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Full Length Research Paper

Structure and regeneration of *Parkia biglobosa* (Jacq.) R. Br Ex G Don in Mount Mandara, Cameroon

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A study was carried out in the Mount Mandara area to assess the populations and renewal status of *Parkia biglobosa*, a multi-purpose species. The study was aimed at contributing to the sustainable management of this resource. Transect methods were used to count individuals of *P. biglobosa* in fields. Results showed that the density of *P. biglobosa* was 7 individuals ha⁻¹. The basal area of *P. biglobosa* individuals varied as follows: Mokolo area (21.41 m² ha⁻¹) and Roua-plateaux Zoulgo (20.06 m² ha⁻¹). In these zones, big trees of *P. biglobosa* with large diameters were quite numerous. The average diameters were 79.14 ± 5.04 cm, and the average height was 13.60 ± 0.96 m. The structure of the distribution among the diameters generally was bell shaped, but the distributions varied in each zone. The greatest number of individuals was observed in the [50-70 cm] and [70-90 cm] diameter classes, with a remarkable increased presence of individuals in class [130-150 cm]. The scarcity of the individuals in the diameter range of 10 to 30 cm was noted in the various zones, with their entire absence in the Roua- Plateaux Zoulgo and Méri zones. The rate of regeneration of *P. biglobosa* was 13 %. This rate was very weak compared to the socio-economic importance of this species. This regeneration rate will not ensure continued population strength of this multipurpose species. This species is proven of importance, but its capacity of regeneration was too low to ensure its sustainability. It becomes imperative to develop effective strategies for its regeneration and conservation.

Key words: Population, dynamic, sustainable management, multi-purpose, *Parkia biglobosa*, Mount Mandara.

INTRODUCTION

In sub-Saharan Africa, trees play an essential role in the life of the rural population. They provide essential

foodstuffs, drugs, fodder, and wood-energy and contribute to the maintenance and stability of ecosystems

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(Le Houérou, 1980; Akpo, 1998; FAO, 2005; Gning, 2008; Mbow, 2008; Bakhoun et al., 2012b; Ngom, 2013; Sarr et al., 2013). *Parkia biglobosa* belongs to a group of privileged forest species, because of their multiple and advantageous uses to the local population (Aubreville, 1950). It is the most common species of the agroforest systems and agrosylvicultures, which occupy a significant place in the semi-arid zones and sub humid regions for its capacity of great use to the local populations (Boffa, 2000; Sacande and Clethero, 2007). Its distribution area encompasses the African, Asian and of South American continents. In the Mount Mandara, *P. biglobosa* also seems to be one of the privileged species because of its multiple uses. The populations generally use its fruit as food in wetting period (Bergeret and Ribot, 1990; Boffa, 2000). Its seeds are fermented to make a cooking spice named "soubala" or "dawa dawa", made up of a black colour, strong odour, nutritive, tasty and high protein value (Bonkougou et al., 2002; Steinkraus, 1996a). The medicinal uses of *P. biglobosa* are diversified. It is used to cure digestive tract (diarrhoea, dysentery, abdominal pain) diseases, cardiovascular system diseases, traumatism and burns, infectious diseases (shingles, malaria, boils, yellow fever, measles, chicken pox, oedemas), paediatric pathologies, symptoms and syndromes (faintness, tiredness, headaches, pain of hip, rheumatism, excessive loss of weight, elephantiasis, beginning of the paralysis (Koura et al., 2011; Mamadou, 2013; Adejumo et al., 2014). The fruits and fermented seeds are consumed and sold. The income generated thus contributes to the improvement of the household's economy and the reduction of poverty (Giffard, 1974; Boffa, 2000; Mamadou, 2013; Adejumo et al., 2014; Koura et al., 2011). The *P. biglobosa* shell fruits or other fruit residue provide many environmental services: it provides a way for the ground to be de-salted for rice way repair and contributes to habitat protection against termite attacks, and acts to suppress bad grass (Mamadou, 2013). Since the repetitive drought cycles, Sahelian farmers have harvested wild fruits and use it to avoid food shortage or as auxiliary sources to cover the food deficit periods (Bergeret and Ribot, 1990; Boffa, 2000; Sawadogo et al., 2008). In the Mounts Mandara, *P. biglobosa* is strongly exploited by the populations for its fruits and particularly its seeds in the transformation of "Dawa dawa". These products are the subject of a trans-border trade. The exploitation of the fruits and seeds of this species tends to intensify the increases in pressure on the resource. It becomes an imperative to provide necessary information for the sustainable management of this important plant. In spite of the significant scientific work load devoted to *P. biglobosa* in its phytogeographical area (Beaulaton and Gutierrez, 2002; Douma et al., 2010; Koura et al., 2013), no scientific study was devoted to the demography and the regeneration status of this species in the Mount Mandara in the Sahelian Zone of Cameroon. However, this species

has particular socio-economic importance because of the utilisation of its various products by local populations. This study brings acknowledgement on the potential availability of this species in order to develop the suitable strategies for its conservation and its sustainable management in the Mount Mandara.

The study's aim is to contribute to the sustainable management of this multi-purpose species in the Sudano-Sahelian zone. More specifically it aims to: (1) to determine the structure of the populations of *Parkia biglobosa*; (2) enumerate the seedling and the rejections on stumps for the assessment of its regeneration.

MATERIALS AND METHODS

Study area

Investigations were carried out in the Mount Mandara located between 10°45' 0 " N. and 13°40' 0 " E. In the administrative plan, it covers two regions: North and Far-North and extending on 4 divisions (Mayo-Sava, Diamare, Mayo-Tsanaga in the Far-North region and Mayo-Louti in the North region) (Figure 1). The climate of the Mount Mandara was characterized by the Sudano-Sahelian type and modified by the orogenic effects. Two varieties were known: in the North, the climate was Sahelo-Sudanian type with a period of four months of rainfall and in the South, the Sudano-Sahelian type with a period of five months of rainfall. The annual average rainfall was about 800 mm in the Northern part and 1100 mm in the Southern part. The temperatures oscillated between 13°C (January) and 38°C (April); the annual average temperature was 28°C. This vegetation was strongly degraded under the pressure of the anthropogenic actions (cutting for firewood, charcoal, wood service, bush fires and overgrazing) and the climatic threat (Letouzey, 1985; Westphal et al., 1985). The economic activities of the zone were based on extensive agriculture subsistence, extensive breeding, exploitation of resources, craft industry and the small trade, generating a substantial income for the poor rural families.

Data collection

To carry out this study, 40 villages were selected along the Mandara Mountains chains, according to the criteria of accessibility, type of dominant activities, demographics and the presence of some ethnic groups. These villages were gathered in 5 zones as follows: Mokolo; Roua-Plateau Zoulgo; Bourha-Plateau Kapsiki; Hina-Guider; Méri. The dendrometric data were collected along transects of 2000 m length and 20 m of width, on the whole 36 transects were established in various milieu within the basal mount and some mountainous zones. The sites (5 zones) of the investigation were as follows: Mokolo (24 ha), Roua-Plateau Zoulgo (48 ha), Bourha-Plateau Kapsiki (44 ha), Hina-Guider (16 ha), and Méri (12 ha). The stems of *P. biglobosa* were counted in all milieus such as the valleys, plates, plains and in relation to altitude. This species grows in flat terrain, with more than 1000 m of altitude. Along transects, the circumference (≥ 10 cm) of all *P. biglobosa* trees was measured at breast height 1.3 m from the ground, using a decametre. The height and the diameter of the tree archway size were estimated and measured. All anthropogenic traces were observed and noted on each tree. Inside transects, plots of 20 m x 20 m were established randomly and used to count the seedling and rejections. A distance of 100 m was made between plots. The seedlings, and the trees that had been cut but rejected and left

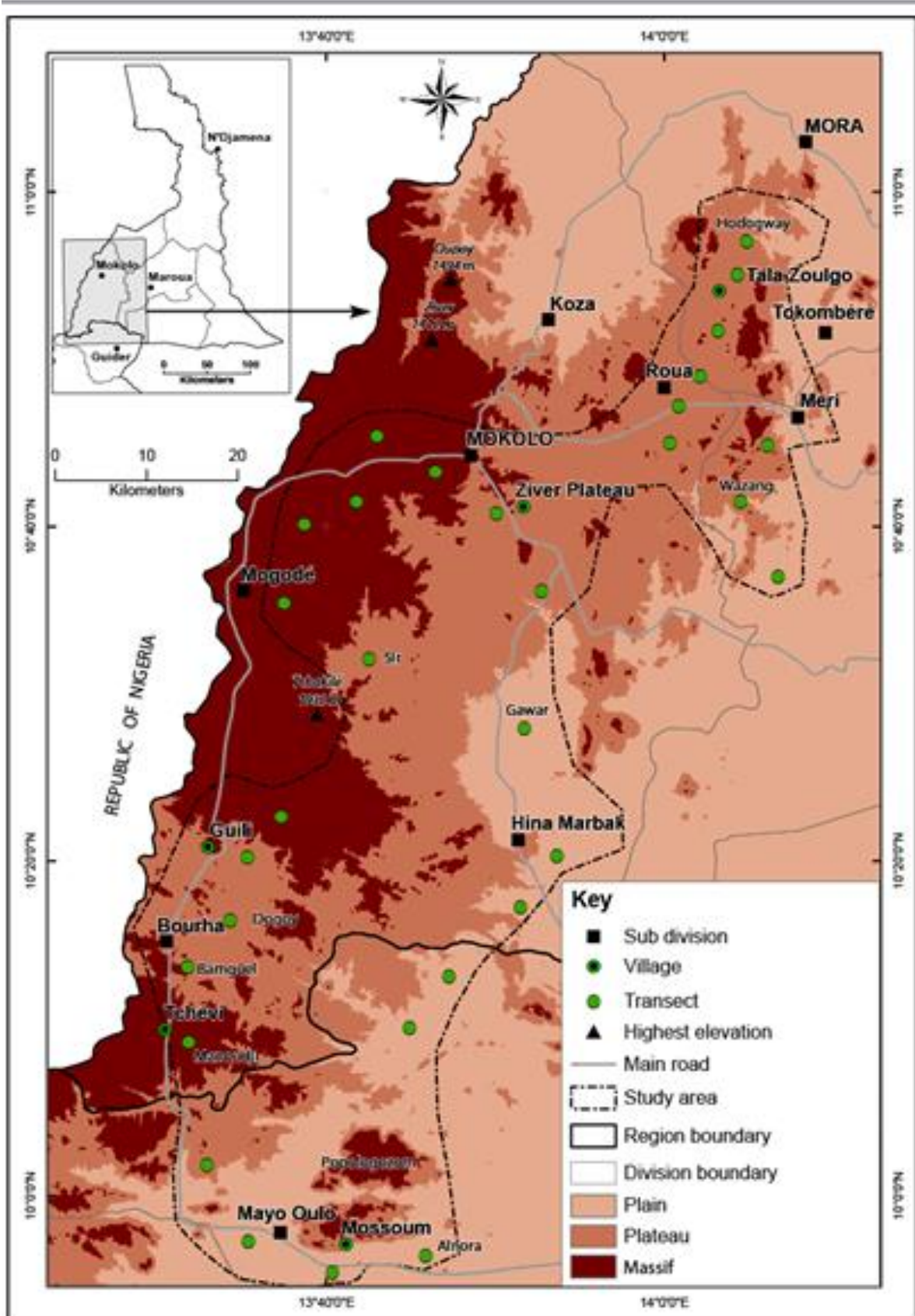


Figure 1. Study area and the transects itinerary in the Mount Mandara.

Table 1. Dendrometric parameters of *P. biglobosa* in the Mount Mandara.

Zone	Density	Average diameter and bar charts (cm)	Basal area (m ² /ha ⁻¹)	Average height and bar charts (m)	Average tree archway (m)
Mokolo	4.29 ^b	87.16 ^a (28.66)	21.41 ^a	14.76 ^a (5.12)	14.05 ^a (5.21)
Roua-Plateaux Zoulgo	6.9 ^a	81.69 ^a (25.89)	20.06 ^a	14.56 ^a (4.25)	14.13 ^a (6.48)
Bourha-Plateaux Kapsiki	11.70 ^a	79.01 ^a (37.11)	19.40 ^a	12.15 ^a (3.68)	11.53 ^a (5.41)
Hina-Guider	1.38 ^b	74.80 ^a (31.10)	18.37 ^a	13.48 ^a (4.31)	10.95 ^a (4.42)
Méri	1.25 ^b	73.04 ^a (27.01)	17.94 ^a	13.07 ^a (2.74)	10.20 ^a (3.97)

According to the analyses of variance, the same letters in the same column means similarity and dissimilarity in the contrary cases. The numbers between parentheses are the bar charts.

standing, were counted. The trees that had been cut and the dead trees were also counted.

Data analysis

The dendrometric parameters in particular, the diameter of the tree, basal area (G) and the density of *Parkia biglobosa* were calculated. The basal area (G) was the sum of the surface of the cross sections of *Parkia biglobosa* individuals measured at 1.30 m. It was determined by the formula (Sourou et al., 2016):

$$G = \sum_{i=1}^n C_i^2 / 4\pi$$

with G, basal area of the trees within a transect and Ci, the circumference.

Tree density was determined as the number of *P. biglobosa* trees counted per hectare. The regeneration rate and the death rate of the population of *Parkia biglobosa* were calculated and expressed as a percentage. Diameter structure of *P. biglobosa* population was measured by grouping individual trees into 20 cm diameter class sizes, height distributions and tree archways. The distribution was analysed and displayed in the various explored zones using histograms in Microsoft Excel. Other variables such as means and percentages were determined using ANOVA in STAGRAPHICSPLUS 5.5.

RESULTS

Distribution of the population of *P. biglobosa* in the Mount Mandara

In the five explored zones, a total of 993 individuals of *P. biglobosa* were counted. The total density of the individuals in the Mount Mandara was 7 individuals ha⁻¹. In this vegetation, the distribution of *P. biglobosa* varies in the various explored zones as follows: Bourha-plateau Kapsiki (11.70 individuals ha⁻¹), Roua-plateau Zoulgo (6.87 individuals ha⁻¹), Mokolo (4.29 individuals ha⁻¹), Hina-Guider (1.38 individuals ha⁻¹), and Méri (1.25 individuals ha⁻¹) (Table 1). This variability was shown to be statistically significant (p= 0.0021) using analysis of variance. A low density of the populations of *P.*

biglobosa was noted in Hina-Guider (1.38 individuals ha⁻¹) and Méri (1.25 individuals ha⁻¹). These zones were located between the altitude and the plain zones where the rainfall decreases and the vegetation is strongly degraded.

In the Mount Mandara, the average diameter of *P. biglobosa* was 79.14 (5.04) cm. In the five explored zones, the average diameter varies from 73.04 (27.01) cm in Meri to 87.16 (28.66) cm in Mokolo (Table 1). The average diameter was respectively of 79.01 (37.11), 81.69 (25.89) and 74.80 (1.10) cm in Bourha-Plateau Kapsiki, Roua-Plateau Zoulgo and Hina-Guider zones. The difference between the average diameters in the zones was not significant. The average height of the trees was 13.60 ± 0.96 m. This height varied in the various zones from 12.15 ± 3.68 m in Bourha-Plateau Kapsiki to 14.76 ± 5.12 m in Mokolo. The average heights were not significantly different in the various zones. The average tree archway sizes was 12.17 (1.62) m. The tree archway size also varied from 14.13 (6.48) m to 10.20 (3.97) m. These results showed that, in the Mount Mandara, *P. biglobosa* was a large tree. It develops a great height, a large tree archway size and a large diameter. Pluviometry and the anthropogenic pressure seem to be the determining factors for the blooming of this species in its phytogeographic area.

In the Mount Mandara, basal area of *P. biglobosa* in the various zones varies from 17.94 m² ha⁻¹ in Méri to 21.41 m² ha⁻¹ in Mokolo. The individuals of *P. boglobosa* had a significant cover in the Mokolo (21.41 m² ha) and Roua-plateau Zoulgo (20.06 m² ha⁻¹) zones. In these zones, the big trees of *P. biglobosa* with the large diameter were rather numerous. These two zones were characterized by the population's culture in conservation of this species; they were developed for a long period of time in the farmer land and habitation area.

Structure in diameter of *P. biglobosa* in the Mount Mandara

The structure of distribution in diameters of the individuals of *P. biglobosa* of the Mounts Mandara

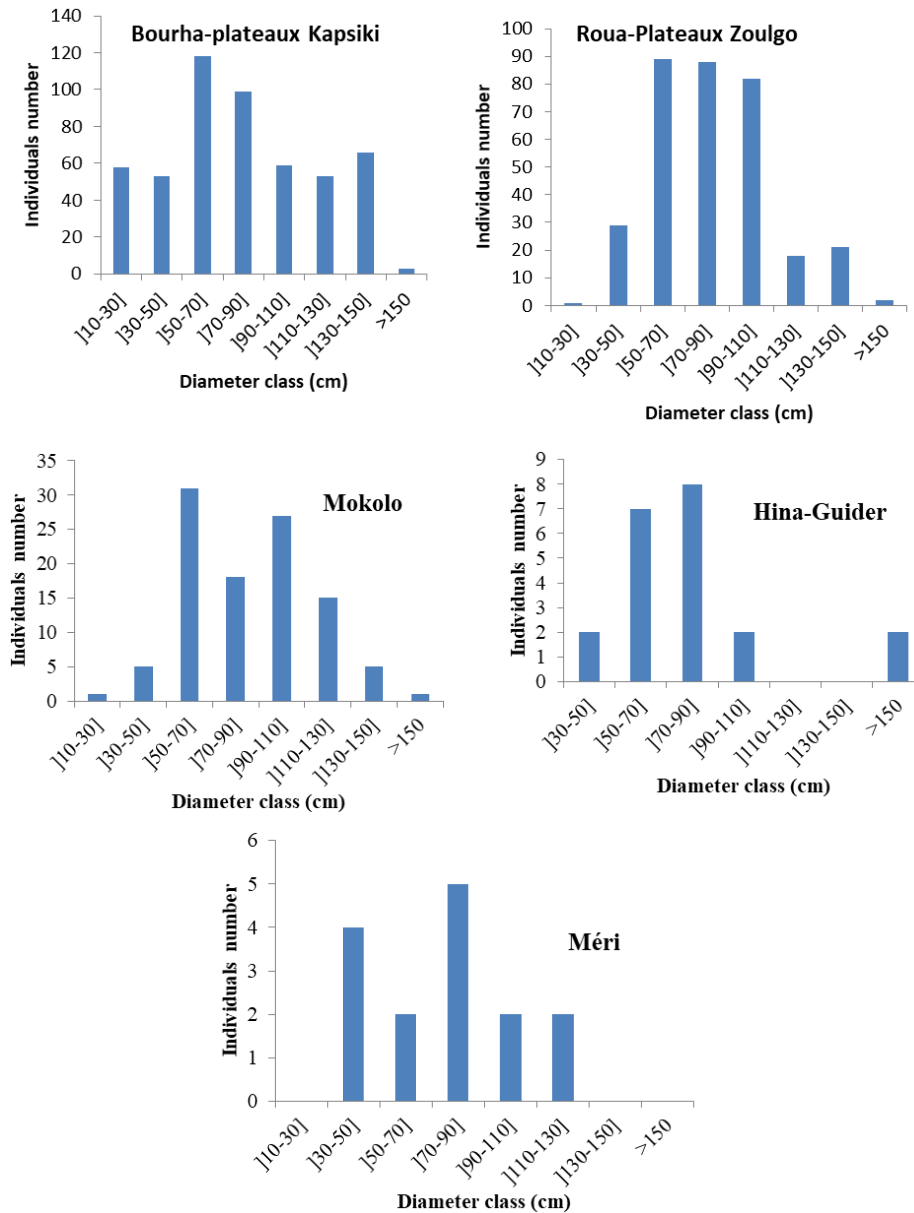


Figure 2. Diameter distributions of *P. biglobosa* in the Mount Mandara.

presented in general a bell shape, but this distribution varied in each zone (Figure 2). They were observed in the diameter classes of [50-70 and 70-90] with a remarkable presence of the individuals in the class [130-150] in Bourha-Plateau Kapsiki with the absence of individuals in this class of Hina-Guider. In the Roua-Plateau Zoulgo zone, the greatest number of individuals was counted between the diameter classes of [50-70] and [90-110]. In the Méri zones, the greatest number of individuals was enumerated in the diameter were noted in the diameter classes of [50-70] and [90-110]. The scarcity of the individuals in diameter from 10 to 30 cm was noted in the various zones with a whole lack in Roua-Plateau

Zoulgo and Méri zones. The individuals of *P. biglobosa* generally form a part of the agricultural landscape. Thus, the structures in diameter of the populations of this species translated the importance of its conservation by the local populations, as far as its socio-economic importance is concerned. In a general way, the individuals of large diameters (≥ 150 cm) are rare. The two categories of individuals, regenerated stems and large trees did not have a homogeneous distribution in all the zones. The scarcity or the absence of individuals of small diameter showed that, the natural regeneration of the species was difficult. The seedlings had difficulties of being maintained because of the effects of the dryness

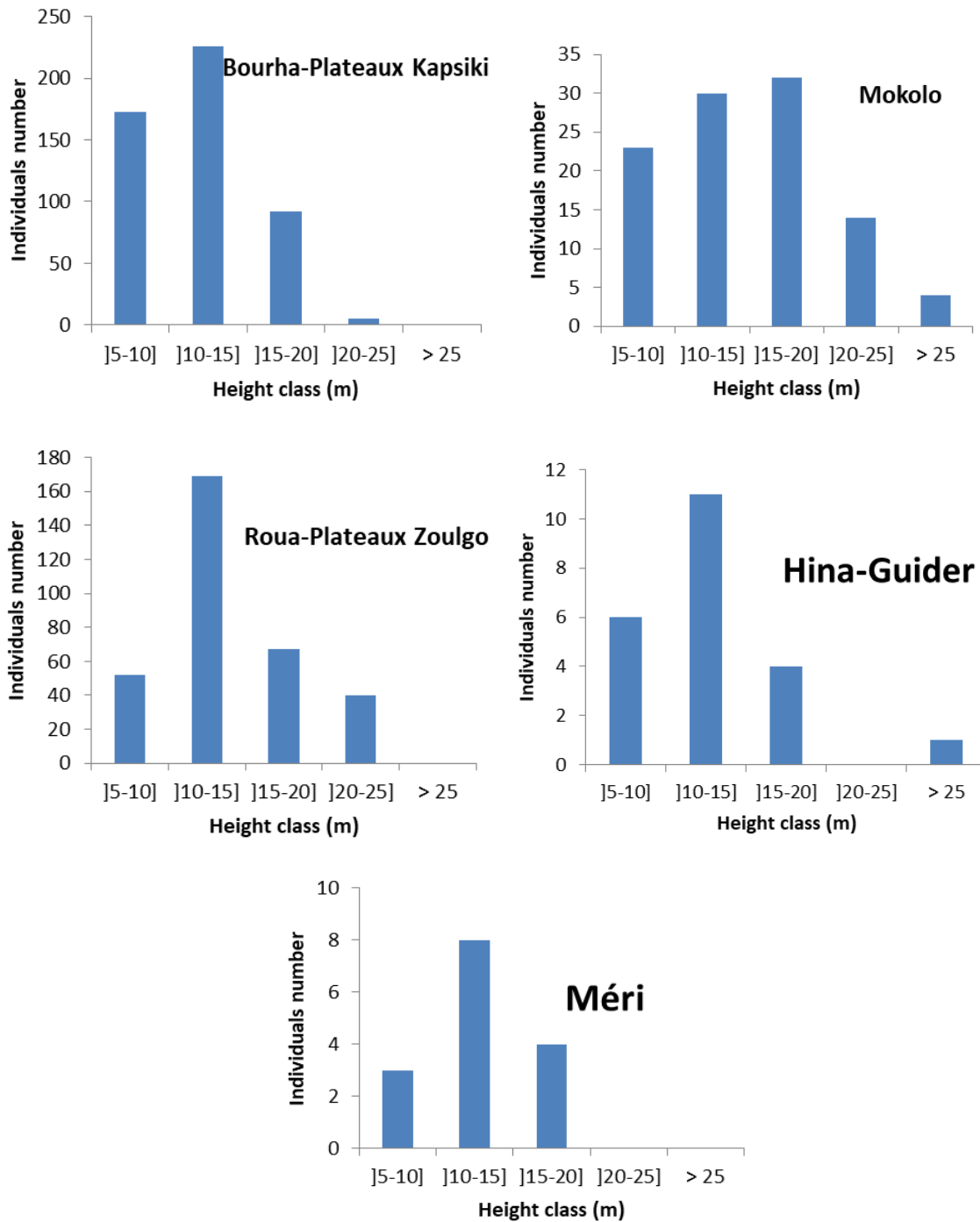


Figure 3. Vertical distribution of *P. biglobosa* in the Mount Mandara.

and the pasture where they were growing during the long period of the dry season. The strong anthropogenic pressure remains also a factor of the regressive evolution of the *P. biglobosa* populations in the Mount Mandara. Under these conditions, it could be an imperative to take important measures in favour of the conservation and the sustainable management of this multipurpose plant.

