickness is also considered to be a good indicator to determine the effect of local site environments since it has critical functions for tree growth and survival (i.e., protection of stems to abiotic and biotic threats and reduction of respiration). In the present study, the FOS showed greater bark thickness than the other two study sites. The thicker bark at FOS could be due to the environmental conditions since trees could be exposed to more extreme climatic conditions (wind and snow loads) directly, and therefore trees may need to produce thicker barks that provide biomechanical strength to the stems. Previous studies have also suggested that thicker barks could contribute almost 10% mechanical strength to the trunks and branches (Niklas, 1999; Paine et al., 2010). Contrary to the FOS, the AAS and NRS sites had slightly thinner barks which could be due to the sampling density. As stated above, sapling density and bark proportions were found negatively associated with each other such that higher sapling density caused thinner barks. It can be suggested that the AAS and NRS sites had thinner barks due to their greater sapling density. This could be also considered to be an adaptive strategy of trees to their local environments since higher sapling density could provide a protective role for tree growth thus not all saplings could face harsh environments directly. Previous studies however have shown that bark generally gets thicker when trees have greater stem diameters (Paine et al., 2010; Williams et al., 2007). The findings of this study however differed from previous studies since we found greater diameters in the AAS and NRS sites which showed lower bark thickness than FOS. The difference may partly be explained by different environmental situations in each site.

Tree ring width also plays a decisive role to understand general interactions between tree growth and site conditions since it shows a tree's ecological responses to its local environments (Fritts, 1966; Rossi et al., 2009). Previous studies have shown that the growth rate of trees could be measured by the tree ring widths such that trees show larger stem diameters and heights due to the greater amounts of tree ring formations (Bogino & Bravo, 2009; Campelo et al., 2013; Rigling et al., 2001). This study revealed similar findings with previous studies. AAS and NRS had greater stem height and diameter growths than FOS and also produced wider tree rings in their stems than FOS. On the other hand, the wider tree rings in the AAS and NRS sites were explained by the proportion of pith in this study. AAS and NRS showed more than two times greater ring widths than FOS due to their greater proportion of pith.

In general, the wider tree rings were also related to the cambial activity since higher cell production rates could produce wider tree rings (Fritts et al., 1991; Rathgeber et al., 2011). In this study, TLW and TWT were greater in AAS and these findings may partly explain why tree rings were wider in this study site. The larger lumen areas and wall thickness could also suggest that earlywood proportion is higher in AAS, thus saplings may provide better water transport, and more likely increase their water storage capacity for growth and development (Vieira et al., 2009; Ziaco et al., 2014a, 2014b). Prior studies also suggested that the fast growing of trees in height results in a greater proportion of earlywood, thus produce larger tracheid lumen widths to provide higher water storage (Fengel & Wegener, 1989; Zhang and Chui,1996; Makinen et al., 2002; Zhu et al., 2007). In this study, we also found that the AAS and NRS study sites had greater sapling growths due to their larger tracheid lumen widths. However, each study site exhibited quite similar tracheid length and width in their saplings. A further study with more focus on tracheid sizes between different site conditions is therefore suggested.

Surprisingly, although the FOS study site had greater basic wood density in their saplings, we did not find significant differences in the basic wood density of saplings between the AAS, NRS, and FOS study sites. Wood density however is an important parameter to determine the wood quality and shows the volume growth of trees under different site conditions. In general, denser woods have a thicker cell wall with smaller lumen areas (Barnett & Jeronimidis, 2003; Fritts, 2001; Gibson et al., 1988). Previous studies found that greater density provides greater strength and stiffness to tree structure thus trees can withstand environmental loads for a long time (Montes et al., 2017; Özden & Ennos, 2018; Smith & Chui, 1994). In this study, denser woods in the FOS study sites could be related to study site conditions since saplings growing at the FOS site may be exposed to more environmental factors; therefore, they could produce stronger woods in their stems to survive.

Conclusions

The findings of this study showed that site treatments and overstory openness both play a crucial role in the establishment and development of black pine. Therefore, the timing and intensity of overstory removal and site preparation should be considered when regenerating black pine forests. The present study also clearly showed that saplings growing in different site conditions exhibited site-specific morphological and anatomical traits. Particularly, abandoned agricultural land could be considered to produce favorable conditions for tree growth and development in this study, since tree ring formation and certain morpho-anatomical characteristics (pith proportion and tracheid lumen width) were greater in this site. Therefore, we can suggest that adaptability and tree growth performance to future harsh ecological conditions in abandoned agricultural land are comparable to a naturally regenerated forest. Trees growing in abandoned agricultural land could more likely be able to reserve their soil minerals and water efficiently in extreme environmental conditions due to their greater morphological and anatomical plasticity. Future monitoring is needed to observe the development of black pine over time on the abandoned agricultural land, because the long-term influence of these lands is unknown for these forests. Initial findings in this study would create a basis for our knowledge on the natural conversion of abandoned agricultural lands into black pine forests. However, additional studies with a higher number of samples would be recommended. Furthermore, similar researches could be conducted in other abandoned agricultural lands occupied with different tree species.

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Author contribution FK and SÖ conceived the idea, developed the framework, collected data, and compiled the literature; EFL worked with the writing and editing.

Availability of data and material The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Code availability NA

Declarations

Ethics approval NA

Conflict of interest The authors declare no competing interests.

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