Structure in height of *P. biglobosa* in the Mount Mandara

The distribution in height of the individuals of *P. biglobosa* followed a pattern of bell shape. It was roughly the same pace as that of the structure in diametric distribution with some particular characteristics (Figure 3). In Bourha-

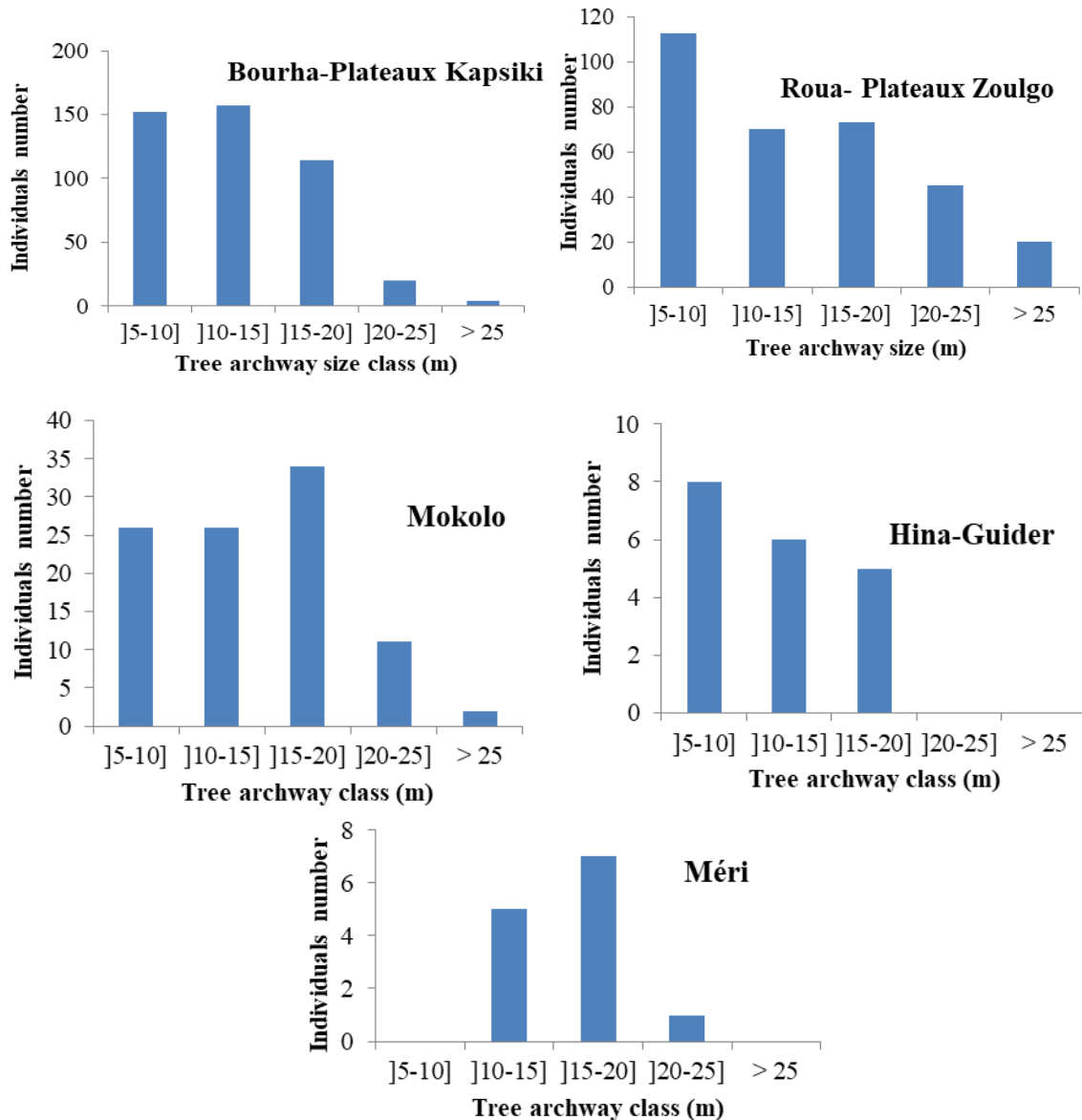


Figure 4. Distribution of the tree archway size study zones.

Plateaux Kapsiki, ZoulgoRoua-Plateau, Hina-Guider and Méri zones, the greatest number of individuals was noted in the class height [10-15 m]. Whereas in Mokolo zone, the classes height [10-15] and [15-20] presented the greatest number of individuals. According to the distribution in diameter, the individuals' height was rarely reduced, 10 m than 20 m were rare. Nevertheless, it should be noted that the younger individuals were more numerous in the Bourha-Plateau Kapsiki and Mokolo than in the other explored zones.

The tendency of the histograms of tree archway size distribution showed that the majority of the individuals concentrate in a general way in the classes [5 -10] and [10 -15]. The variation was very remarkable in Mokolo

and Méri zones where the record number of individuals concentrated in the classes of [10-15] and [15-20] with a scarcity of the individuals in the classes of [5 -10] and ≥ 25 (Figure 4). Broadly, the greatest number of individuals was concentrated around the average of tree archway size (12.17 ± 1.62 m). This variability was shown by the calculation of variance with $p = 0.0161$. These results illustrated the behaviour and the interest which the population of the various zones had on this specie. In certain zones like Mokolo, Roua-plateau Zoulgo, and Hina- Guider, the farmers practised the upholding of the old and unproductive trees by pruning for the renovation and a resumption of fructification, thus contributing to the reduction of the size of the tree archway size.

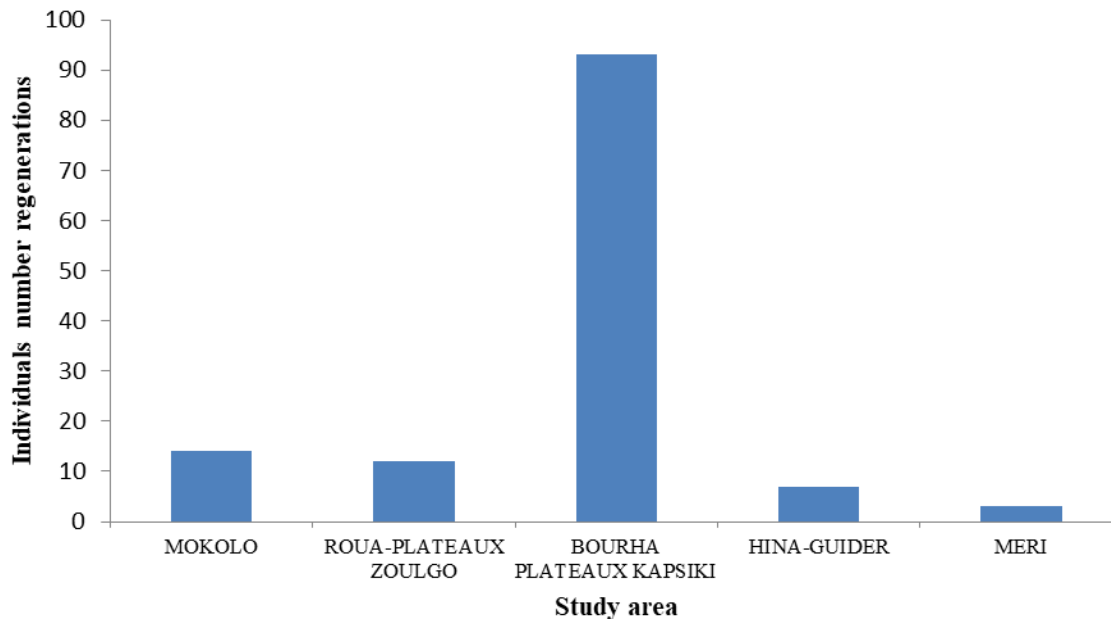


Figure 5. Regeneration status of *P. biglobosa* in the Mount Mandara.

Regeneration status of *P. biglobosa* in Munts Mandara

In the Mounts Mandara, the regeneration rate of *P. biglobosa* was 13%. In the Bourha-Kapsiki zone, the regeneration rate was 9.36% (Figure 5). In the four other zones, the youthful individuals are rare: Mokolo (1.40%), Roua-Plateau-Zoulgo (1.20%), Hina-Guider (0.7%), and Meri (0.3%). The distribution of the seedlings and rejections were significantly different in the five studies zones with $p = 0.0024$. The regeneration rate noted in Bourha-Kapsiki explained the interest which the local populations had for the conservation of *P. biglobosa*.

DISCUSSION

Distribution of population of *P. biglobosa* in the Mount Mandara

The density of *P. biglobosa* was 7 individuals ha^{-1} in the Mount Mandara, this density varied from 11.70 individual ha^{-1} to 1.25 individual ha^{-1} in the various explored zones. These results were near to those obtained by Gbédji (2003) (1 to 5 individual ha^{-1}) in the *P. Biglobosa* parks in Benin; Douma et al. (2010) (1.64 individual ha^{-1}) in a study in Sahelo Sudanese area in Niger and Douamba (2006) (8 individuals ha^{-1}) in Sudano-Guinean zones. These authors justify the low densities obtained due to the fact that, individuals of *P. biglobosa* were subjected to a strong anthropogenic pressure. The wood of this species was required in craft industry to fabricate arts objects. On the other hand, these values of density were

definitely lower than those obtained by Koura et al. (2013) (39 to 70 individuals ha^{-1}) in various zones in Benin obtained by Ouédraogo (1995) (5 to 25 individuals ha^{-1}) and those of Thiombiano et al. (2010) (37 individuals ha^{-1}), whose work were undertaken in various zones in Burkina Faso. According to these authors, the zones where the density of *P. biglobosa* were high, it had a great socio-economic and cultural value in the life of the population. It was the case noted in the present study where the densities were significant in the zones of Bourha-Plateau Kapsiki and Roua-Plateau Zoulgo, where the plant was of remarkable socio-economic interest for the population. The same observations were made by Koura et al. (2011) where the densities of *P. biglobosa* were high, local knowledge were diversified. In spite of the rocky field, *P. biglobosa* had a remarkable capacity of adaptation in mountainous zone. *P. biglobosa* developed a root system and succeeded in being clutched with the blocks of rocks. In the Mounts Mandara, the volume of rainfall was particularly significant upon the influence of altitude on the climate, making this milieu favourable for the development of *P. biglobosa*. It was found in altitude 1000 m and also in the plains. The studies of Douamba (2006) in Sudano-guinean zone concluded that *P. biglobosa* developed in flat grounds on the clay soils, muddy and muddy argilo that, *P. biglobosa* was not identified in altitude. According to the various results, it was possible to affirm that *P. biglobosa* developed well in a plain and in altitude on various types of ground in the Sudano-Guinean and Sudano-Sahelians zones.

In the Mounts Mandara, *Parkia biglobosa* was a large tree, but a weak number of the regeneration individuals to

ensure the renewal of the species were noted. However, *P. biglobosa* developed also a large diameter and great height. These observations were in agreement with Douma et al. (2010) a study of the Tamou rural district in Niger, which were shown that 80% of the listed individuals were large trees and noted a weak recruitment of the juvenile individuals. The occurrence of old trees was remarkable and indicated an elderly of this species populations and a low capacity of regeneration. Seignobos (1982) shows that the abundance of old trees can be considered as an index of threat to the disappearance of this species in the agroforests system of the zone and the result of a shaping by the anthropic activities.

The basal area of *P. biglobosa* in the various zones of Mounts Mandara, varied from 17.94 to 21.41 m² ha⁻¹, respectively in Méri and Mokolo zones. The individuals of *P. biglobosa* had a significant coverage in Mokolo (21.41 m² ha⁻¹) and Roua-plateau Zoulgo (20.06 m² ha⁻¹) zones. In these zones, *P. biglobosa* trees with large diameter were quite numerous. Because of the socio-economic importance of this species, the populations long times had developed the ethnicity of its conservation since a long time ago. These results were higher than those of Koura et al. (2013) which found 4.93 to 12.38 m² ha⁻¹ in *P. biglobosa* parks in Benin.

The structure of distribution in diameter of *P. biglobosa* in the Mounts Mandara shows that the greatest number of individuals was observed in the diameter class of [50-70] and [70-90] with a remarkable presence of individuals in class [130-150] in Bourha-Plateau Kapsiki zone with the lack of individuals in this class in Hina-Guider. The scarcity of individuals in diameter 10 to 30 cm was noted in the various zones with quasi absence in Roua- Zoulgo Plateau and Méri. The individuals of *P. biglobosa* were found in the agricultural landscape. These results were confirmed by those of Koura et al. (2013) which had shown a similar structure in diameter and concluded that, the structures in diameter of *P. biglobosa* population translated the impact of the management of this species by farmers. The local management did not worry about the regeneration status of the species and presented a stunning deficit in the lower diameter classes ranging between 10 and 20 cm (Koura et al., 2013). The scarcity or the absence of the seedling individuals showed that the natural regeneration of *P. biglobosa* was difficult. Seedlings had difficulties of being preserved because of the effects of dryness and pasture which they faced during the long period of the dry season. The strong anthropogenic pressure remains also an important factor of the regressive evolution of the *P. biglobosa* populations in the Mounts Mandara. It was an imperative under these conditions of taking measures in favour of the conservation and the sustainable management of the plant. If provisions were not taken for the sustainable management, the species will extinct. The same reports were made by Thiombiano et al. (2010) on ageing and

instability of *P. biglobosa* populations in Burkina Faso. Sambou et al. (2007) in Senegal and Oumarou et al., (2009) in Burkina Faso also found that *P. biglobosa* populations in savannas had a regressive structure. Human activities through the anarchistic exploitation of trees and the introduction of animals seem to be the causes of this regression (Douma et al., 2010).

Regeneration status of *P. biglobosa* in the Mount Mandara

The regeneration rate of *P. biglobosa* in the Mount Mandara was weak and could not permit ensure the sustainability of this significant species. The seedlings were confronted during a year to the climatic threat such as dryness and weak rainfall and to the anthropogenic activities pressure. The survival of the seedlings between the rainy and dry seasons was difficult. Even if the seeds germinated in the raining season, during the dry season, the seedlings in their majority perished. Indeed, seedlings were grazed, trampled by animals in pasture, farmers exploited intensively *P. biglobosa* seeds; this dropped the potentiality of seeds germination in the fields, was and a probable scarce of seeds sprouted to ensure the regeneration. But it should be noted that, in the Mount Mandara the regeneration of *P. biglobosa* was natural and non-participative taking into account the interest which the populations developed around the plant. It was an imperative to develop the adequate strategies to stimulate the populations to contribute significantly to the assisted regeneration of *P. biglobosa*. The regeneration by rejections after the cutting of *P. biglobosa* was not identified in fields. However, the renovation of the old individuals following a pruning to obtain young branches able to produce more fruits was more frequent in the share of the study zones. This strategy of conservation contributed to an increase in the productivity of the individuals existing but was not of great importance in the regeneration of the potential on individuals of *P. biglobosa* in the Mounts Mandara. Ouédraogo (2006) and Oumarou et al. (2009) shows in their work that the seedlings undergo the combined action of the dryness, fires and predatory (cattle, insects, rodents, mushrooms).

Conclusion

The enumeration of the potential available of *P. biglobosa* along transects, in the various zones of the Mount Mandara, showed that this species was well distributed. This distribution correlated the socio-economic importance that farmers in the various zones had on this species and took part in its conservation. *P. biglobosa* sprouted as well on flat ground as in altitude up to 1000 m. *P. biglobosa* was a large tree in the Mount Mandara, it developed a large diameter. *Parkia biglobosa* populations

were composed in majority of the growing old individuals of big size. The structure of distribution and the potential regeneration of *P. biglobosa* showed that the renewal of plant was weak and that the population was subjected to anthropogenic activities and the climatic risks which impact on the sustainability of the species. However, the populations practised the renovation of old trees by pruning to obtain younger branches able to produce more fruits. This strategy of conservation contributed to an increase in the productivity of existed individuals of *P. biglobosa* in the Mount Mandara. It was an imperative to develop the strategies of regeneration and conservation *P. biglobosa* in the Mount Mandara. The popularization of the products of the plant, a plan of afforestation, the production of nursery trees and the encouragement of farmers in the preservation of the potential availability were advisable.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Moisture requirements for *Pinus sylvestris* seeds: Variation in germination among provenances from different elevations

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Patterns of climatic variables are likely to change significantly in the coming century, with important but poorly-understood consequences for plant communities. This experiment determined how seeds of *P. sylvestris* collected from different elevations in two geographical regions performed when placed in simulated moisture stress conditions. Eight provenances from two regions (Norway and France) were chilled for zero and four weeks at $4\pm 1^\circ\text{C}$ and then germinated at 10 and 20°C , at water potentials of -0.3, -0.6 and -0.9 MPa. Moisture stress resulted in delayed germination and reduced germination capacity, and there was no germination at a water potential of -0.9 MPa. Provenances from high elevations were the most susceptible to moisture stress.

Key words: Moisture stress, water potentials, simulated moisture conditions, germination capacity, temperature effects.

INTRODUCTION

Changes in climate are predicted to alter the geographic distributions of plant species, in part by affecting where individuals can establish from seed. Climate models predict that dry periods will become more common in Central Europe, the Western USA and East Asia (Eastaugh, 2011), and this may affect seed germination and seedling establishment. Germination begins with water uptake (imbibition), and imbibition rate is determined by the availability of water, the permeability of seed structures regulating the uptake of water, and conditions during hydration. Both factors can, separately or jointly, affect the germination capacity and germination

rate (Baskin and Baskin, 1998). Each plant species appears to have its own specific range of water potential requirements, below and above which no germination occurs, and optimum water potential at which germination is fastest.

The values of water potential ranged from zero (pure water) to large negative numbers such as -100 MPa for air-dry seeds. Increasing osmotic and matric effects reduce the total water potential to lower (more negative) values. The water potential of seeds imbibed sufficiently for germination has generally been considered to be high. Accurate models are required to describe seed responses

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Table 1. Geographic and climatic details for the eight *P. sylvestris* provenances used in this experiment.

Provenance	Elevation (m)	Mean annual temperature (°C)	Mean annual precipitation (mm)	Year of collection
Norway A1	420	2.8	1204	2006
Norway A2	250	2.8	1204	2006
Norway A3	230	3.4	1958	2006
Norway A4	150	5.0	1204	2006
France A1	330	9.7	759	2006
France A2	280	9.7	759	2006
France A3	280 to 250	10.2	759 to 711	2009
France A4	250	10.6	711	2006

Within region (Norway and France) provenances are shown in order of decreasing elevation.

to temperature and water potential and make effective predictions of field germination. Bradford (1990) introduced the idea of hydrotime for explaining germination. Hydrothermal germination models offer a useful way of understanding why and how seed germination varies under different moisture and temperature conditions, and among different seed populations (Bloomberg et al., 2009). Germination of a given fraction of seed is prevented if water potential is below the base water potential for that fraction. Germination rate increases with an increase in the distance between base water potential and actual water potential up to the optimum water potential (Meyer and Allen, 2009).

One technique for studying the effect of water potential on germination is to simulate moisture stress conditions using solutions of variable concentrations of large molecules such as polyethylene glycol (PEG) (Falusi et al., 1983). Polyethylene glycol is an inert, water-binding polymer with a non-ionic and virtually impermeable long chain and is not toxic to plants (Amooy and Fazlollahnejad, 2014). Polyethylene glycol of high molecular weight (PEG 8000) is more appropriate than smaller molecules such as PEG 4000 for creating moisture stress, because germination capacities of seed in polyethylene glycol 8000 and in soil with the same water potential are approximately equal (Rana et al., 2017). Studies have demonstrated that significant differences exist among seed sources in germination ability under moisture stress levels. There have been few studies to determine if seeds collected from different environmental conditions, particularly from areas of high and low rainfall, demonstrate variation in moisture stress tolerance. The work described here shows the effects of moisture stress on germination of *Pinus sylvestris* provenances from four elevations in Norway and four in France.

MATERIALS AND METHODS

Table 1 shows the geographic and climatic details for the eight

provenances from two regions (Norway and France, wet and dry climates respectively) of *Pinus sylvestris* used in this study. The provenances were selected to represent an elevation range from two parts of the species' range and seed was purchased from a government tree seed centre and a commercial tree seed supplier in Norway and France, respectively. Seeds of the France A3 provenance were collected from an elevation range that included the locations of the France A2 and A4 provenances.

Seed treatment: Target moisture content and chilling

At the start of experiment, all seeds to be chilled for any duration before germination testing were adjusted to a target moisture content of 30%. This is the moisture content at which seeds of *P. sylvestris* are optimally chilled (Afroze and O'Reilly, 2013). Target moisture content allows moist seeds to break dormancy and do not allow any radicle growth. The weight of water necessary to achieve this target moisture content was calculated using the following formula:

$$w = s - \frac{(m \times s)}{1 - tmc} - s.$$

Where w = weight of water to add (g); s = dry weight of seed (g); m = moisture content (fresh weight basis) of seed prior to treatment; tmc = target moisture content (% fresh weight basis) (Jones and Gosling, 1994).

The moisture content of seeds was determined using the oven dry method. Once the target moisture content (30±2 %) was achieved the polythene bags containing the seeds were loosely tied to allow gas exchange but restrict water loss and transferred to a walk-in cold dark room set at 4±1°C for the required chilling duration. The seeds were inspected and gently shaken once in a week to allow uniform imbibition and air circulation (Afroze and O'Reilly, 2013).

Moisture stress treatments

Polyethylene glycol (PEG 8000) was dissolved in deionised water to make solutions with water potentials of -0.3, -0.6 and -0.9 MPa. The equation used to determine the amount of PEG to be used for a given water potential is:

$$\Psi = 1.29 \times (\text{PEG})^2 \times T - 140 \times (\text{PEG})^2 - 4.0 \times (\text{PEG}).$$

where Ψ = water potential; PEG = polyethylene glycol concentration (g l^{-1}).

Deionised water (0.0 MPa) was used as the control. Water

potentials of the solutions used in germination boxes were monitored by measuring samples of the solutions using a HR-33T Dew Point Microvoltmeter and Sample Chamber C-52 (Wescor model).

Experimental procedure and analysis

Two chilling durations (zero and four weeks) at one chilling temperature (4°C) were used in this experiment. Seeds were germinated at two temperatures in two germination chambers, one set at 10°C and the other 20°C. Each temperature had fifty seeds. Filter paper in germination boxes were moistened with 150 ml of the appropriate PEG 8000 solution or deionised water. PEG solutions at a germination temperature of 20°C were renewed weekly by pouring out the existing solution and adding 150 ml of fresh solution in every germination box. Germination boxes were inspected daily. Germinated seeds were counted and discarded immediately; this avoided any recounting of the same germinants. The final germination count was done on day twenty-one. Germination capacity and rate of germination were calculated using the following formulas:

Germination capacity: $100 \times S/T$

Where S is the cumulative number of germinated seeds at the end of the experiment and T is the total number of sown seeds.

Germination rate: $(\sum Ni/Di)/S$

Where Ni is the number of seeds that germinated on day Di.

Statistical analyses were conducted at two levels. Firstly, differences in germination capacity and germination rate among seed sources and treatments (chilling temperatures and chilling durations) as factors were tested with a two-way ANOVA. None of the germination data from any of the experiments required transformation. Where differences among seed sources and / or treatments were significant ($P < 0.05$) a multiple comparison *post hoc* test was performed (Tukey test) to determine the significance of pairwise differences between means. Data analyses were conducted using Genstat (14th Edition, VSN International Ltd) and SPSS (20th Edition).

RESULTS

Norway: Germination capacity

Germination capacity was generally higher in seeds from low elevations, after chilling for four weeks at 4°C, at lower levels of water stress (water potentials of 0.0 and -0.3 MPa) and at 20°C. Without chilling and at 10°C, germination capacity ranged between 0 and 29%, and was highest in the lowest elevation (Norway A4) provenance. After four weeks chilling and at 10°C, germination capacity ranged between 0 and 78.5%, and highest germination capacity was in the second lowest elevation (Norway A3) provenance (Figure 1a and b). Without chilling and at 20°C, germination capacity ranged between 19 and 81.5%, and was highest in the two lower elevation (Norway A3 and A4) provenances. After four weeks chilling and at 20°C, germination capacity ranged between 30.5 and 96%, and was highest in the second lowest elevation (Norway A3) provenance (Figure 1c and

d). Germination at a water potential of -0.6 MPa occurred only at 20°C (Figure 1c and d).

Germination decreased as moisture stress increased and was completely inhibited at a water potential of -0.9 MPa. Analysis of variance showed that there were significant ($P < 0.001$) differences in germination capacity among provenances, chilling durations, moisture stress levels and germination temperatures (Table 2). There were significant interactions between all factors. The provenances were significantly different from each other, except for comparisons between Norway A3 and Norway A4 (Table 3).

Norway: Germination rate

Germination rate was generally higher in provenances from low elevations, after four weeks chilling at 4°C, at lower levels of moisture stress (water potentials of 0.0 and -0.3 MPa) and at 20°C. Without chilling and at 10°C, germination rate ranged between 0 and 0.7 day⁻¹, and was highest in the lowest elevation (Norway A4) provenance.

After four weeks chilling and at 10°C, germination rate ranged between 0 and 1.8 day⁻¹, and was highest in the second lowest elevation (Norway A3) provenance (Figure 2a and b). Without chilling and at 20°C, germination rate ranged between 0.3 and 1.9 day⁻¹, and was highest in the lower elevation (Norway A3 and A4) provenances. After four weeks chilling and at 20°C, germination rate ranged between 0.7 and 2.2 day⁻¹, and was highest in the second lowest elevation (Norway A3) provenance (Figure 2c and d). Table 4 shows the results of analysis of variance of germination rate. Germination rate was significantly ($P < 0.001$) different among provenances, chilling durations, moisture stress levels and germination temperatures. There were significant interactions between all factors except between provenance and chilling and between provenance and temperature (Table 4). The provenances were significantly different from each other, except for comparisons between Norway A3 and A4 (Table 5).

Norway: Cumulative germination

There was no germination of any provenances in the -0.6 MPa moisture stress treatment (Figure 3). Germination curves differed chiefly in the time at which germination started. Without chilling, germination started at 19-20 days in the control (0.0 MPa) moisture stress treatment and at day 21 in the -0.3 MPa treatment (Figure 3a and b). After four weeks chilling, germination started at 11 to 13 days in the control (0.0 MPa) moisture stress treatment and at 15-18 days in the -0.3 MPa treatment (Figure 3c and d). The difference in the start of germination between moisture stress levels was greatest for the provenance from the lowest elevation (Norway A4).

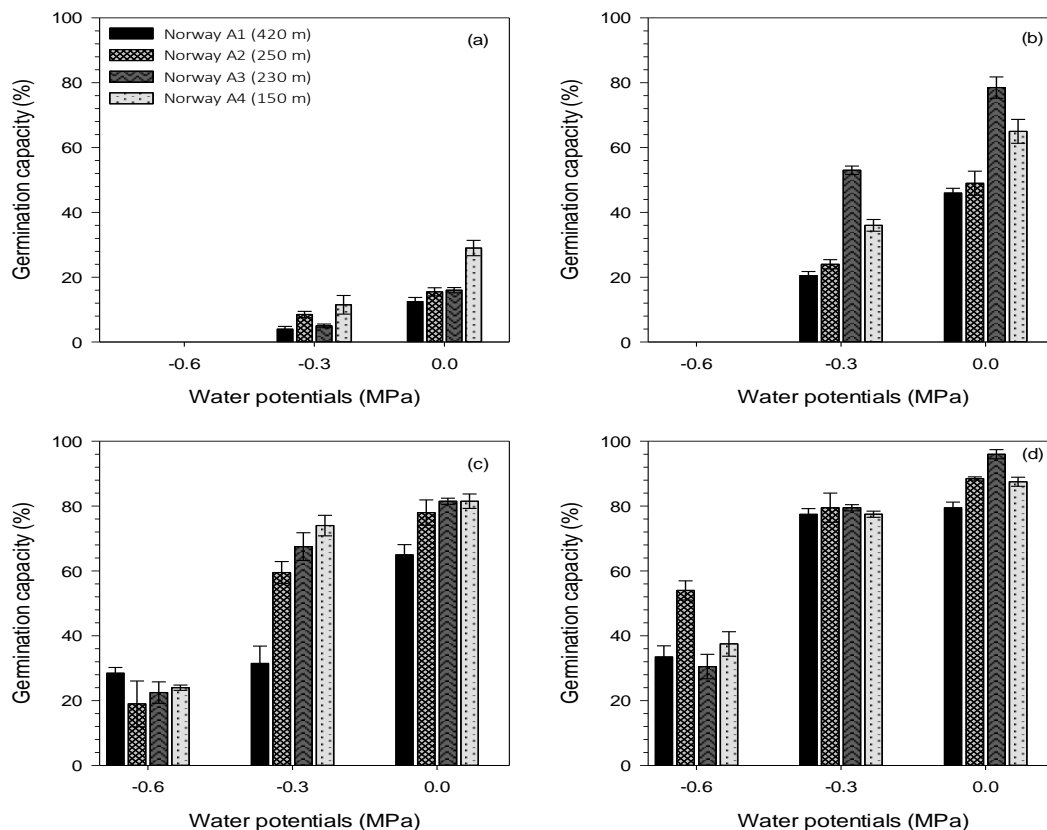


Figure 1. Germination capacity at water potentials of 0.0, -0.3 and -0.6 MPa in four provenances (elevations) of *P. sylvestris* from Norway: (a) zero weeks chilling, germination at 10°C; (b) four weeks chilling, germination at 10°C; (c) zero weeks chilling, germination at 20°C and (d) four weeks chilling, germination at 20°C.

Table 2. Analysis of variance of germination capacity of four provenances (elevations) of *P. sylvestris* from four altitudes in Norway after two chilling durations, at three moisture stress levels and at two temperatures.

Source of variation	d.f	F-value	P-value
Provenance	3	46.921	***
Chilling	1	559.712	***
Moisture stress	2	943.830	***
Temperature	1	2647.172	***
Provenance × chilling	3	7.356	NS
Provenance × moisture stress	6	51.513	***
Provenance × temperature	3	5.891	**
Chilling × moisture stress	2	25.547	***
Chilling × temperature	1	26.499	***
Temperature × moisture stress	2	4.078	*
Provenance × chilling × moisture stress × temperature	23	25.271	***
Error	144		
Total	191		

NS, not significant; *, ***, significant $p < 0.05$ and $p < 0.001$, respectively.

Figure 4 shows the cumulative germination of Norwegian provenances at 20°C after two chilling durations and at three moisture stress levels (water potentials of 0.0, -0.3

and -0.6 MPa). Without chilling, germination started at 5 to 7 days in the control (0.0 MPa) moisture stress level treatment, at 7 to 8 days in the -0.3 MPa treatment and

Table 3. Pairwise comparisons (Tukey's test) of germination capacity of four provenances (elevations) of *P. sylvestris* from four elevations in Norway.

Provenance	Norway A1	Norway A2	Norway A3
Norway A2	***		
Norway A3	***	***	
Norway A4	***	***	NS

Provenances are shown in order of decreasing elevation. NS, not significant; *, ***, significant $p < 0.05$ and $p < 0.001$, respectively.

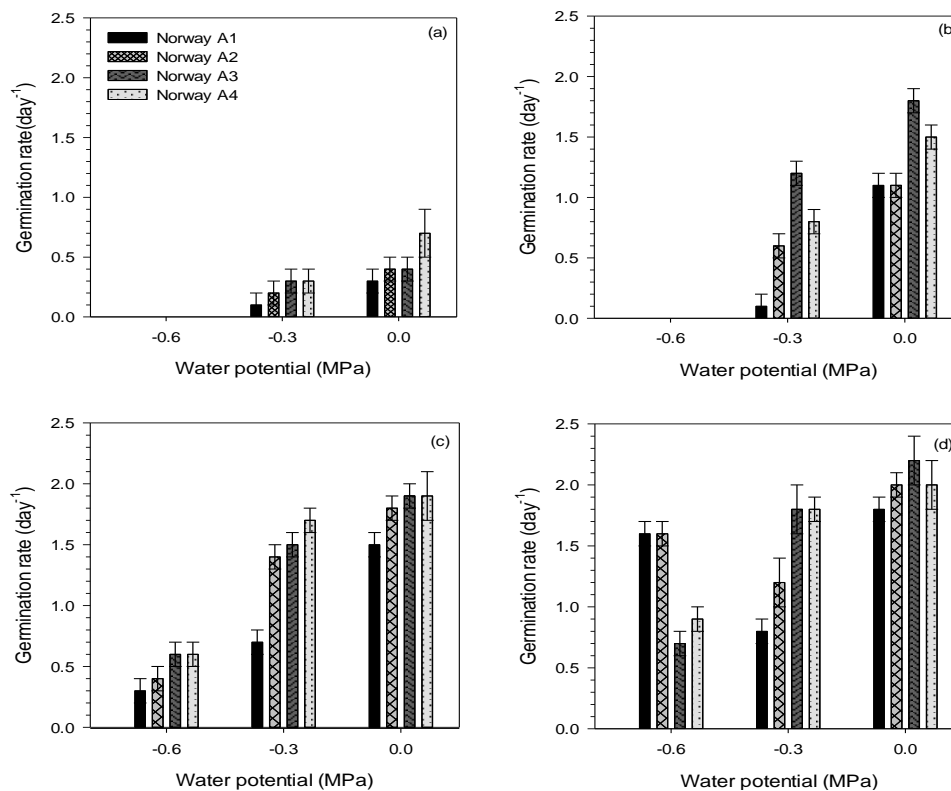


Figure 2. Germination rate at (water potentials of 0.0, -0.3 and -0.6 MPa in four provenances (elevations) of *P. sylvestris* from Norway: (a) zero weeks chilling, germination at 10°C; (b) four weeks chilling, germination at 10°C; (c) zero weeks chilling, germination at 20°C and (d) four weeks chilling, germination at 20°C.

at 16 to 18 days in the -0.6 MPa treatment (Figure 4a to c). After four weeks chilling, germination started at 3 to 4 days in the control, 4 to 6 days in the -0.3 MPa treatment and at 6 to 10 days in the -0.6 MPa treatment (Figure 4d and f). The difference in the start of germination between moisture stress treatments was greatest for the provenance from the lowest elevation (Norway A4).

France: Germination capacity

Germination capacity was generally higher in the provenances from lower elevations after chilling for four

weeks at 4°C, at lower moisture stress levels (water potentials of 0.0 and -0.3 MPa) and at 20°C. Without chilling and at 10°C germination capacity ranged between 0 and 41%, and was highest in the lower elevation (France A3 and A4) provenances. After four weeks chilling and at 10°C, germination capacity ranged between 0 and 87.5%, and was also highest in the provenances from the lower elevation (France A3 and A4) (Figure 5a and b). Without chilling and at 20°C, germination capacity ranged between 0 and 94%, and was highest in the lower elevation (France A3 and A4) provenances. After four weeks chilling and at 20°C germination capacity ranged between 19.5 and 97.5%,

Table 4. Analysis of variance of germination rate of four provenances (elevations) of *P. sylvestris* from four elevations in Norway after two chilling durations, at three moisture stress levels and at two temperatures.

Source of variation	d.f	F-value	P-value
Provenance	3	46.921	***
Chilling	1	559.712	***
Moisture stress	2	943.830	***
Temperature	1	2647.172	***
Provenance × chilling	3	7.356	NS
Provenance × moisture stress	6	51.513	***
Provenance × temperature	3	5.891	**
Chilling × moisture stress	2	25.547	***
Chilling × temperature	1	26.499	***
Temperature × moisture stress	2	4.078	*
Provenance × chilling × moisture stress × temperature	23	25.271	***
Error	144		
Total	191		

NS, not significant; *, ***, significant $p < 0.05$ and $p < 0.001$, respectively.

Table 5. Pairwise comparisons (Tukey's test) of germination rate of four provenances (elevations) of *P. sylvestris* from four elevations in Norway.

Provenance	Norway A1	Norway A2	Norway A3
Norway A2	***		
Norway A3	***	***	
Norway A4	***	***	NS

Provenances are shown in order of decreasing elevation. NS, not significant; *, ***, significant $p < 0.05$ and $p < 0.001$, respectively.

and was again highest in the lower elevation (France A3 and A4) provenances (Figure 5c and d).

Germination at a water potential of -0.6 MPa only occurred at 20°C and there was very little germination without chilling. Germination decreased as moisture stress increased and was completely inhibited at a water potential of -0.9 MPa. Analysis of variance showed that there were significant ($P < 0.001$) differences in germination capacity among provenances, chilling durations, moisture stress levels, and germination temperatures (Table 6). There were significant interactions between all factors except between provenance and chilling and between provenance and temperature (Table 6).

The provenance from the highest elevation (France A1) was significantly different from the other three provenances, which were not significantly different from each other except between France A2 and France A4 (Table 7).

France: Germination rate

Germination rate was generally higher in provenances

from lower elevations, after four weeks chilling at 4°C , at lower levels of moisture stress (water potentials of 0.0 and -0.3 MPa) and at 20°C . Without chilling and at 10°C germination rate ranged between 0 and 0.8 day^{-1} , and was highest in the lower elevation (France A2, A3 and A4) provenances. After four weeks chilling and at 10°C germination rate ranged between 0.0 and 1.9 day^{-1} , and was highest in the provenances from lower elevations (France A3 and A4) (Figure 6a and b). Without chilling and at 20°C , germination rate ranged between 0.0 and 2.1 day^{-1} , and was highest in the provenances from the lower elevations (France A3 and A4). After four weeks chilling and at 20°C germination rate ranged between 0.4 and 2.2 day^{-1} , and was also highest in the provenances from the lower elevations and the lowest elevation (France A3 and A4) provenances (Figure 6c and d). Germination rate was significantly ($P < 0.001$) different among provenances, chilling durations, moisture stress levels and germination temperatures (Table 8).

Results of pairwise comparisons between provenances are shown in Table 9. The provenance from the highest elevation (France A1) was significantly different from the other three provenances, which were not significantly different from each other except between France A2 and

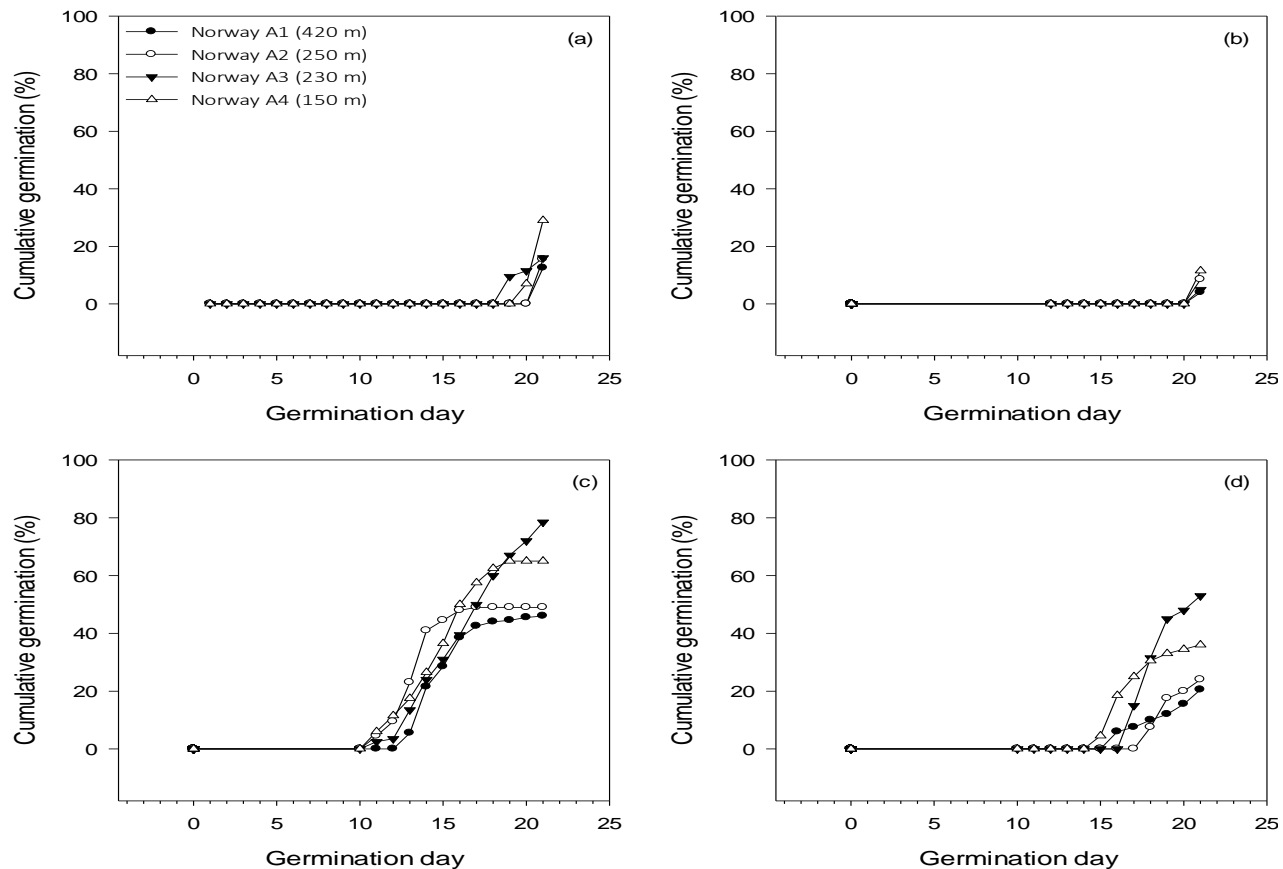


Figure 3. Cumulative germination of four provenances (elevations) of *P. sylvestris* from Norway at 10°C after two chilling durations (zero and four weeks) and at two moisture stress levels (water potentials of 0.0 and -0.3 MPa): (a) zero weeks, 0 MPa; (b) zero weeks, -0.3 MPa; (c) four weeks, 0 MPa and (d) four weeks, -0.3 MPa.

A4.

France: Cumulative germination

Without chilling, germination started at 16-18 days in the control (0.0 MPa) moisture stress level treatment and at 19-21 days in the -0.3 MPa treatment. The provenance from the highest elevation (France A1) did not germinate at all in the -0.3 MPa treatment (Figure 7a and b). After four weeks of chilling germination started at 7 to 10 days in the control treatment and at 12 to 16 days in the -0.3 MPa treatment (Figure 7c and d). The difference in the start of germination between moisture stress treatments was greatest for the provenance from the lowest elevation (France A4).

Cumulative germination of four provenances from France at 20°C after two chilling durations (zero and four weeks) and at three water stress levels (water potentials of 0.0, -0.3 and -0.6 MPa) is shown in Figure 8. Without chilling, germination started at 4 to 5 days in the control (0.0 MPa) treatment and at 6 to 9 days in the -0.3 MPa treatment (Figure 8a and b). Only the France A2 provenance germinated in the -0.6 MPa treatment,

starting on day 11 (Figure 8c). After four weeks chilling, germination started at three days in the control, at 4-5 days in the -0.3 MPa treatment and at ten days in the -0.6 MPa treatment (Figure 8d to f). There were no obvious differences in response to moisture stress levels among provenances.

DISCUSSION

The results reported here clearly demonstrate both variation in germination behaviour among provenances from different elevations and the effect of moisture stress on germination of seeds. Seeds from lower elevations had a higher germination capacity and rate than those from higher elevations, as observed for seeds of some high-elevation woody species studied by Meyer and Monsen (1991). Seeds from low elevations may germinate more rapidly and have a higher germination capacity because they are less dormant than seeds from high elevations (Pratap and Sharma, 2010). However, according to Fernández-Pascual et al. (2013), seeds from high elevations may germinate better and faster because of their reduced dormancy compared with seeds from low

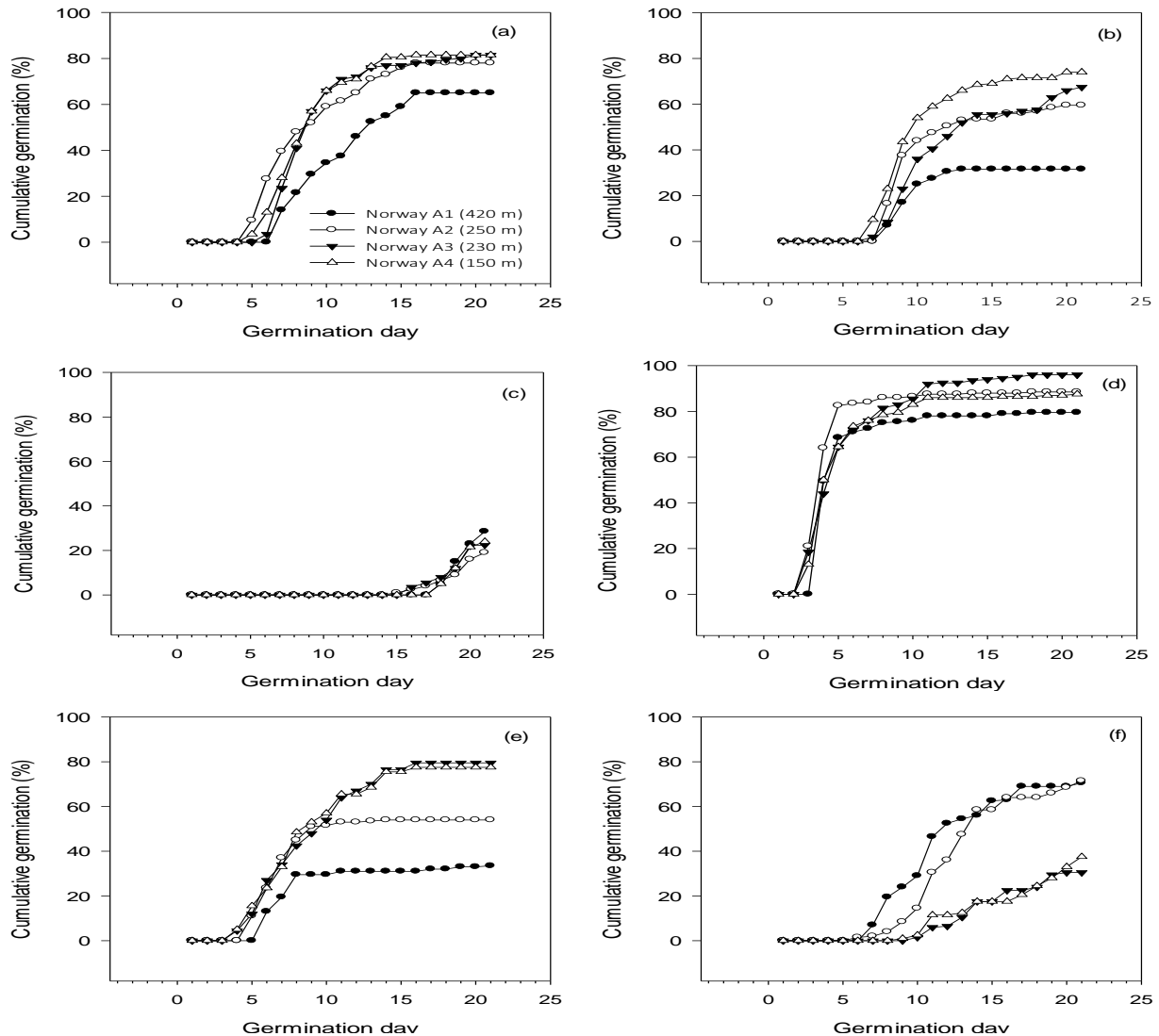


Figure 4. Cumulative germination of four provenances (elevations) of *P. sylvestris* from Norway at 20°C after two chilling durations (zero and four weeks) and at three moisture stress levels (water potential of 0.0, -0.3 and -0.6 MPa): (a) zero weeks, 0 MPa; (b) zero weeks, -0.3 MPa; (c) zero weeks, -0.6 MPa; (d) four weeks, 0 MPa; (e) four weeks, -0.3 MPa and (f) four weeks, -0.6 MPa.

elevations, as is the case for *Calluna vulgaris*, *Erica cinerea* and *Erica vagans*. Additionally, Vera (1997) argued that seeds from higher elevations might receive more chill units than seeds from lower elevations due to the decrease in temperature as elevation increases, resulting in faster germination and higher germination capacity. Vera's (1997) contentions are not in agreement with findings of this experiment, in which higher-elevation provenances showed delayed and poorer germination than lower-elevation provenances. These results agree with the findings of Falusi et al. (1983), who reported a decrease in germination capacity of *Pinus brutia* seed with increases in moisture stress. Falusi et al. (1983) further observed that in *Pinus halepensis* an increase in moisture stress to -0.2 MPa lowered germination capacity

considerably, while at a water potential of -0.8 MPa germination was reduced to approximately 25%. Ne'eman et al. (1999) showed that germination of *Rhus coriaria* was reduced by 80% at a water potential of -0.26 MPa. In studies on *Pinus nigra* by Topacoglu et al., (2016), increasing water stress also reduced germination, while Zhu et al. (2006) reported that increasing water stress decreased germination rate in *Pinus sylvestris*. Gholami et al. (2010) obtained similar results for four wild almond species and concluded that PEG prevents water uptake by seed tissues. According to Ahmadloo et al. (2011), the major reason for reduced and delayed germination under conditions of moisture stress is an increase in the length of the lag phase between imbibition and radicle emergence, since the increase in seed water

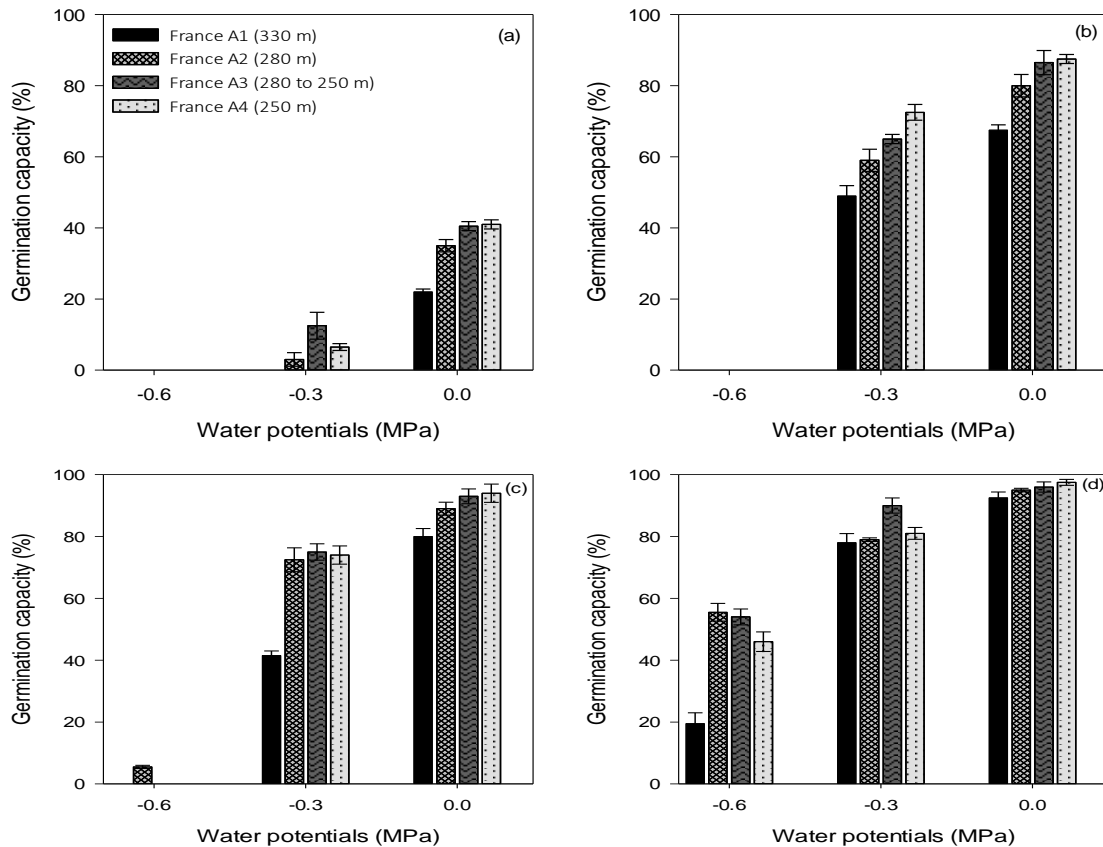


Figure 5. Germination capacity at water potentials of 0.0, -0.3 and -0.6 MPa in four provenances (elevations) of *P. sylvestris* from France: (a) zero weeks chilling, germination at 10°C; (b) four weeks chilling, germination at 10°C; (c) zero weeks chilling, germination at 20°C and (d) four weeks chilling at 20°C.

Table 6. Analysis of variance of germination capacity of four provenances (elevations) of *P. sylvestris* from four elevations in France after two chilling durations, at three moisture stress levels and at two temperatures.

Source of variation	d.f	F-value	P-value
Provenance	3	114.711	***
Chilling	1	2245.853	***
Moisture stress	2	4069.718	***
Temperature	1	3088.410	***
Provenance x chilling	3	1.441	NS
Provenance x moisture stress	6	9.502	***
Provenance x temperature	3	7.333	***
Chilling x temperature	1	56.009	***
Chilling x moisture stress	2	109.413	***
Moisture stress x temperature	2	80.819	***
Provenance x chilling x moisture stress x temperature	23	57.690	***
Error	144		
Total	191		

NS, not significant; *, ***, significant $p < 0.05$ and $p < 0.001$, respectively.

content proceeds slowly during this period. However, at 20°C and a water potential of -0.3 MPa, most (> 70%) of

the seeds in the current experiment germinated, showing that seeds can germinate when subjected to some

Table 7. Pairwise comparisons (Tukey's test) of germination capacity of four provenances (elevations) of *P. sylvestris* from four elevations in France.

Provenance	France A1	France A2	France A3
France A2	***		
France A3	***	NS	
France A4	***	***	*

Provenances are shown in order of decreasing elevation. NS, not significant; *, ***, significant $p < 0.05$ and $p < 0.001$, respectively.

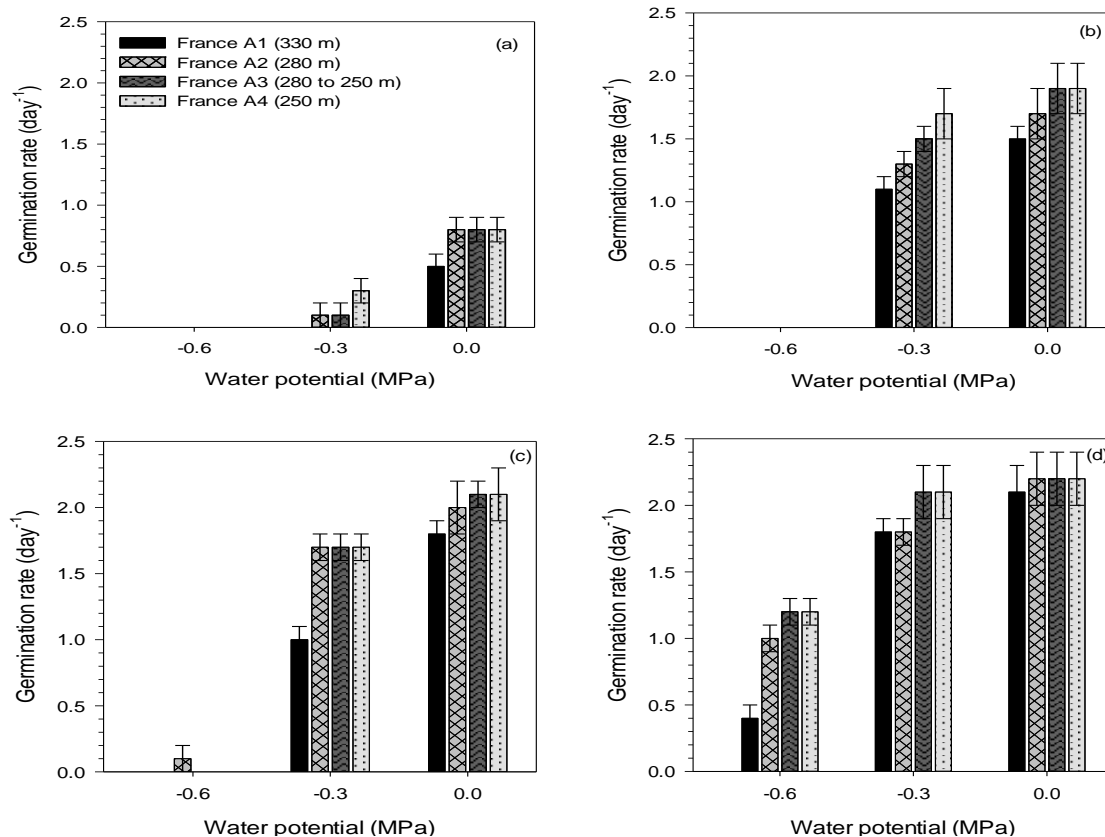


Figure 6. Germination rate at (water potentials of 0.0, -0.3 and -0.6 MPa in four provenances of *Pinus sylvestris* from Norway: (a) zero weeks chilling, germination at 10°C; (b) four weeks chilling, germination at 10°C; (c) zero weeks chilling, germination at 20°C and (d) four weeks chilling at 20°C.

moisture stress.

Research by Mataruga et al. (2010) on five provenances of *Pinus nigra* showed that seeds coming from harsher environments took up lower amounts of water than those coming from environments that are more suitable for the species, suggesting local adaptation and the development of well-adapted land races. However, during the germination stages high-elevation provenances are vulnerable to changes in the physical environment, and selection by soil moisture may be important. High-elevation provenances typically germinate soon after snowmelt in the year after seed production

(Körner, 2013). Following germination early in the growing season, seedlings are more likely to experience favourable soil moisture levels and temperatures, and a growing season long enough to allow accumulation of reserves before the onset of winter. Intraspecific differences in moisture stress tolerance may be associated with environmental differences among sites where populations are found, and are often interpreted in terms of responses to ecological factors (Tilki and Huseyin, 2007). The growing season is shorter and temperatures are lower at high elevations than at low elevations. Faster germination may facilitate the

Table 8. Analysis of variance of germination rate of four provenances (elevations) of *P. sylvestris* from four elevations in France after two chilling durations, at three moisture stress levels and at two temperatures.

Source of variation	d.f	F-value	P-value
Provenance	3	68.477	***
Chilling	1	1475.202	***
Moisture stress	2	2734.137	***
Temperature	1	2204.311	***
Provenance × chilling	3	1.973	NS
Provenance × moisture stress	6	5.320	***
Provenance × temperature	3	4.642	**
Chilling × temperature	1	46.500	***
Chilling × moisture stress	2	81.234	***
Moisture stress × temperature	2	72.277	***
Provenance × chilling × moisture stress × temperature	23	36.296	***
Error	144		
Total	191		

NS, not significant; *, ***, significant $p < 0.05$ and $p < 0.001$, respectively.

Table 9. Pairwise comparisons (Tukey's test) of germination rate of four provenances (elevations) of *P. sylvestris* from four elevations in France. Provenances are shown in order of decreasing elevation.

Provenance	France A1	France A2	France A3
France A2	**		
France A3	***	NS	
France A4	***	***	NS

Provenances are shown in order of decreasing elevation. NS, not significant; *, ***, significant $p < 0.05$ and $p < 0.001$, respectively.

emergence, establishment, growth and survival of seedlings at high elevations (Mondoni et al., 2012). Higher germination at 20°C than at 10°C favours germination late in spring, when temperatures are higher and there is a lower probability of late-season frosts that could damage new seedlings. Pre-chilled seeds from low-elevation provenances do not show much difference in germination between 20 and 10°C, suggesting that seeds from lower-elevation provenances could germinate at lower temperatures, resulting in earlier seedling establishment and allowing the development of a root system appropriate for surviving summer drought conditions. Earlier germination over a wider range of environmental conditions may confer a greater chance of successful establishment of natural regeneration. Conversely, slow germination could help seeds avoid short periods favorable for germination within a long period unsuitable for seedling growth. Additionally, seeds that do not germinate in the presence of water stress can help the population to spread germination over the year.

Numerous studies have shown that germination patterns reflect the seasonal and climatic environments which occur naturally during the germination period

(Dwyer and Erickson, 2016). The higher germination capacity shown by provenances of *P. sylvestris* from low elevations seems to be adaptive. At low elevations winters are relatively mild, with short periods of snow cover. Hence, the main risk of seedling death is from moisture stress as summer progresses. Seeds of all provenances, whether chilled or not, reached a very low germination capacity when incubated at 10°C, possibly because this temperature is close to base temperature and the thermal time accumulated by the end of the incubation period was not enough to permit the germination of most of the provenances. Additionally, the low germination capacity may suggest that seeds had not completely broken their primary dormancy. Reduction in germination with increased moisture stress suggests that moisture levels in the seeds were below the base water potential, which may lead to degradation and inactivation of essential hydrolytic and other groups of enzymes (Wilson, 1971). These results are consistent with suggestions that when water potential is reduced, germination will be delayed or prevented depending on the extent of its reduction (Boydak et al., 2003). The results revealed some differences, which were not tested

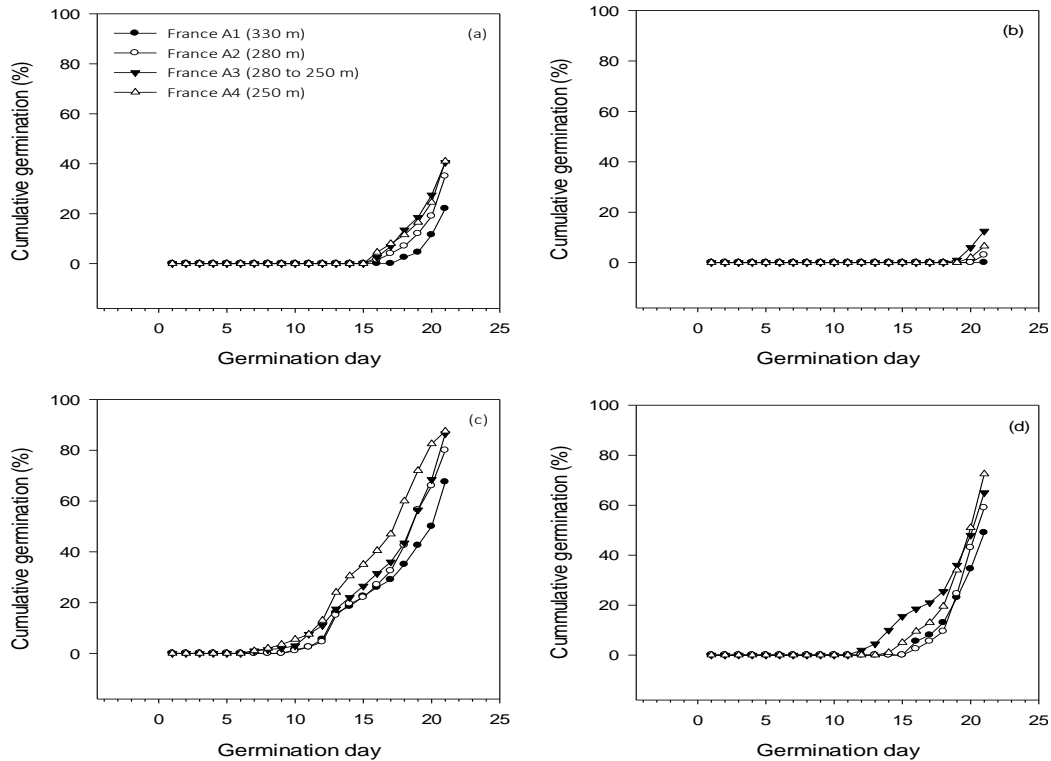


Figure 7. Cumulative germination of four provenances (elevations) of *Pinus sylvestris* from France at 10°C after two chilling durations (zero and four weeks) and at two moisture stress levels (water potentials of 0.0 and -0.3 MPa): (a) zero weeks, 0.0 MPa; (b) zero weeks, -0.3 MPa; (c) four weeks, 0.0 MPa and (d) four weeks, -0.3 MPa.

statistically, between seeds from Norway and France. Germination of both Norwegian and French seeds tended to decrease with an increase in elevation of provenance. At the highest moisture stress level at which germination occurred (-0.6 MPa), Norwegian provenances were more tolerant than French provenances. These findings suggest that in drier climates the chances of seedling survival are increased by delaying germination until suitable moisture conditions are present for seedling establishment. They also suggest that germination of France provenances is more sensitive to declining moisture levels than germination of Norwegian provenances. Germination success of provenances from higher elevations in France was significantly reduced at high moisture stress levels. This is concerning, as climate change predictions are for drier conditions at higher elevation locations in France in the future, suggesting that natural regeneration may be at risk. However, according to Holtmeier and Broll (2007), pine species are migrating to higher elevations and pine recruitment around old seed trees appears to be promoted by climatic warming. The importance of water, temperature and their interaction on *P. sylvestris* germination is clear. Low germination at 10°C suggests that relatively high temperatures may be critical for success of germination during periods of low water potential, for example late

spring and early summer. This experiment has demonstrated that *P. sylvestris* provenances from wet and dry regions (Norway and France) can germinate at various temperature and moisture stress levels, but that a temperature of 20°C and water potentials below -0.9 MPa (for Norwegian provenances) and -0.6 MPa (for provenances from France) are needed to reach high levels of germination. The low elevation provenances (Norway A3 and A4, France A3 and A4) were the most moisture-stress-tolerant provenances. The highest-elevation provenances Norway A1 and France A1 were the least tolerant of moisture stress. Seeds from low elevations which have high tolerance of moisture stress play an important role in the regeneration of this species in a future with decreased amounts of precipitation. In order to refine the selection of provenances offering the best potential under these conditions, it would be useful to continue investigations on the tolerance of moisture stress to the seedling stage, since establishment also depends on the ecological conditions prevailing at the time of seedling emergence.

Conclusion

Germination is one of the most critical periods in the life

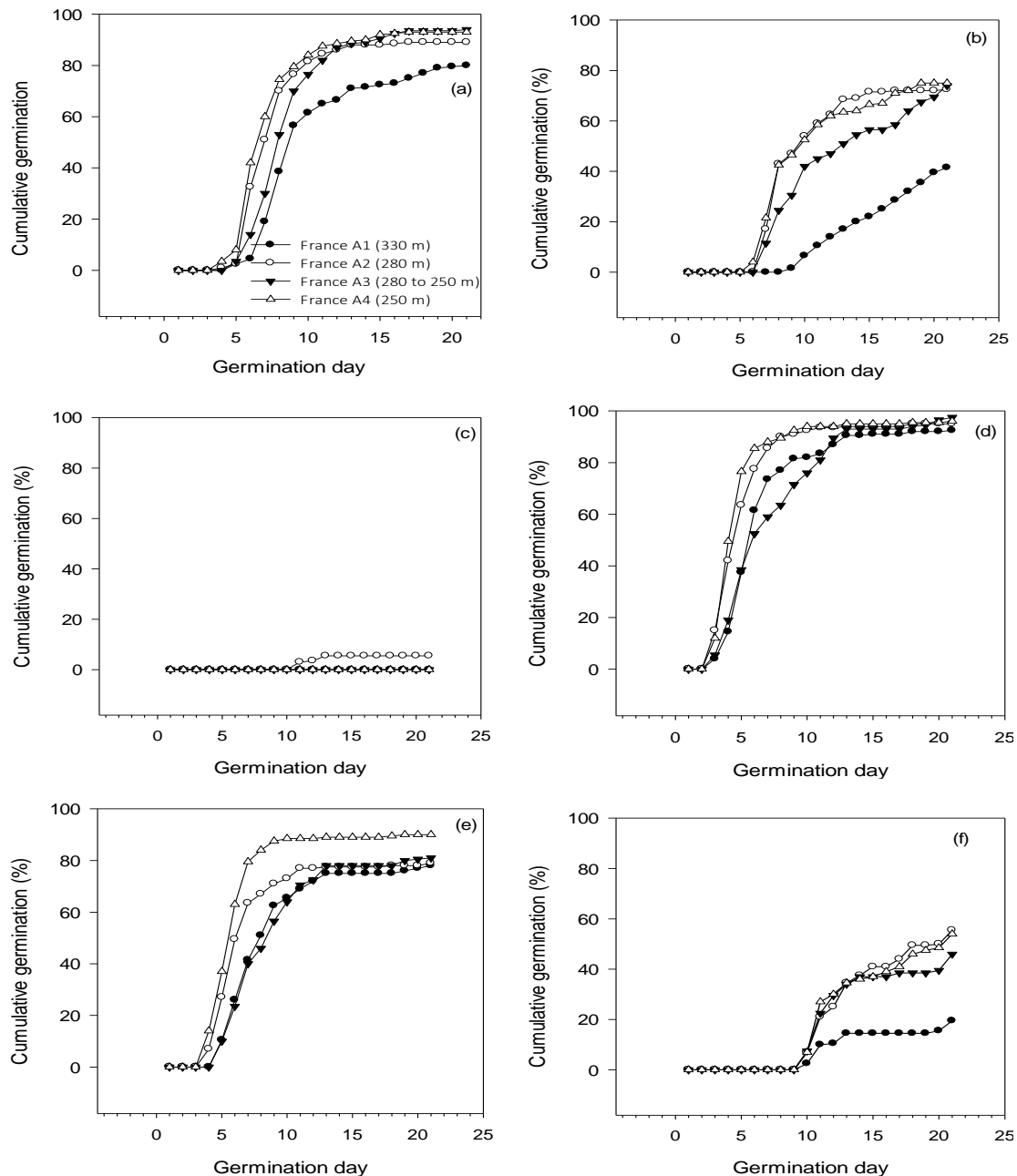


Figure 8. Cumulative germination of four provenances (elevations) of *Pinus sylvestris* from France at 20°C after two chilling durations (zero and four weeks) and at three moisture stress levels (water potentials of 0.0, -0.3 and -0.6MPa): (a) zero weeks and 0.0 MPa; (b) zero weeks and -0.3 MPa; (c) zero weeks and -0.6 MPa; (d) four weeks and 0.0 MPa; (e) four weeks and -0.3 MPa and (f) four weeks and -0.6 MPa.

cycle of plants. Under water stress, low water potential is a determining factor inhibiting seed germination. Water stress delays the beginning of germination of *P. sylvestris* seeds in relation to the control treatment. Provenances from high elevations were the most susceptible to moisture stress. These results provide useful data that will facilitate the development of strategies for the selection of provenances that are more tolerant towards water stress in a changing climate.

CONFLICT OF INTEREST

The author has not declared any conflict of interest.

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Review

Current distribution, regeneration and management practice of *Hagenia abyssinica* in different agroforestry systems of Ethiopia: A review

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***Hagenia abyssinica* is a multipurpose tree, which is sparsely distributed in the highland of Ethiopia. It is used for illness remedy, furniture, fuel, and as a soil additive. This review provides information on the present status and management practice of *H. abyssinica* in Ethiopia to give urgency care other than that given to introduced species for future conservation. The foliage and flowers have a higher quality of N and P for managing soil fertility. However, market availability is diminished. Its healing potential is widely accepted within the society and it is one of the dominant plants in the Boda Montane and Gole Forest. The plant in Hararge of Ethiopia is highly threatened through settlement changes and agriculture expansion. The Gemechis and Menagesha Amba Mariam forest is comprised of seedlings and saplings, but the Chilimo and Gole Natural Forest shows poor regeneration. The Doshke and Gedo forest is represented by aged trees; and the species is under risk. In some areas of Kofele, Bale, Holeta, Legambo and Debark of Ethiopia, it is cultivated in the homegardens for multiuse. Therefore, the diversity is highly shrinking and urgent actions will be needed to conserve it.**

Key words: Conservation, forest, *Hagenia abyssinica*, homegarden, regeneration, threatened.

INTRODUCTION

Hagenia abyssinica (Bruce) J.F. Gmel. is a multiuse tree that belongs to the genus *Hagenia* and family Rosaceae. It is one of the Afromontane endemism tree plants in Ethiopia. However, its abundance and distribution are highly reduced by anthropological factors and the tree is sparsely distributed in Montane Africa (Feyissa et al., 2005). It is found in Sudan, Ethiopia, Malawi, Zambia, Zimbabwe, Tanzania, Kenya, Uganda, Burundi, Rwanda and Democratic Republic of Congo (Tariku, 2018). The

tree was once abundant in the semi-humid mountain woodlands of Ethiopia with an altitudinal range between 2,450 and 3,250 m.a.s.l (Feyissa et al., 2005). According to Ayele et al. (2017), the species were growing within an altitudinal range of 1,850 to 3,700 m.a.s.l. It plays a great role for the economic, ecological, and social aspects, serving as a source of medicine, construction, furniture, income generation, fencing, Bee and animal fodder, fuel wood, and soil fertility (Akale et al., 2019).

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Table 1. Macronutrient composition of the foliage, flower, and stem in *Hagenia abyssinica*.

Foliage (mg g ⁻¹ dry matter)		Flower bud (mg g ⁻¹ dry matter)		Stem (mg g ⁻¹ dry matter)	
N	30.07	N	27.20	N	12.94
P	3.71	P	4.54	P	1.54
K	21.22	K	22.04	K	16.05
Ca	9.69	Ca	5.54	Ca	8.57
Mg	2.38	Mg	2.53	Mg	1.68
S	2.03	S	2.70	S	1.50
Ratios of C, lignin and soluble phenolics to N (mg g⁻¹)					
Foliage			Flower bud		
C:N		15.49	C:N		17.99
Lignin:N		1.79	Lignin:N		2.84
Soluble phenolics:N		5.71	Soluble phenolics:N		8.80
Soluble phenolics + lignin:N		7.50	Soluble phenolics + lignin:N		11.64

Source: Kindu et al. (2008).

The enormous significance of the species for these purposes leads to its being threatened in the country due to over-exploitation (Bekele and Reddy, 2014). The main aim of this review was to give brief information on the present status of *H. abyssinica* in different agroforestry systems of Ethiopia to give attention to the urgency of its conservation beyond that of introduced species.

ECOLOGICAL SIGNIFICANCE OF *H. ABYSSINICA*

The species is mostly used for soil conservation and as reported by Kiros et al. (2016), who also indicated that the translocation rates of major nutrients in the green leaves of *Hagenia* trees were in the order of N > P > K (56 to 61% for N, 54 to 59% for P, and 31 to 42% for K) (Table 1). This shows that the concentration of N and P is higher in the green leaves and C to N ratio was highest in the foliage (Kindu et al., 2008). This reveals that the species has high quality of foliage and flower bud for managing soil fertility. The plant is comprised of large crowns that continually shed leaves; and it is easily decomposable. Similarly, Akale et al. (2019) observed a high amount of leaf foliage and shedding at the base of the plants, which can act as a soil additive in Legambo district of South Wollo, Ethiopia. Therefore, it is advisable to apply green leaves in order to improve soil quality. However, the report by Cronin et al. (2013) stated that the land covered by *H. abyssinica* at Jeldu (Blue Nile basin) transitioned too sparsely with a spread and rapid expansion of *Eucalyptus globulus*. Therefore, the government gives priority for afforestation of the species to recover the previous ecology.

COMMERCIAL STATUS OF *H. ABYSSINICA*

According to Akale et al. (2019), female informants gave

high value for *H. abyssinica* due to its preferred marketable value, which accounted for around 1500 (ETB) Birr per single stem. Helmut et al. (2014) reported that each seller sold an average of 2.9 taenicides (*H. abyssinica*) in 1973, but the extent diminished to 0.2 in 2014 at Merkato, Addis Ababa (capital city of Ethiopia). In 2010 at Bale, a quarter of kilo-dried flowers were sold for one Ethiopian Birr and also used quintals to exchange, which were sold with a range of 60 to 100 Ethiopian Birr per quintal (Assefa et al., 2010). Lulekal et al. (2013) reported that a jug of inflorescence was sold for four Birr in Ankober District, North Shewa Zone of Amhara, Ethiopia. As indicated by farmers in the study of Soromessa and Kelbessa (2014), the seeds of the plants were purchased for 12 Birr per kg, which is a low price when compared to: *Juniperus procera* (50 Birr/kg), *Podocarpus falcatus* (20 Birr/kg), and all are more expensive than *Prunus africana* (7 Birr/kg) at Chilimo Forest of Central Ethiopia. According to Ethiopia Forestry Research Center (IBC, 2012), 9,004,788 *H. abyssinica* seedlings were grown from 61.2 kg of seeds within a period of five years (2006/2007-2010/2011). Results in all the study sites showed that the medical value of the species is still available in the national and local markets for a low price, but the amounts are reduced due to overutilization of the plant in the past and present time.

MEDICINAL VALUE OF *H. ABYSSINICA*

Many people of the countries of Africa, Asia, and Latin America depend on medicinal plants to meet some of their primary health care needs (Bekele and Reddy, 2014). According to Assefa et al. (2010), more than 80% of the Ethiopian people are reliant on plants for their health service. Many studies confirm that the species are used to treat several human and livestock ailments (Assefa et al., 2010; Tariku, 2018). Similarly, Assefa et al.

(2010); Reta (2013); Bekele and Reddy (2014) and Atnafu et al. (2018) confirming that all parts of the species (Bark, Leaf, Flowers, Seeds, Fruits, and Wood) were used to treat ailments by the local people. In Selale (North Shoa, Ethiopia), the society used the fruit and seeds to treat tapeworm (Reta, 2013). Mekuria et al. (2018) reported that extractions of flowers and leaves were used to treat hepatotoxicity, diarrhea, gastritis, and optic atrophy. Currently, many traditional healers used the species to treat ailments in the countryside (Table 2).

In the study of Mekuria et al. (2018) at a Teaching Hospital in Gondar of Ethiopia, 26.9% of 240 respondents used the species as a traditional remedy. Similarly, in the Ankober (North Shewa Zone, Ethiopia), 93.75% of the total respondents used the plant as a remedial agent and it provided the best healing potential in the area (Lulekal et al., 2013). Assefa et al. (2010) stated that 43% of the respondents in Bale, 40% in Kofele and 30% in Debark used self-made remedies and it is widely accepted within the society for its hygienic effect. Now, most societies in all of the study areas are utilizing case remedy. However, many studies have shown that the tree is shrinking in occurrence, and is over exploited, through various anthropogenic impacts, due to the practical usefulness of the species in its many forms.

CURRENT POPULATION OF *H. ABYSSINICA* AND ITS CONSERVATION STATUS

In the Gole Forest, *H. abyssinica* is a highly dominate plant, next to *Juniperus procera*, *Myrsine melanophloeos* and *Hypericum revolutum*; each with an importance value index of 78.2, 41.3, 31.1 and 29.1, respectively (Hailemariam and Temam, 2018). The large basal area (8.8 m²/ha) of *H. abyssinica* contributed to its status as a high relative dominance species (17.9), next only to *Juniperus procera* (33.3) (Hailemariam and Temam, 2018). Therefore, it is one of the most dominant and ecologically important tree species in the Gole Forest. Similarly, at Boda dry evergreen mountain forest, West Shewa of Ethiopia, *H. abyssinica* with *Podocarpus falcatus* and *Juniperus procera* are the dominant dry evergreen trees in the montane forest (Fikadu and Melesse, 2014). The published report for Legambo District (Chiro Kebele) South Wollo, Ethiopia by Akale et al. (2019) revealed that *Hagenia* is the dominant native tree species next to *J. procera*.

However, in the Hararge Area of Eastern Ethiopia, *H. abyssinica* was highly threatened and all the respondents classified such indigenous plants under highly vulnerable species (Abdala et al., 2017). As Bekele and Reddy (2014) reported, the current population of the species is declining through settlement changes and land clearing for agriculture. The Borana communities, recalling their childhood times, reported that there were originally plenty of *H. abyssinica* trees in the surrounding forests, but has now become reduced significantly (Bekele and Reddy,

2014). Similarly, in the reports of Getachew and Biruk (2014), the Yeraba Priority State Forest (Amhara Region, Ethiopia) is represented by the least frequent occurrence of *H. abyssinica*; and this implies that this particular species is rare and it may be extinct from the area in the near future. Currently, the diversity and abundance of such endemic plants are highly decreased. According to the result of Wami et al. (2016), the Forest Department and District Agriculture Department in Gedo, West Shewa of Ethiopia, have established a strong tree plantation programme for replenishment of trees such as *J. procera*, *H. abyssinica*, *Rhus glutinosa* and *Cordia africana* in every year in the degraded forest area. Therefore, other areas of the country will use similar tree farm programmes as developed by Gedo, West Shewa of Ethiopia in order to recover the species

Most studies confirm that conservation of the species and its planting programs have been given little attention. However, certain aspects of traditional thought play a significant contribution to the conservation of indigenous trees. For example, in the report of Belay et al. (2014), the farmers in Debark District, northern Ethiopia believed that *H. abyssinica* attracts lightning during the rainy season, thereby preventing lightning strikes of people, livestock, or houses.

CURRENT REGENERATION STATUS OF *H. ABYSSINICA* IN ETHIOPIA

The occurrence of *H. abyssinica* trees were the least recorded species, in the Menagesha Amba Mariam Forest (Central Highlands of Shewa, Ethiopia) compared to the other trees (Tilahun et al., 2015), and it is one of the most important tree species in the area, which was represented by both seedlings and saplings. Similar studies at Gemechis Natural Forest (West Hararge Zone, Oromia, Ethiopia) by Dawud et al. (2018) showed, the presence of seedlings (11.5) and sapling (4.5) indicated they were more numerous than mature or older ones of the species. This showed that *H. abyssinica* has good regeneration status in the Gemechis and Menagesha Amba Mariam Forest than the other studied areas. Similarly, in the homegardens at Legambo district of south wollo Ethiopia both shrubs and trees of the species were recorded by Akale et al. (2019). In some homegardens, also consist of both seedling and sapling stages of growth, thus further revealing its regeneration status.

However, Tegene et al. (2018) revealed that seedlings and saplings of *H. abyssinica* in the Doshke Forest of Chencha, Gamo Gofa Zone of Ethiopia, are absent among the regeneration group. Similarly, Wami et al. (2016) confirm that the species in the Gedo Forest (West Shewa of Oromia, Ethiopia) were not represented by seedlings or saplings. This showed that the aged trees of *Hagenia*, are the only ones of the species represented in the forests, and *H. abyssinica* is under high risk of loss

Table 2. Medicinal value of parts of *Hagenia abyssinica* in the different area of Ethiopia.

Bale (Oromiya National, Regional State, Southeastern Ethiopia)				
Barks	Flowers	Roots	Leaves	Woods
Fever/cough	Intestinal Worms		Diarrhea	
Stomachache	Healing Wound	Stomachache	Typhoid	Stomachache
Cold (bronchitis)	Epilepsy*		Cough	
Livestock disease	Evil eye		Livestock disease	
Kofele (Oromiya National Regional State, Southeastern Ethiopia)				
Bark	Flower	Root	Leaf	Wood
Dermatology	Intestinal Worms	Stomachache	Diarrhea	
Malaria	Hepatitis*	Abdominal pain		Stomachache
Stomachache	Sexual Transmitted Diseases	Throat disease	Livestock disease	
Livestock disease	Harms related to Bile	Cancer*		
Debark (Amhara Regional State, Northern Ethiopia)				
Bark	Flower	Root	Leaf	Wood
Livestock disease	Intestinal Worms	Severe stomach pain	Healing injured	-
Abaya (Borana Zone, Oromia Regional State, Ethiopia)				
Bark	Flower	Root	Leaf	Wood
Fever/ Cough	Intestinal Worms	Stomachache	Diarrhea	
Stomachache	Wound healing	Abdominal pain	Typhoid	Stomachache
Cold/ bronchitis	Epilepsy	Throat disease	Cough	
Dermal diseases	Hepatitis	Cancer	Healing injured	
Hawassa (College of Teacher Education campus, SNNPRS)				
Leaf	Flowers	Seeds		
Tapeworm, Wound	Ascariasis	Tapeworm, Typhoid		

Source: Assefa et al. (2010); Bekele and Reddy (2014); Atnafu et al. (2018)

(Figure 1). Therefore, these situations call for conservation actions through prioritizing. Similarly, as shown by the results of Hailemariam and Temam (2018), the species in the Gole Natural Forest (West Arsi Zone, Oromia Regional State of Ethiopia) exhibited poor regeneration status and the forest includes mainly seedling stages of *H. abyssinica*. Likewise, in the Chilimo Forest of Central Ethiopia, the species were represented by the seedling stage (Soromessa and Kelbessa, 2014). Therefore, both the Gole and Chilimo forests show poor regeneration potential, and the ecosystem is mainly represented by aged trees.

The status of *H. abyssinica* in the Homegardens

According to Akale et al. (2019), 62 individuals of the species were recorded from 32 out of 75 informants in their homegarden at Chiro kebele of Legambo district, South Wollo Ethiopia. In the report of Assefa et al. (2010), 30% in Kofele, 26% in Bale, (Oromiya, Southeastern Ethiopia) and 7% of the informants in

Debark (Amhara, Northern Ethiopia) have planted *H. abyssinica* in their home garden and include transplanted seedlings from nearby forests in their backyards. Nevertheless, the majority of the informants (74% in Bale, 70% in Kofele and 93% in Debark) have not planted this *H. abyssinica* in their homegardens and this fundamental problem is due to the lack of seedlings in their nearby sites. Therefore, the societies in the study areas have endless interest in planting the indigenous tree in their homegardens (Assefa et al., 2010). Similarly, Mekonnen et al. (2014) reported the presence of *H. abyssinica* in the farmer's homegardens of Holeta Town, Oromia National, Regional State, Ethiopia, which are used for medicinal purposes. Therefore, cultivation in the homegardens plays a crucial role in the conservation of the species and the government supports and encourages homegarden cultivation practices.

CONCLUSION

H. abyssinica is highly distributed in the highlands of



Figure 1. Status of Kosso in homegardens at Chiro kebele, South Wollo of Ethiopia (Akale et al., 2019).

Ethiopia such as Boda dry evergreen montane forest, Chilimo, Doshke, Gedo, Gole, Gemechis, Hararge, Yeraba Priority State Forest and Menagesha Amba Mariam forest. The species has a high nutrient quality of foliage and flower buds for managing soil fertility. The shed leaves are easily decomposable and it is advisable to apply the green leaves in order to improve the soil quality. In all the study sites, the medicinal value of *H. abyssinica* is still available in local markets for a low price. Most of the traditional healers used the Bark, Leaf, Flowers, Seeds, Fruit, and Wood of the plant to treat several ailments. The Gemechis and Menagesha Amba Mariam Forest is largely comprised of seedlings and saplings, and has good regeneration status. However, the Doshke, Gedo and Hararge Forests were not described as having seedlings or saplings; and these forests are under risk of loss. The status of *H. abyssinica* in Gole and Chilimo forests is represented largely by seedling stages; and this shows poor regeneration ranks. Thus, the situation calls for conservation measures through prioritization. Some societies in the Kofele, Bale, Debark, Legambo and Holeta Town cultivated the species in their homegardens and seem to have endless interest. Such tasks should be encouraged and there is a need for agricultural experts to help them. However, most of the previous studies confirm that the *H. abyssinica* are under risk in the highlands of Ethiopia and need a high conservation priority in the case of Ethiopia.

RECOMMENDATIONS

According to the above published evidence, because of the diversity and frequency of *H. abyssinica* at the country level it is overexploited and shrinking over time. Formerly, such indigenous species were the dominant tree in the upper Montane forest belt, but now they are only found as scattered trees and restricted in the highlands. This is due to the over dominance of an exotic species (*Eucalyptus* tree) occurring in Ethiopia; and the Agroforestry sectors have given little attention to the indigenous plants. Therefore, the government and Agroforestry sectors have to give high priority for plantation programs to conserve the native species:

- (1) Lowland adaptability techniques of the species should be expected (range shift gaps).
- (2) The highly vulnerable forest should be given priority conservation.
- (3) National and international *ex-situ* conservation gaps of the species should be achievable.
- (4) Homegarden plantations should be given high attention.
- (5) Implementing of *H. abyssinica* nursery sites in each region should be established for the rehabilitation and conservation programs.
- (6) Implementing *H. abyssinica* plantation programs in the country.

- (7) Building tissue culture academic institution for the preservation of the species.
 (8) Finally, people need to be encouraged to have *H. abyssinica* in their gardens.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Floristic composition, vegetation structure and regeneration status of Wabero forest, Oromia Regional State, Southeastern Ethiopia

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Wabero forest is one of the very few remnant moist evergreen montane forests in Southeast Ethiopia. However, basic data on vegetation structure and composition of the forest that could provide valuable information for possible conservation measures is lacking. The main objective of this study, therefore, was to investigate the woody species composition, vegetation structure and regeneration status of the forest. In the study 30 quadrants of 20 m by 20 m were used for woody species data and 5 m by 5 m for sapling and seedling in a nested form. The sample quadrants were laid down along transects at a distance of 300 m from each other. Diameters at breast height (DBH), basal area, frequency and importance value index were used for analysis of vegetation structure. In the study, a total of 47 woody plant species belonging to 43 genera and 33 families were recorded. The major families were Rubiaceae and Rutaceae each represented by four species followed by Sapindaceae, Moraceae and Oleaceae each represented by three species. The rest of the 28 families were represented by one or two species, only. The mean density of all the woody species recorded in the study site was 21,737 ha⁻¹. *Diospyros abyssinica*, *Podocarpus falcatus*, *Strychnos mitis*, *Celtis africana* and *Coffea arabica* were found to be the most dominant and frequent species with the highest importance value index. Results of regeneration status of the forest, which was analyzed by comparing number of seedling and sapling with mature trees, revealed that the forest is under good regeneration status. Therefore, based on the results of the study, awareness creation and implementation of participatory forest management strategy is recommended in order to sustain the forest.

Key words: Basal area, diameters at breast height (DBH), importance value index, Montane forest, woody species composition.

INTRODUCTION

Ethiopia is one of the tropical countries with highest floral diversity (Motuma et al., 2010). Between; 6,000-7,000 species of higher plants are estimated to exist in the country of which about 780-840 (12-13%) plant species are estimated to be endemic (Demel, 2001). However,

these biologically rich resources of Ethiopia are vanishing at an alarming rate due to extensive deforestation. It was mentioned that once upon a time about 35% of Ethiopia's land area was covered with forest (EFAP, 1994). Early 1950's, the area covered by forest in Ethiopia



Figure 1. Map of Wabero forest, Dello-mena district, Oromia.

estimated to be 9 million ha (16% of the land area) and in the 1980's; it was reduced to 3.6% and by 1989 to 2.7% (Gessesse, 2010). Even though several factors contribute for destruction of natural forests in Ethiopia, agricultural expansion is probably the dominant force (Mulugeta and Demel, 2006). This will result in the loss of biodiversity resources along with their habitats (Senbeta and Denich, 2006). Moreover, destruction of forests has an adverse effect on the environment, such as soil degradation, soil erosion and alteration of natural resources. Since forest serves as a source of food, household energy, construction and agricultural material, tourism and recreation values and medicines, its loss would have impact on the socio-economic setup of the communities (Hundera et al., 2007).

Ecological investigation of species composition and vegetation structure of a forest is essential to provide information on type of species of the forests, identify economically and ecologically useful plant species as well as to identify the most threatened ones for management and conservation (Tesfaye et al., 2013). The present study, therefore, stems from this understanding and aimed at studying species

composition and vegetation structure of Wabero forest.

MATERIALS AND METHODS

Description of the study area

The study was conducted on Wabero forest, which is located in Dello-mena district of Bale zone of Oromia Regional National State (Figure 1). The forest is located between 6° 27' 240" to 6° 31' 673" N latitude and 39° 42' 883" to 53° 26' 320" E. Its altitudinal range is between 1500-2500 m above sea level. The mean annual rainfall of the area is 700 mm. The mean annual minimum and maximum temperatures of the area ranges between 21 and 38°C while the mean annual temperature is about 29.5°C.

Sampling design

A systematic sampling technique was used to collect vegetation data in the Forest. In the present study, quadrats of 20 m × 20 m size were laid for woody species and subplots of 5 m × 5 m for seedling and sapling study. The subplots were laid at the four corners and one at the center of the large quadrat. Plots were laid systematically at every 300 m along transect lines, which were 1000 m apart from each other.

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Table 1. Density and percent contribution of five woody species in Wabero forest.

Species name	Density (ha ⁻¹)	%
<i>Strychnos mitis</i>	2521.42	11.60
<i>Diospyros abyssinica</i>	2350.82	10.82
<i>Celtisafricana</i>	2314.10	10.65
<i>Coffea arabica</i>	2093.32	9.63
<i>Podocarpusfalcatus</i>	1882.50	8.44
The rest 46 species	10574.89	48.64
Total	21737.00	100

Data collection

In each quadrat, all plant species were recorded. In addition, woody plant species occurring outside quadrats laid were also recorded only as 'present', but not used in the subsequent data analysis. Plant specimens were identified and deposited at the National Herbarium of Ethiopia (ETH), Addis Ababa University. In each plot, diameters at breast height (DBH) of all woody plant species with DBH \geq 2.5 cm were measured. Individuals having DBH less than 2.5 cm were counted. Diameter tape was used to measure DBH of all the woody plant at 1.3 m above the ground. GPS (Garmin eTrex) was used to take location and altitude data.

Data analysis

Species importance value (IVI) and basal area were used to describe vegetation structure, tree density, frequency, and DBH. DBH was classified into the following 7 classes: Class-1: 2.5-10 cm, Class-2: 10.1-20 cm, Class-3: 20.1-50 cm, Class-4: 50.1-80 cm, Class-5: 80.1-110 cm, Class-6: 110.1-140 cm, and Class-7: >140 cm (Tesfaye et al., 2013). Basal area was calculated using the formula: $BA = \pi (d/2)^2$, where d is diameter at breast height. Density (number of individuals of a species within the sample area) was computed on hectare basis. Frequency calculated by dividing number of plots in which species "n" occurs by total number of sampled plots. In addition, relative density was calculated by dividing density of species "n" by total density of all species multiplied by 100. Relative dominance was calculated by dividing dominance for species "n" over total dominance of all species and multiplied by 100. Relative frequency was estimated by dividing the frequency value for species "n" by total of all frequency values for all species and then multiplied by 100. Finally, Importance Value Index (IVI) was computed as a summation of relative density, relative dominance and relative frequency.

RESULTS AND DISCUSSION

Floristic composition

A total of 47 woody plant species belonging to 33 families were identified in the 30 plots. Among these, families Rubiaceae and Rutaceae were relatively the two most dominant species-rich families, each represented by four species. Sapindaceae, Moraceae and Oleaceae were the second most species-rich families represented by three species. All the other families were represented only by one or two species (Appendix Table 1).

Vegetation structure

For the description of the vegetation structure of Wabero forest woody species whose DBH > 2.5 cm were selected and measured. The total density of woody species of the forest was estimated to be 21.737 ha⁻¹. The five densest species in the forest were *Strychnos mitis* (2521.42 density ha⁻¹), *Diospyros abyssinica* (2350.82 density ha⁻¹), *Celtis africana* (2314.1 density ha⁻¹), *Coffea arabica* (2093.32 density ha⁻¹), *Podocarpus falcatus* (1882.5 density ha⁻¹). These five species contributed to 51.4% and the remaining 46 species together accounted for 10574.89 density ha⁻¹ (48.6%) (Table 1). Among all woody species, *Allophylus abyssinicus* and *Acalypha volkensii* were species with the lowest densities (13.2 and 15.7 individuals ha⁻¹, respectively).

DBH class distribution

The different DBH class distribution of woody species in Wabero forest is given in Figure 2. The number of individuals in DBH class less than 10 cm is 5007 per ha. The number of individuals gradually decreases toward the higher DBH classes as the DBH class size increases. DBH class distribution of all individuals in different size classes showed a reversed J-shape distribution (Figure 2). The current funding suggests the existence of good reproduction and recruitment potential of woody species in Wabero forest. The result is in agreement with that of Tesfaye et al. (2013).

Basal area

The total basal area of Wabero Natural Forest is 88.89 m²ha⁻¹ (Table 2). About 43.06 m²ha⁻¹ (48.43%) of the total basal area was covered by eight tree species: *D. abyssinica* (9.11%), *P. falcatus* (9.49%), *S. mitis* (7.03%), *Mimusops kummel* (6.12%), *Filicium decipiens* (4.34%), *Olea welwitschii* (4.22%), *C. africana* (4.08), and *Galiniera saxifrage* (4.04). However, the remaining 43 species together contributed for 45.83 m²ha⁻¹ (51.57%) of total basal area. The species with the largest contribution

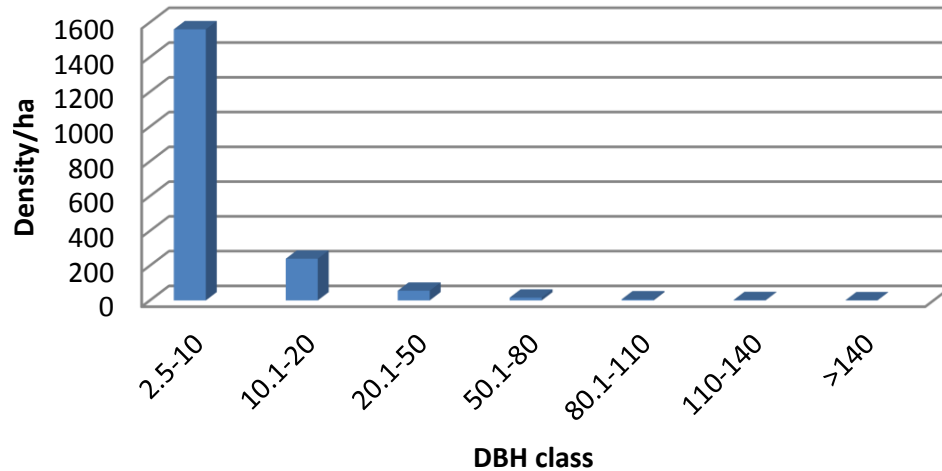


Figure 2. Density of individuals under different DBH classes.

Table 2. The highest basal area (m^2ha^{-1}) of species.

S/N	Scientific name	Basal area (m^2ha^{-1})	%
1	<i>Podocarpus falcatus</i>	8.44	9.49
2	<i>Diospyros mespiliformis</i>	8.09	9.11
3	<i>Strychnos mitis</i>	6.25	7.03
4	<i>Mimusops kummel</i>	5.44	6.12
5	<i>Filicium decipiens</i>	3.86	4.34
6	<i>Olea welwitschii</i>	3.75	4.22
7	<i>Celtis africana</i>	3.63	4.08
8	<i>Galiniera saxifraga</i>	3.60	4.04
9	The remaining 43 species contributed	45.83	51.57
	Total	88.89	

Table 3. The five highest frequented species in study area.

S/N	Scientific Name	Family of species	Relative frequency (%)
1	<i>Celtisafricana</i>	Ulmaceae	6.83
2	<i>Diospyros mespiliformis</i>	Ebenaceae	6.68
3	<i>Coffea arabica</i>	Rubiaceae	6.30
4	<i>Strychnos mitis</i>	Podocarpaceae	6.19
5	<i>Podocarpus falcatus</i>	Loganiaceae	5.87

in basal area can be considered the most important woody species in the forest.

The basal area of Wabero Forest ($88.89 \text{ m}^2 \text{ ha}^{-1}$) is greater than basal area of many forests in Ethiopia. As Tesfaye et al. (2013) reported that the value of Masha Anderacha ($49.80 \text{ m}^2 \text{ ha}^{-1}$), Dindin ($49 \text{ m}^2 \text{ ha}^{-1}$), Alata-Bolale ($53.33 \text{ m}^2 \text{ ha}^{-1}$) and Jibat Natural Forest ($60.9 \text{ m}^2 \text{ ha}^{-1}$). However, basal area of Wabero Natural Forest is less than that of Belete Natural forest ($90.6 \text{ m}^2 \text{ ha}^{-1}$).

Frequency

Frequency is the number of quadrats in which a given species occurred in the study area. Based on the results the five most frequently observed woody species in the forest (Table 3) were *C. africana* (6.83%), *D. abyssinica* (6.68%), *C. arabica* (6.3%), *S. mitis* (6.19%), and *P. falcatus* (5.87%). Moreover, the three lowest percent frequency species were *Trichilia emetic*, *A. abyssinicus*

Table 4. Important Value Index (IVI) of the first five most important woody species.

S/N	Scientific name	RD (%)	RF (%)	RDO (%)	IVI
1	<i>Diospyros mespiliformis</i>	10.87	6.68	9.87	27.40
2	<i>Strychnos mitis</i>	11.68	6.19	7.62	25.40
3	<i>Podocarpus falcatus</i>	8.66	5.87	10.30	24.80
4	<i>Celtis africana</i>	10.58	6.83	4.43	21.80
5	<i>Coffea arabica</i>	9.62	6.30	3.21	19.10

RD=Relative Density, RDO=Relative Dominance, RF=Relative Frequency.

and *A. volkensii* and each constituting less than 0.25% relative frequency. Frequency gives an approximate indication for homogeneity and heterogeneity of vegetation. As Lamprecht (1989) pointed out that high value in high frequency and lower value in the lower frequency classes indicate vegetation homogeneity. Conversely, high percentage of number of species in the lower frequency class and low percentage of number of species in the higher frequency classes indicates high degree of floristic heterogeneity (Simon and Girma 2004).

Species importance value index (IVI)

Based on their IVI value, *D. abyssinica* (27.4), *S. mitis* (25.4), *P. falcatus* (24.8), *C. africana* (21.8) and *C. arabica* (19.1), respectively, were the leading, the highest IVI, and ecologically most significant woody species in Wabero Natural forest (Table 4). These species might also be the most successful species in regeneration, pathogen resistance, growth in shade, and competition with other species, least preferred by animals, least attracted by pollinators and seed predators that facilitate seed dispersal within the existing environmental conditions of the study area. In contrast, the woody species, which exhibited the lowest IVI values (IVI < 1), were *Ficus sycomorus*, *Ficus thonningii* Blume, *Carrisa spindrum*, *Trema guineensis*, *Vernonia amygdalina*, *A. volkensii*, and *A. abyssinicus*. According to Curtis and McIntosh (1951), IVI gives a more realistic figure of dominance from structural point of view. It is also useful to compare the ecological significance of species (Lamprecht, 1989) where a high IVI value indicates that the species sociological structure in the community is high. As a result, ecologists consider IVI as one of the most important tool in a vegetation study (Curtis and McIntosh, 1951).

Population structure of the forest

The study revealed that the woody species recorded from the study site demonstrated more or less the same patterns of population structures. The documented woody species have shown stable population structures

composed of the highest density of individuals at the lowest DBH class followed by gradually declining densities of individuals with increasing DBH classes (Figure 3). Information on population structure of a tree species indicates the history of the past disturbance of that species and the environment; and hence, is used to forecast the future trend of the population of that species. According to Simon and Girma (2004), population structure has significant for sustainable use and conservation of the forest.

Regeneration status of the forest

Regeneration status of the forest was analyzed by comparing seedling and saplings with mature trees according to Dhaukhandi et al. (2008) and Tiwari et al. (2010). In Wabero forest the seedling, sapling, and matured tree accounted for 69, 23, and 8% density ha⁻¹ of individuals, respectively (Figure 4). Density ha⁻¹ of individuals of species showed that the seedling >sapling> matured tree in the forest. According to Dhaukhandi et al. (2008), the density values of seedling and saplings are considered as regeneration potential of the species. Based on the criteria of Dhaukhandi et al. (2008) and Tiwari et al. (2010) the forest of Wabero is categorized among forests with good regeneration potential.

Conclusion

Wabero forest is one of the remnant natural forests in southeastern Ethiopia. It consists of 47 woody species belonging to 33 families. Rubiaceae and Rutaceae were the two most species-rich families. Among the collected woody species, *D. abyssinica*, *P. falcatus*, *S. mitis*, *C. africana* and *C. arabica* were found to be the most dominant and frequent species with the highest importance value index. Results of regeneration status of the forest, which was analyzed by comparing number of seedling and sapling with mature trees, revealed that the forest is in good regeneration status. Therefore, the government, NGO and the community should work jointly in order to save and use the forest resources in a sustainable manner.

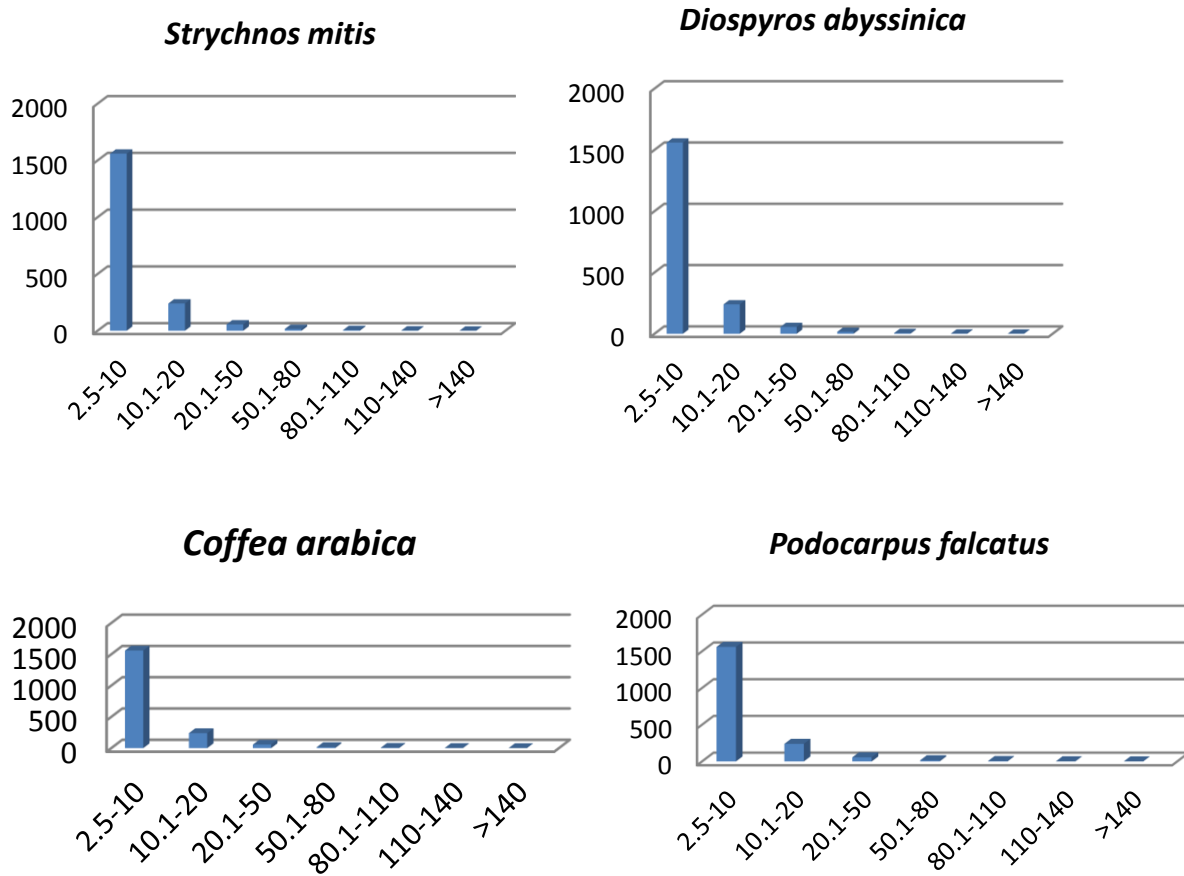


Figure 3. Vegetation structure of five woody species in Wabero forest.

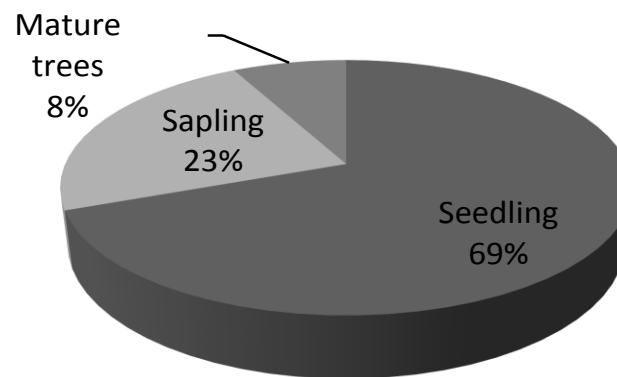


Figure 4. Density ha⁻¹ of seedling, sapling and mature trees.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Appendix Table 1. List of woody plant species in Wabero forest.

Scientific name	Family name
<i>Acalypha volkensii</i>	Euphorbiaceae
<i>Allophylus abyssinicus</i>	Sapindaceae
<i>Apodytes acutifolia</i>	Icacinaceae
<i>Bersama abyssinica</i>	Melanthaceae
<i>Buddleia polystachya</i>	Buddlejiaceae
<i>Calpurina aurea</i>	Fabaceae
<i>Canthiune uryoides</i>	Rubiaceae
<i>Capparis tomentosa</i>	Capparidaceae
<i>Carrisa spindrum</i>	Apocynaceae
<i>Cassipourea malosana</i>	Rhizophoraceae
<i>Celtis africana</i>	Ulmaceae
<i>Citrus aurantium</i>	Rutaceae
<i>Coffea arabica</i>	Rubiaceae
<i>Cordia africana</i>	Boraginaceae
<i>Croton macrostachyus</i>	Euphorbiaceae
<i>Diospyros abyssinica</i>	Ebenaceae
<i>Dobera glabra</i>	Salvadoraceae
<i>Ehretia cyamosa</i>	Boraginaceae
<i>Euclea schinperi</i>	Ebenaceae
<i>Fagaropsis angolensis</i>	Rutaceae
<i>Ficus sur</i>	Moraceae
<i>Ficus sycomorus</i>	Moraceae
<i>Galiniera saxifraga</i>	Rubiaceae
<i>Mimusops kummel</i>	Sapotaceae
<i>Myrsine africana</i>	Myrsinaceae
<i>Mytenus senegalensis</i>	Celastraceae
<i>Ococotea kenyensis</i>	Lauraceae
<i>Olea capensis</i>	Oleaceae
<i>Olea welwitschii</i>	Oleaceae
<i>Olinia rochetiana</i>	Oliniaceae
<i>Podocarpus falcatus</i>	Podocarpaceae
<i>Pouteria adolf-friedericii</i>	Sapotaceae
<i>Poyiscias fulva</i>	Araliaceae
<i>Pruns africanus</i>	Rosaceae
<i>Rhamnus prinoids</i>	Rhamenaceae
<i>Rhus ruspolii</i>	Annacardiaceae
<i>Rothmanniaur celliformis</i>	Rubiaceae
<i>Schrebera alata</i>	Oleaceae
<i>Strychnos mitis</i>	Loganiaceae
<i>Syzigium macrocarpum</i>	Myrtaceae
<i>Teclea nobilis</i>	Rutaceae
<i>Trema guineensis</i>	Ulmaceae
<i>Trichilia emetica</i>	Meliaceae
<i>Vepris dainelli</i>	Rutaceae
<i>Vernonia amygdalina</i>	Astreaceae
<i>Vernonia leopoldi</i>	Astreaceae
<i>Warburgia ugandensis</i>	Canallaceae

Full Length Research Paper

Do internal roadways influence tree diversity and density in a reserve forest? - A reality check at BRT Tiger Reserve, Karnataka, India

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Roadways are inevitable components in any human managed landscape across the world, with the sole objective to facilitate transportation. Reserve forests are not an exception to this situation. A roadway being a potential factor in influencing biodiversity, this study checks the possibility of relationship between the distance of tree communities from roads and the basic characteristics (diversity and density) of the communities in a protected forest. 30 distinct plots were studied across two ranges in BRT Tiger Reserve, Karnataka. For each plot, its average distance and minimum distance from a road and also from specific road types were calculated. In order to check the relationship of distance from road and community characteristics, correlation tests were carried out. It is inferred that the distance from internal roads and community characteristics does not possess any relationship in a reserve forest scenario. Though the possibility of influence by internal roads over tree communities is rendered nil in this study, cautious execution of road-based operations with-in the reserve forests in future is highly recommended.

Key words: Biodiversity conservation, correlation analysis, distance from roads, forest landscapes.

INTRODUCTION

Roads are ingenious creations of humans that make livelihood easier at a large scale. But, on an ecological basis, roads also play a crucial role over determining the state of biodiversity, in several ways. Predominantly, road edges display a variety of physical and chemical changes as a result of road operation and vehicle transports (Forman, 2000; Delgado et al., 2007; Parris and Schneider, 2008; Hoskin and Goosem, 2010) and are characterized by altered micro-climatic conditions, hydrology regimes, soil composition, increased levels of

light and pollutants in the air, soil and water (Jaeger and Fahrig, 2004). Hence, the constraining factors along roadsides, such as alteration of normal hydrological flows, the introduction of chemicals or salts, vehicle exhaust emissions, wind gusts, excessive light, dust, etc., may play a vital role in promoting/demoting the spread of plants along roadsides, by causing variations in the habitat, environment or ecology (Rentch et al., 2005). Roads in general constitute a partial to complete barrier for movement of many organisms, depending on the

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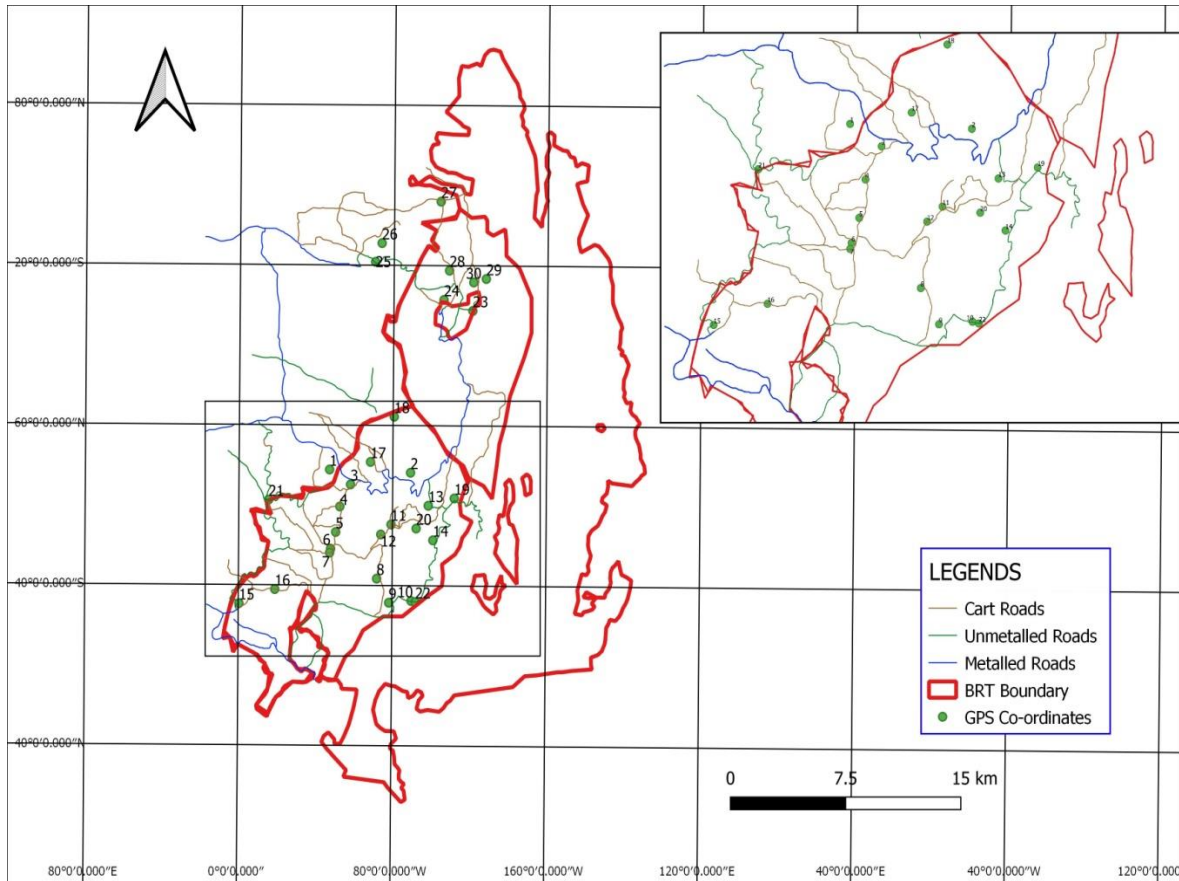


Figure 1. GIS map showing BRT boundary, internal roadways and plots (refer to Table 1).

mobility and behavior of taxa and on road characteristics, influencing changes in plant-animal interactions (Jaeger and Fahrig, 2004). Also, under the influence of possible elevated CO_2 in the roadside vicinities, plants may grow faster and spread efficiently by enhanced photosynthetic activity (Miquel et al., 2004) or through enhanced root formation/activity (Madhu and Hatfield, 2013), leading to variations in leaf or root biomass, respectively. Alternatively, elevated CO_2 might also cause retarded growth of plants, by its negative effect on stomatal conductance/respiration, affecting the transpiration driven mass flow of Nitrogen in the soil (Daniel and Wang, 2008; Uddling, 2015).

Understanding the role of protected areas in conserving biodiversity is a prime objective in conservation biology. Anthropogenic activities within these protected areas, in particular, roads, can alter the spatiotemporal dynamics of biological diversity there. However, our understanding of how the presence and position of roads affects tree communities and paves way for biodiversity conservation is limited. One major positive role played by roads is the facilitation of forest monitoring and biodiversity conservation (M'Woueni et al., 2019). And, low-traffic roads can also lead to increase in habitat heterogeneity

in any structurally poor forest (Salek et al., 2010). Though the overall capacity of roadways is known, its influential potential in a reserve forest scenario is quite unexplored. Hence, this study aims to bring about clarity in the subject, by considering BRT Tiger Reserve. It is a well-protected reserve forest of over 540 km^2 in India, at the confluence of the Western Ghats and the Eastern Ghats mountain ranges.

MATERIALS AND METHODS

Using stratified random sampling protocol in a GIS platform (ArcGIS 10.6.1), 30 distinct spots were marked across two ranges, viz., K. Gudi and Yelandur, in BRT Tiger Reserve, Karnataka. Following standard protocols (Reddy et al., 2018), 0.1 ha plots were established at the derived spots, irrespective of their distance from the roads, and tree inventory data were collected for each plot, during December 2018 to January 2019 (Figure 1).

Later, using GIS (QGIS 3.8), with the help of georeferenced toposheet maps (SOI maps- Karnataka, Tamil Nadu 57H4, 58E1, 58E2, 58E5) and shape file of BRT, distinct lines were obtained for metalled roads, unmetalled roads and cart roads, separately, across the two covered ranges. The GPS points for the plots were laid over as a layer, and their perpendicular distances from the roads were calculated efficiently. For each plot, its total average distance and minimum distance from a road was calculated. In

Table 1. GPS coordinates (in degree decimals) for the plots.

Plot	N	E	Plot.	N	E
1	11.90791	77.07389	16	11.84043	77.04111
2	11.90606	77.12222	17	11.91218	77.09833
3	11.89948	77.08639	18	11.93768	77.1125
4	11.88704	77.08	19	11.89156	77.14833
5	11.87264	77.0775	20	11.87458	77.12556
6	11.86323	77.07444	21	11.89091	77.0375
7	11.86099	77.07389	22	11.83303	77.125
8	11.84631	77.10194	23	11.99738	77.15944
9	11.83271	77.10917	24	12.00359	77.14222
10	11.83367	77.1225	25	12.02511	77.10139
11	11.87687	77.11056	26	12.03567	77.10528
12	11.87123	77.10444	27	12.05905	77.14056
13	11.88742	77.13278	28	12.02012	77.14556
14	11.86792	77.13556	29	12.01539	77.1675
15	11.83248	77.01972	30	12.01344	77.16

addition, average distance and minimum distance from specific road types were also calculated, expressed in meters.

Each of the plant characteristics variables, namely, density and richness, were calculated by counting the number of trees per 0.1 ha plot area and the number of different species within the sampling plot, respectively.

In order to check the relationship between two variables; that is, distance from road and community characteristics, correlation tests (Pearson, 1896; Spearman, 1904) were used.

RESULTS

The respective variables for tree density, richness and distances from roads (in meters) for the 30 plots analyzed in this study are presented in Table 2. Tree density ranged from 7 to 62, and richness varied from 3 to 14 tree species per sampling plot area. Average distance from a main road and minimum distance from a main road occurred only for three Plots (Sl. Nos. 1 – 3). The average distance from a main road ranged from 315 to 1012 m, and the minimum distance from a main road ranged from 315 to 669 m. The average distance from an unmetalled road ranged from 24 to 1196 m; and for the minimum distance from the unmetalled road the range also was the same, 24 to 1196 m. On the whole, as may be expected, there were more sample sites that had cart roads in this locale. The average distance from a cart road, and the minimum distance from a cart road, both ranged from 0.006 to 2504 m. Correlation coefficients for the variables of plant characteristics (density and richness) related to geographic variables (road distances) are presented in Table 3. A correlation coefficient (r or r_s) can range in value from -1 to +1. The larger the absolute value of the coefficient, the stronger the relationship between the variables. As shown in Table 3, none of the correlation values are sufficiently large in value to be

considered significant. Therefore, evidently, there is no detectable relationship between the distance from road and plant community characteristics for the data collected in this study.

DISCUSSION

In human settlement landscapes, non-forest woody vegetation represents an important component of green infrastructure in the agricultural areas, where natural and semi-natural forest cover has only a low land use proportion (Tóth et al., 2016). Urban afforestation produces a great enhancement in the flora of cities; but if improperly planted, they generate disturbances (de Oliveira et al., 2018). Street trees are a significant component of urban zones, which can remove a significant amount of CO₂ from the surroundings annually, equivalent to the removal of several thousand cars from the road (McPherson et al., 2016). Most urbanized municipalities have a surplus of non-native species and also trees belonging to native species, as street trees (Thomsen et al., 2016). Narrow roads, usually in congested residential neighborhoods, have fewer trees, smaller-sized tree species and a lower species diversity compared to wide roads (Nagendra and Gopal, 2010). Whereas, in disturbance-prone, human accessible forest landscapes, logging roads can trigger forest degradation by reducing the integrity of the ecosystem and providing access for encroachment, as most logging roads are abandoned after timber harvesting (Kleinschroth et al., 2016). Inter-correlated suite of changes can occur along the margins of the logging roads, like increases in understory foliage density, decreases in sapling density and decreases in tree species richness (Malcolm and Ray, 2000). The

Table 2. Respective tree density, richness and distances from roads (in meters) for the considered plots.

Plot	Density	Richness	Total avg. dist. from road	Total min. dist. from road	Avg. Dist. from M.R.	Min. dist. from M.R.	Avg. dist. from U.R.	Min. dist. from U.R.	Avg. dist. from C.R.	Min. dist. from C.R.
1	7	3	920	669	1012	669	-	-	828	677
2	12	6	982	480	982	480	-	-	-	-
3	25	10	229	142	315	315	-	-	142	142
4	24	6	133	27	-	-	-	-	133	27
5	21	6	270	107	-	-	-	-	270	107
6	60	12	166	141	-	-	-	-	166	141
7	33	7	3.2	3.2	-	-	-	-	3.2	3.2
8	62	5	152	152	-	-	-	-	152	152
9	33	9	357	268	-	-	268	268	446	446
10	53	10	24	24	-	-	24	24	-	-
11	24	8	7.5	7.5	-	-	-	-	7.5	7.5
12	42	9	14	14	-	-	-	-	14	14
13	36	9	512	269	-	-	269	269	755	755
14	45	8	207	207	-	-	207	207	-	-
15	14	3	146	50	-	-	50	50	242	242
16	14	7	0.006	0.006	-	-	-	-	0.006	0.006
17	41	13	521	260	-	-	-	-	521	260
18	51	8	1850	1196	-	-	1196	1196	2504	2504
19	30	14	417	153	-	-	153	153	681	681
20	17	7	472	451	-	-	-	-	472	451
21	11	4	57	47	-	-	57	47	-	-
22	39	9	18	18	-	-	18	18	-	-
23	38	12	120	120	-	-	120	120	-	-
24	37	6	623	364	-	-	823	823	423	364
25	18	8	55	55	-	-	55	55	-	-
26	9	7	661	448	-	-	873	873	448	448
27	28	10	149	122	-	-	-	-	149	122
28	23	10	174	174	-	-	-	-	174	174
29	35	12	501	501	-	-	-	-	501	501
30	33	11	541	307	-	-	-	-	541	307

Avg.- Average; Min.- Minimum; Dist.- Distance; -, not applicable; M.R.- Metalled Road; U.R.- Unmetalled road; C.R.- Cart Road.

distance to logging roads and to unlogged forest can influence post-logging recovery, emphasizing

the importance of edge effects in previously logged forests (Katovai et al., 2016). Most

importantly, along roads, shade-tolerant aliens are to be monitored and removed as they can

Table 3. Correlation results for the considered variables.

X	Y	Pearson correlation coefficient (r)	Spearman correlation coefficient (r _s)
Density	Total Avg. dist.	-0.012	-0.081
Density	Total Min. dist.	0.019	-0.035
Richness	Total Min. dist.	-0.069	0.048
Richness	Total Avg. dist.	-0.072	0.008
Density	Avg. Dist. from metalled road	0	0
Density	Avg. Dist. from unmetalled road	0.140	-0.049
Density	Avg. Dist. from cart road	0.203	0.088
Richness	Avg. Dist. from metalled road	0	0
Richness	Avg. Dist. from unmetalled road	-0.141	-0.155
Richness	Avg. Dist. from cart road	0.042	0.185
Density	Min. Dist. from metalled road	0	0
Density	Min. Dist. from unmetalled road	0.143	-0.033
Density	Min. Dist. from cart road	0.211	0.088
Richness	Min. Dist. from metalled road	0	0
Richness	Min. Dist. from unmetalled road	-0.138	-0.139
Richness	Min. Dist. from cart road	0.031	0.152

Avg.- Average; Min.- Minimum; Dist.- Distance.

potentially invade natural forests (Heinrichs et al., 2018).

In a protected area, not much study of the impact of roads on tree communities has been carried out, except for the compulsive report on dynamics of trees at traditional agroforestry parks in a West African biosphere reserve (M'Woueni et al., 2019). A significant decline of tree density was reported, affected by their proximity to monitoring roads and human populations, as local people continually extracted timber. Also, tree density was found comparatively higher in farms close to the monitoring roads used by park rangers to patrol the park, indicating the importance of conservation surveillance. In the present study, an assessment of road impacts on trees, within a well-protected reserve forest, is presented for 30 randomly selected plots. Cardinaly, the data show that internal roads are not influential on the density and richness of the tree communities in the well-protected forest landscape examined in this study. However, a strong emphasis is made to retain the pristinity of forests by managing internal road-based operations to be as minimal and non-destructive as possible.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Species-based approach to biodiversity conservation: An innovative step in Madagascar

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Known for its diversity and high endemism, half of Madagascar's floral richness is sheltered in the Island's eastern evergreen dense rainforest; including key, threatened and socio-economically valuable species. The vulnerability of such habitat, combined with overexploitation of timber, threatens the livelihoods of forest communities and conservation of biological diversity. Up till date, the main actions regarding Madagascar's biodiversity are leading at the ecosystem level through protected area systems, without necessarily ensuring real conservation of all key species. These are rarely considered as a criterion for effective protected area management. Although decisions on long-term conservation and genetic improvement require detailed scientific and economic knowledge of species, very limited data are available. This research study sets out to promote the development of scientific knowledge of six key endemic species belonging to lowland rainforest ecosystems, to propose a variant of models for the conservation and sustainable use of their genetic resources. The research was conducted using several methods including ecological studies of the habitats of these key species, demographic analysis, and monitoring of their biological and phenological behaviors. The results obtained made it possible to identify three priority groups of key species for which specific conservation and sustainable use measures are proposed.

Key words: key species ecology, biology and physiology, conservation and sustainable use, evergreen dense rainforest.

INTRODUCTION

Madagascar, world-renowned for the value of its ecosystems, fauna and flora, is commonly regarded as one of the world's "megadiversity hotspots" (Mittermeier et al., 1998; Toillier, 2018). Forest biodiversity is

particularly original and very rich in fauna and flora endemic species. Concerning plant species specifically, Malagasy flora is highly diverse with more than 12,000 species of higher vascular plants, of which 84% are

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endemic to the country [Schatz, 2001; Callmander et al. (2011) in Freeman et al. (2014)]. These forest plant species provide a wide range of critically important ecosystem services such as climate regulation, biomass production, water supply and purification, pollination, and the provision of habitats for other forest species (Brockerhoff et al., 2017). They also constitute an exceptional heritage for local populations because of the goods and services they provide (Abdourhamane et al., 2013) and their usefulness for various products such as fuel wood, construction materials, medicine, fodder for livestock, and human food (Langat et al., 2016). Indeed, about one million Malagasy people, particularly the rural poor, depend heavily for their livelihoods on access to forest resources and ecosystems (Angelsen et al., 2014).

Among these plant species are so-called “*key species*” because of their ecological role, their endemism, their socioeconomic value and their state of threat. Madagascar's evergreen dense and low-lying rainforest, which hosts about 57% of country's vascular plant flora, is home to a large number of key species (MEEF, 2014). However, this type of habitat is highly vulnerable due to the pressures and threats it faces, particularly deforestation, slash-and-burn agriculture practices, overexploitation of some timber species (Waeber et al., 2019), as well as the repeated passage of cyclonic events (Lisan, 2015) combined with climate change phenomena. Resource depletion has been increasingly felt in recent years, especially those species of high commercial value, such as rosewood and palisander (Razafintsalama, 2016). As an example, more than twenty species of Malagasy *Dalbergia* species have been classified as “*endangered*” (EN) by IUCN since 1994 and all species of this genus and those of *Diospyros* have been listed in “*Appendix I*” of CITES (Ratsimbazafy et al., 2016). Up to now, in response to this situation, the main actions carried out in favor of biodiversity conservation in Madagascar have been mainly concentrated at the ecosystem level without exploring the added value that the alternative of instituting a species-based conservation approach could produce. Effectively, the ecosystem-based approach is insufficient, because it has not really saved important species.

This research focusing on six key species, belonging to Madagascar's eastern humid forest, is part of the implementation of a project of the Malagasy Ministry of Environment and Sustainable Development (MEDD), in partnership with UN Environment, and is funded by the Global Environment Facility (GEF), for 2017-2022. The aim is to provide the scientific knowledge necessary for the conservation and sustainable use of these species.

MATERIALS AND METHODS

The six key species have been chosen in such a way to represent the most conservation status according to IUCN, in addition to their

socio-economic values and the ecological roles they play. This serves to highlight proposals for specific conservation and sustainable use measures (Table 1 and Figures 1 to 4).

Five protected areas distributed throughout the eastern part of Madagascar, from North to South, were chosen as study areas to better identify intraspecific variability at different latitudes across the natural range of key species distribution (Figure 5). The choice of protected areas relates to the general objective of the project, which is to ensure complementarity between the species-based conservation approach and the ecosystem-based approach. The northeast part includes 3 sites: Pointe à Larrée, Tampolo and Betampona. The first two are New Protected Areas (NPA) established since 2015, covering 770 ha and 675 ha, respectively; while Betampona is an Integral Nature Reserve (INR) established in 1927 and covers 2228 ha. In the south-eastern part, 2 sites belonging to the Atsimo Atsinanana Region have been selected: Manombo, which is a 5320 ha Special Reserve created in 1962; and Mahabo Mananivo, which is a New Protected Area established in 2015 on 2745 ha.

The research topics addressed in this study include the following: limited scientific understanding of key species, vulnerability of their habitat, risk of species loss with the usual ecosystem-based conservation approach, and the total absence of silvicultural practices in natural forests. In order to address this issue and achieve the research objective, the following hypotheses are put forward: “*Changes in ecological habitat conditions affect the viability of species*”; “*Species adopt particular behaviors in relation to changes in the ecological conditions of their environment*”; “*The reproductive biology of target species can be assisted by artificial measures*”.

For this purpose, ecological types were defined across the study sites by combining the following factors: plant community type, habitat degradation status, toposequence and soil type. Key species belonging to each ecological type, thus defined, have been subjected to demographic, phytosociological, phenotypic and biological analysis. To this end, sampling systems were adopted based on the Braun-Blanquet (1965) plot method: 30 square 20 m x 20 m plots were delimited by ecological type at each study site. The choice of plot location is of a rational type, based on the presence of at least one individual of a key species. In each plot, first, the inventory work concerned individuals of all the key species encountered.

Then, with the adapted classification of Blaser and Rakotomanana (1990), inventoried trees were categorized into three groups according to their development stages (Table 2) and the values of the following dendrometric parameters: diameter at breast height (DBH), total height (TH), and biovolume (Vi). This is in order, firstly, to characterize the demography of each target species, reflected by the specific density per ecological type, expressed by the number of individuals present per unit area (Smith, 1963); secondly, to establish the total population structure per species, determined by the distribution curve of the number of individuals according to diametric classes (Rollet, 1969), thus allowing to define their silvicultural compartment in terms of light requirement, called “*temperament*.” The projection of the crown and the shape of the barrel were additional phenotypic characteristics.

Then, 5 seed trees (ST) per species by ecological type were selected, numbered and geolocated to monitor their phenological behavior. The choice was made for trees of good phenotypic quality, vigor and in good sanitary condition. On each selected individual-tree, the followings were documented: different phenophases (period, frequency and duration) defined by Comps et al. (1987), foliation, flowering and fruiting. Fruit and/or seed production capacity and natural seedlings (wildings) availability under the crown of parent plants were estimated during the fruit ripening phenophase to determine the natural reproduction potentiality of each key species. To this end, the following

Table 1. Conservation status and use patterns of key species.

Key species (Family)*	UICN + CITES Status	Use
<i>Asteropeia amblyocarpa</i> Tul. (Asteropeiaceae)(Figure 1)	CR, Critically Endangered	Building timber: manufacture of piles and cabin poles, bridge pillars; Firewood.
<i>Dupuya haraka</i> (Capuron) J.H.Kirkbr.(Fabaceae)(Figure 2)	VU, Vulnerable	Building timber, lumber: manufacture of sleepers, posts and planks; construction of huts; manufacture of canoes and coffins
<i>Dalbergia baronii</i> Baker (Fabaceae)(Figure 3)	VU CITES II, Vulnerable + Appendix II of CITES	Woodworking, wood carving: construction of huts; manufacture of tool handles
<i>Dalbergia chapelieri</i> Baill. (Fabaceae)(Figure 3)	VU CITES II, Vulnerable + Appendix II of CITES	Carpentry and cabinet-making wood. Branches and heartwood used in traditional medicine
<i>Faucherea tampoloensis</i> Aubrév. (Sapotaceae)(Figure 4)	DD, Data deficient	Building timber: manufacture of heavy support structure, bridges, parquet floors, freshwater piles, crosspieces and huts pillars; Firewood.
<i>Tina thouarsiana</i> (Cambess.) Capuron(Sapindaceae)(Figure 4)	EN, Endangered	Carpentry wood, building timber: manufacture of sleepers; used in shipbuilding; Firewood; Leaves and twigs used in traditional medicine.

*According to the Angiosperm Phylogeny Group IV or AGP IV (2016) plant classification system.

parameters were considered and evaluated: number of fruits, seeds and wildings per unit area ($N\ m^{-2}$) on $1\ m \times 1\ m$ square plots around the seed tree along a given axis (Andriambelo, 2007).

During the fruit ripening phenophase, the estimation of the fruit and/or grain production capacity and the availability of natural seedlings (wild) under the crown of the seed plants made it possible to determine the natural reproductive potential of the species. For this purpose, the following parameters were considered and evaluated: number of fruits, seeds and wildings per unit area ($N\ m^{-2}$) on $1\ m \times 1\ m$ square plots around the seed tree along a given axis (Andriambelo, 2007). It was also an occasion to determine the dissemination modes of target species, based on the state of seed dispersal and/or seedling establishment (random, regular, aggregated), as well as their natural regeneration capacity (Rothe, 1964).

Finally, the data collected were analyzed using a series of scientific methods to test research hypotheses, establish the variability of key species across their natural range and identify the differentiation factors in order to propose model variants of conservation and sustainable use. First, the use of a cross-classification table (contingency table) made it possible to identify the ecological preferences of the 6 key species, considering the specific density ($indiv.\ ha^{-1}$). To this end, the existence of a relation between the qualitative variables "key species" and "defined ecological types" was detected by "Chi-square" independence test. Then, an interdependence between these two variables led to a "Correspondence Analysis" (CA) to identify affinities between key species and different ecological types. Subsequently, the "Analysis of variance" (ANOVA) and the "Kruskal-Wallis" non-parametric test were applied to study the variability of target species across their natural range.

The quantitative variables to be explained were the dendrometric parameters (DBH, TH, and Vi). The explanatory ones were constituted by the ecological types where the target species was encountered. ANOVA is a parametric statistic requiring normally distributed data; when this was not the case, or if it concerns discrete qualitative or quantitative variables, such as phenotypic

(crown and bole shape) and biological variables (number of fruits, seeds, wildings per unit area), the non-parametric Kruskal-Wallis test was used (Labreuche, 2010).

For the ANOVA and Kruskal-Wallis tests, the significance level was set at $p \leq 0.05$. The identification of a "significant difference" among the variables at the end of the two types of tests led to an additional test of multiple pairwise comparisons ("post-hoc analysis") to determine the explanatory factors for differentiation and which levels of the independent variable differ from each other level.

RESULTS AND DISCUSSION

Asteropeia amblyocarpa (Asteropeiaceae)

The key species *A. amblyocarpa* was found in Tampolo. The Correspondence analysis (CA), using "symmetric plot" of the rows ("key species") and columns ("defined ecological types") of the contingency table, identified its ecological preferences as well as those of the other target species (Figures 6 and 7).

At the "seed tree" (ST) development stage, *A. amblyocarpa* adopts a particular preference for littoral forest ecosystems on coastal plains. At these locales, the types of alluvial, hydromorphic and sandy soils are favorable for its development.

Generally, if the sand drains easily, therefore dries out and quickly depletes nutrients, one of the forms of adaptation to these habitat types is the presence of mycorrhizae living in symbiosis with this species. For *Dupuya haraka*, ST individuals are



Figure 1. *Asteropeia amblyocarpa* in flowering phenophase (© Rabehevitra).



Figure 2. *Dupuya haraka* in flowering phenophase (© Schatz / Missouri Botanical Garden).



Figure 3. *Dalbergia chapelieri* in flowering phenophase (left) (© Ramananjatovo); *Dalbergia baronii* in fruiting period (right) (© Ramananjatovo).

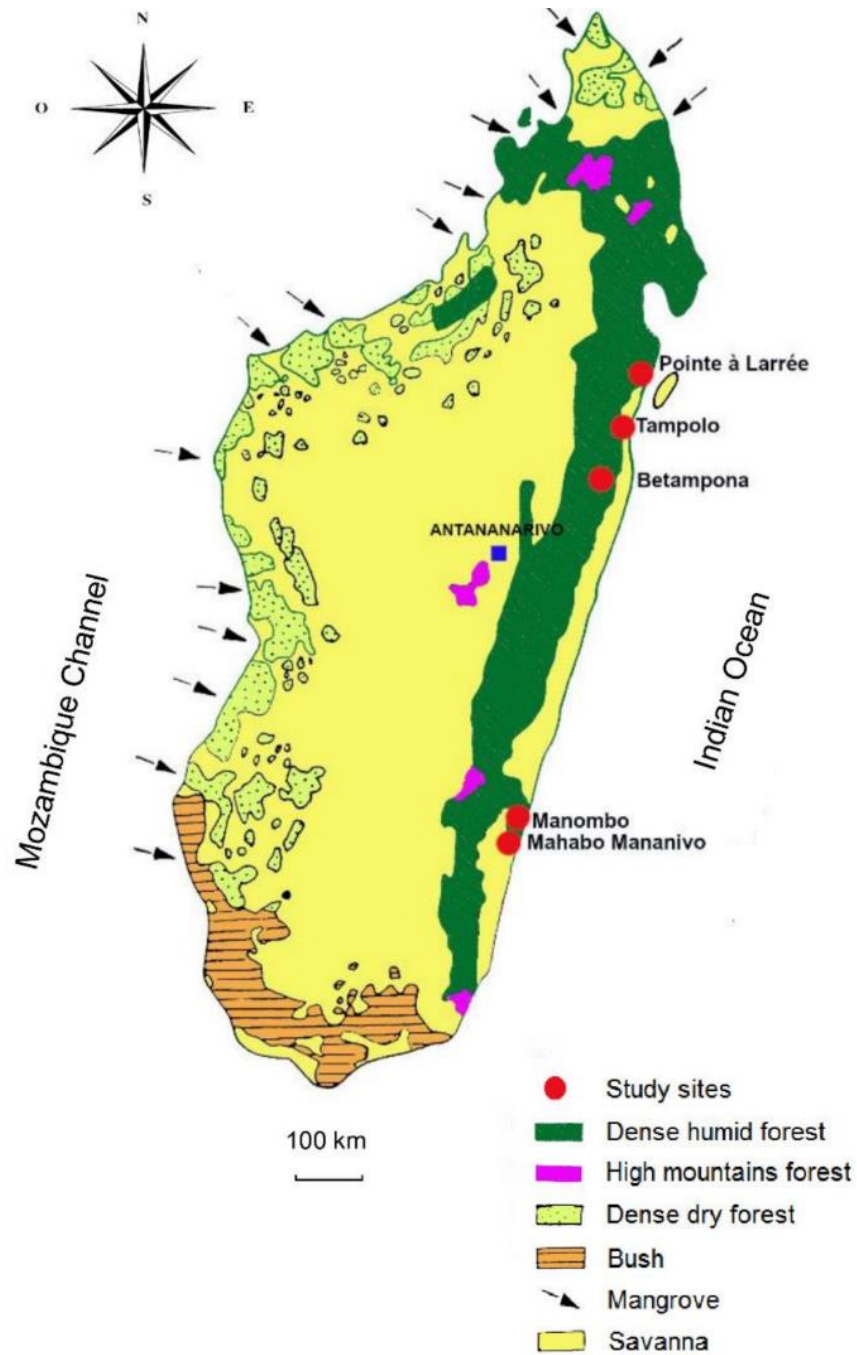


Figure 4. *Faucherea tampoloensis* in flowering phenophase (left) (© Missouri Botanical Garden); *Tina thouarsiana* in fruit ripening phase (right) (© Ramananjatovo).

Table 2. Inventory threshold of individuals encountered in inventory plots.

Class	Codification	Inventory threshold	Development stage
Seed tree	ST	DBH* \leq 10 cm	Large poles, seedling-forest
Earlywood	EW	5 \leq DBH \leq 10 cm	Young pole stands
Regeneration	RG	1 \leq Dc* \leq 5 cm	Regrowth, thicket, sapling

*DBH, diameter at breast height; Dc, root collar diameter (basal diameter).

**Figure 5.** Location of the study sites.

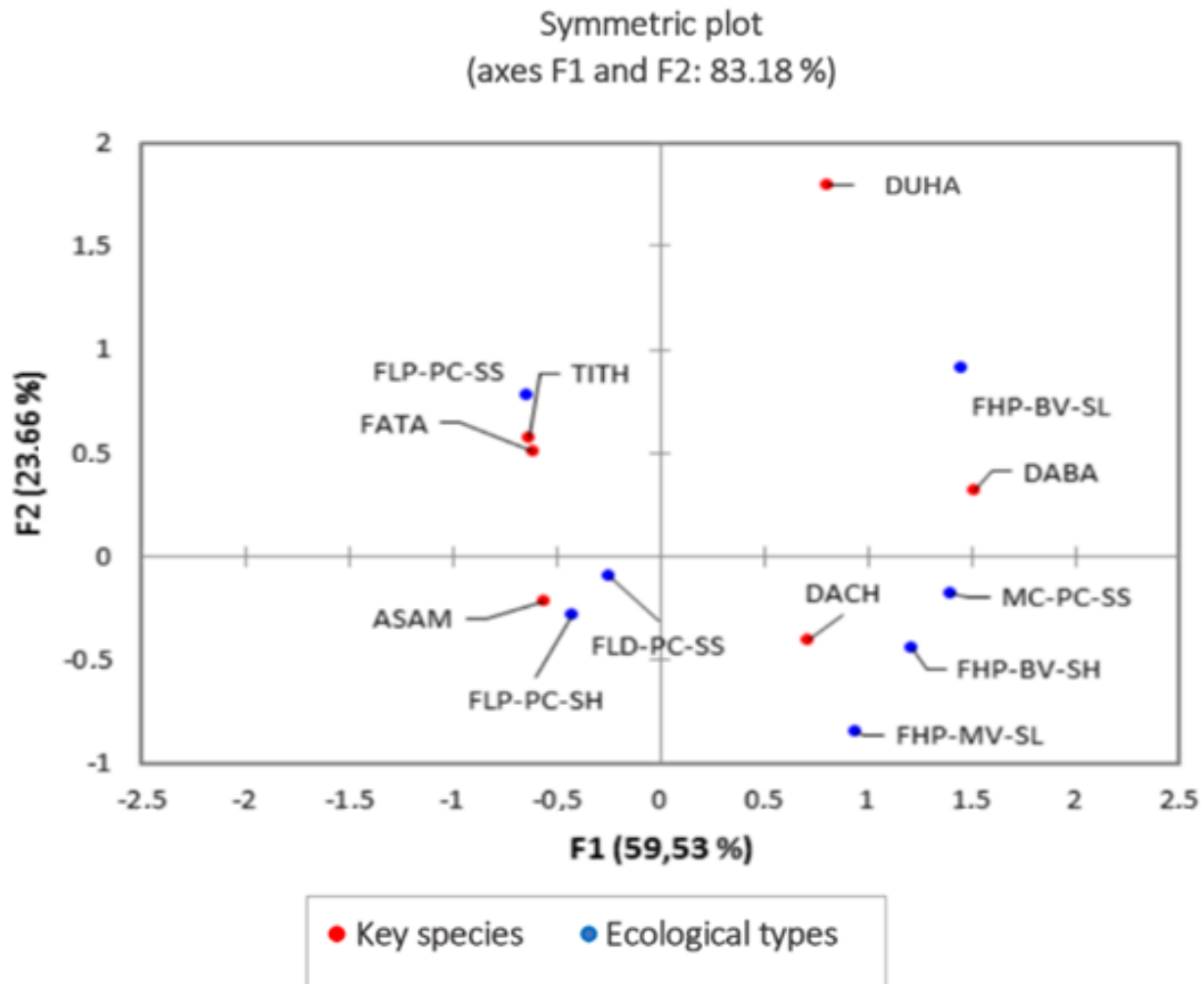


Figure 6. Symmetric plot of the contingency tables between the variables "key species" and "ecological types" for seed tree individuals. Code key. FHP, Dense evergreen humid forest of medium and low altitude; slightly disturbed; FHD, Dense evergreen humid forest of medium and low altitude; degraded; FLP, Littoral forest; slightly disturbed; FLD, Littoral forest; degraded; MC, Mosaic of food crops (tree fields) / BV, Bottom slope; PC, Coastal plain / SH, Alluvial; hydromorphic soil; SL, Red lateritic soil; with concretions; SS, Sandy soil. ASAM, *Asteropeia amblyocarpa*; DUHA, *Dupuya haraka*; DABA, *Dalbergia baronii*; DACH, *Dalbergia chapelieri*; FATA, *Faucherea tampoloensis*; TITH, *Tina thouarsiana*.

isolated and distant from the ecological types defined according to a symmetric plot (Figure 6). This did not allow the ecological preference of the species to be identified. However, the literature suggests that *Dupuya haraka* occurs in humid, and also in deciduous forests, in the Northeastern to the Eastern of Madagascar, mainly between 10-598 masl (Ramanantsialonina, 2019) on sandy, sandy-silt or well-drained limestone soils (Louppe et al., 2008). The two *Dalbergia* species, *D. baronii* and *D. chapelieri*, seem to have the same ecological preferences at the ST development stage. They adapt to several types of formations: littoral forests on coastal plains, low and medium altitude evergreen humid forests, degraded environments, and even in "non-forest" habitat

types such as tree-lined agricultural fields (food crop mosaics). These agricultural fields would probably have been ancient forest ecosystems but have kept living individuals of *Dalbergia*. Seed trees of *Faucherea tampoloensis* and *Tina thouarsiana* form a group with a particular affinity for the ecological types of coastal forests on coastal plains, and seem to adapt well on two types of soils: sandy soils and temporarily flooded alluvial soils.

For natural regeneration individuals (RG), *Asteropeia amblyocarpa*, *F. tampoloensis* and *T. thouarsiana* constitute an isolated group with a particular affinity for littoral forests on coastal plains (Figure 7). Regeneration is absent for *Dupuya haraka*. This species seems to have

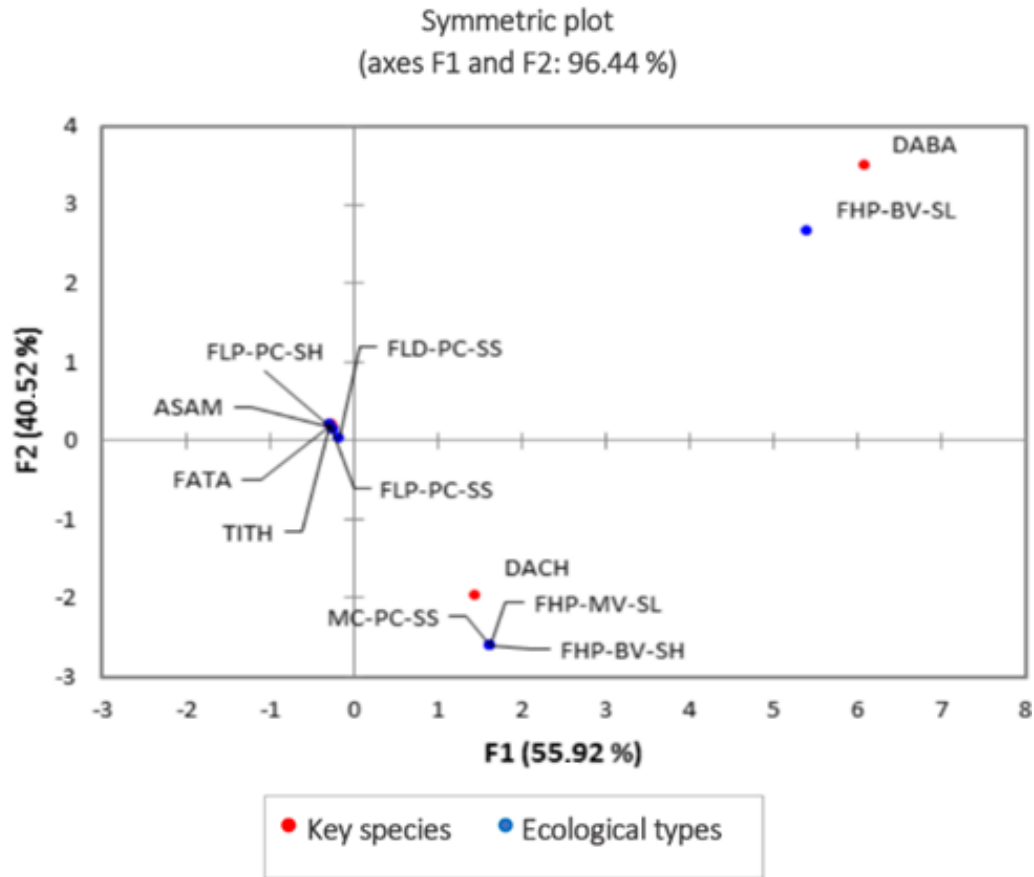


Figure 7. Symmetric plot of the contingency tables between the variables "key species" and "ecological types" for regeneration individuals.

a difficulty in regeneration, and a low potential for reproduction in its natural state. *Dalbergia chapelierii*'s juvenile trees have the same ecological preferences as the ST development stage, which adapts to almost all types of formation: it can be found in humid valleys as well as on drier crest, on lateritic or sandy soil, and even may survive as a shrub after resprouting in secondary vegetation (Contu, 2012). However, *Dalbergia baronii* regenerations are very demanding, adopting a strict preference for low-elevation humid forest types on bottom slopes (Figure 7).

The population of *Asteropeia amblyocarpa* is characterized by a high abundance of natural regeneration compared to adult individuals (Table 3). The potential for regeneration of the species in its natural state is quite good with a rate above 500% according to the Rothe scale (1964). Inventoried trees generally have the same phenotypic characteristics: a tolerable crown shape, more or less circular in plan with some deficiencies in symmetry, and a good conformation of the bole shape; *i.e.*, straight, round and solid, cylindrical, without serious defects. Trees up to 50 cm of DBH have

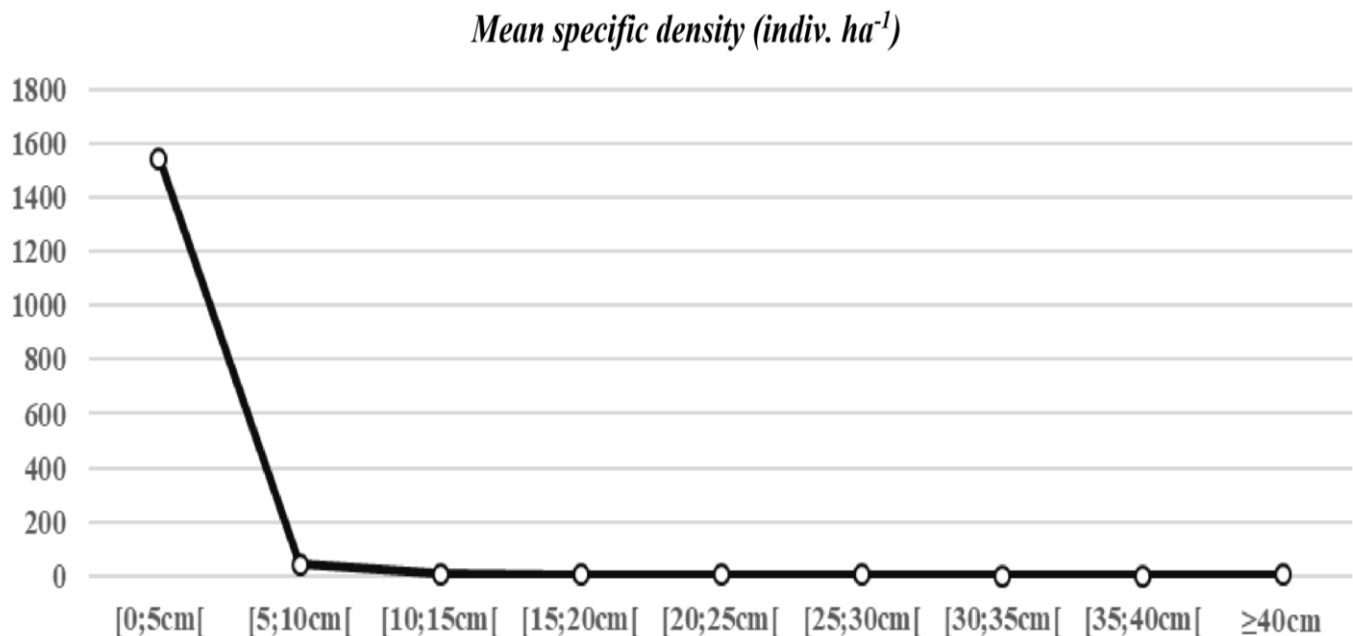
been recorded, but the average DBH of the key species remains quite low (DBHa = 10.77 cm).

The high reproductive potential of the target species *A. amblyocarpa* is partly explained by its phenological behavior, characterized by a year-round observation period of flowering and fruiting phenophases. Seed trees produce abundant flowers and fruits during these phenophases (average production per parent plants $Nm = 42$ mature fruits m^{-2}). The continuity and regularity of the phenological rhythm, spread over most of the year, is announced by Sabatier and Puig (1986) as characteristic of small trees in the forest understorey that thrive under particular edaphic conditions such as hydromorphic soils that reduce climatic seasonality. Indeed, the distribution by diametric class of the trees inventoried in exponential form (Figure 8), according to the Rollet curve (1969), gives the species a sciaphilous character temperament of the undergrowth. As a result, the opening of the canopy due to windthrow and anthropogenic disturbances significantly affects the installation and development of natural seedlings. In disturbed environments, the natural regeneration rate or NRR (Rothe, 1964) can be as high

Table 3. Demographic and dendrometric characteristics of the key species *Asteropeia amblyocarpa*.

Ecological types code	Specific density (indiv. ha ⁻¹)			Dendrometric characteristics (ST individuals)		
	ST	EW	RG	DBHa (cm)	THa (m)	Vi (m ³ ha ⁻¹)
FLP-PC-SH	20	95	2870	10.2 ± 6.7*	4.9 ± 2.1*	4.08
FLP-PC-SS	8	15	480	11.6 ± 5.8	6 ± 1.6	0.4
FLD-PC-SS	10	23	1280	10.5 ± 5.9	6 ± 1.7	0.88

*DBHa, average of the DBH ± standard deviation (n-1); THa, average of the TH ± standard deviation (n-1).

**Figure 8.** Distribution of the mean specific density (indiv. ha⁻¹) of *Asteropeia amblyocarpa* according to diametric classes (cm).

as 1000%. This high rate in disturbed areas can be explained either by the low proportion of adult individuals in relation to regeneration due to their exploitation, or by the fact that the species needs a certain luminosity condition to regenerate. This was explained by Whitmore (1989), that even "*shade-tolerant*" species, able to survive and even grow in the shaded undergrowth, have responded positively to a certain opening of the canopy, a condition allowing them to move on to the next stages (thicket, saplings, young pole stands).

A. amblyocarpa could therefore be a semi-sciaphilous to sciaphilous species. However, with good germination capacity in the natural state, abundant availability of wildings under the crown $N_a = 36$ seedlings/ m²/ indiv. ST, and a high NRR% (NRR > 500%), many regenerative individuals (RG) cannot survive and develop to the "*young*" and "*large poles stands*" stages of development (EW and ST) (Table 3). The aggregate dispersal pattern

of seedlings suggests that the high density of wildings per unit area (m²) has favored natural selection to reduce competition between individuals. But several factors can damage young regrowth during this stage of development, the most important of which are: pathogenic fungi (Augspurger, 1983), insects and vertebrates feeding on leaves or stems (Angulo and Aide, 2000), mortality due to falling debris (Clark and Clark, 1991), water reserve in the soil (Wright, 1992), light exposure (Baraloto, 2001).

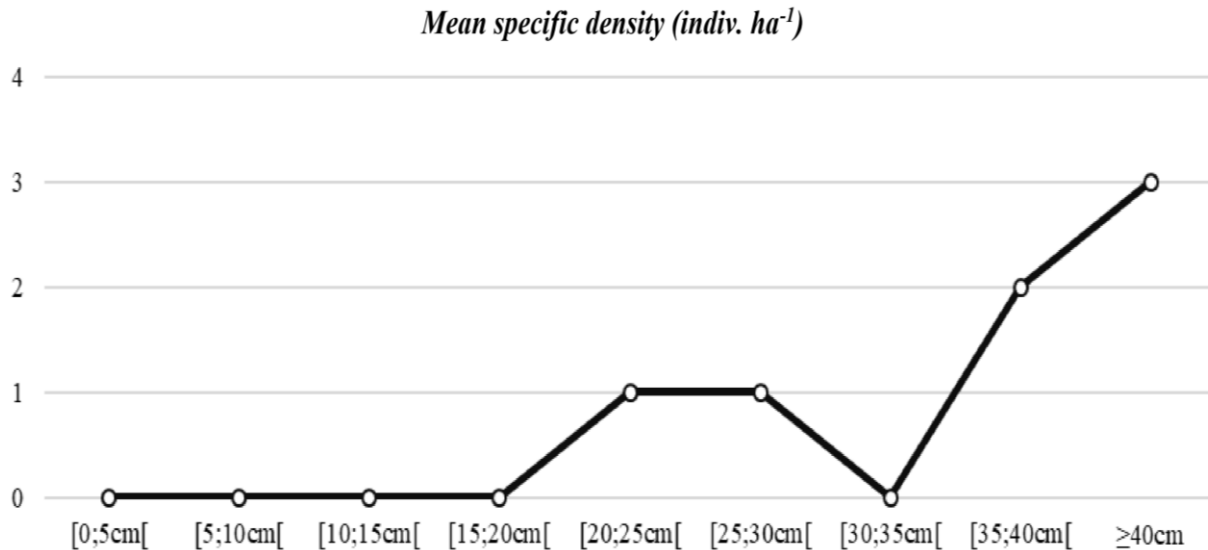
***Dupuya haraka* (Fabaceae)**

The species has been found in Tampo and Betampona. The distribution of individuals recorded in the inventory plots according to diametric classes showed a low abundance of seedlings and the absence of natural regeneration (Table 4).

Table 4. Demographic and dendrometric characteristics of the key species *Dupuya haraka*.

Ecological types code	Specific density (indiv. ha ⁻¹)			Dendrometric characteristics (ST individuals)		
	ST	EW	RG	DBHa (cm)	THa (m)	Vi (m ³ ha ⁻¹)
FLP-PC-SS	5	0	0	55.3 ± 13.9*	22.8 ± 4.89*	9.3
FHP-BV-SL	8	0	0	54.3 ± 22.8	21.3 ± 6.8	18.2

*DBHa, average of the DBH ± standard deviation (n-1); THa, average of the TH ± standard deviation (n-1).

**Figure 9.** Distribution of the mean specific density (indiv. ha⁻¹) of *Dupuya haraka* according to diametric classes (cm).

Over its natural distribution area as a whole, the demographic characteristics of the species reflect its difficulty in regenerating and its very low potential for reproduction in the natural state, with a NRR of 0% (Rothe, 1964) at both sites. Several factors could explain this behavior. During the phenological monitoring of the ST individuals, no flowering or fruiting phenophase was observed. It is therefore assumed that this species exhibits discontinuous and irregular phenological behavior, which, according to Sabatier and Puig (1986), is characteristic of large trees in the forest. Effectively, *Dupuya haraka*'s seed trees share the same phenotypic characteristics throughout its natural range; that is, large individuals of up to 80cm of DBH and up to 30m in TH; high average exploitable wood volume of 13.8m³/ha; good conformation of the tree characterized by a topless, circular, planar, symmetrical, dense and extended crown with a straight, round, cylindrical, flawless and branchless bole. However, the variation in ecological types has no effect on these phenotypic characteristics (ANOVA; DBH: $p = 0.95$, TH: $p = 0.90$, Vi: $p = 0.84$). The species thus has an interannual alternation of production which,

according to Normand (2014), results from a year of high production followed by one or two years of low or no production. Production alternation can be influenced by climatic or endogenous (tree-specific) factors (Goldschmidt, 2005). When fruiting takes place, the conditions of regenerations installation are strongly associated to the physiological behavior of the seeds. Indeed, *D. haraka* seeds have a delayed germination process whose dormancy removal is conditioned by several parameters. They adopt partial dormancy, which after collection and storage for about 6 months has shown a low germination rate of 30-40%, while those stored away from heat and humidity after 1.5-2 years have reached 84% germination rate (Louppe et al., 2008). Thus, the variability of environmental conditions such as soil moisture, litter type and thickness, light quality and temperature has a direct influence on the continuation or termination of the process (Dalling et al., 1998). While the erratic aspect (Figure 9) of the distribution curve of individuals according to diameter classes (Rollet, 1969) determines a pioneering heliophilous temperament ("*shade intolerant*") for this

Table 5. Demographic and dendrometric characteristics of the key species *Dalbergia baronii*.

Ecological types code	Specific density (indiv. ha ⁻¹)			Dendrometric characteristics (ST individuals)		
	ST	EW	RG	DBHa (cm)	THa (m)	Vi (m ³ ha ⁻¹)
FLP-PC-SH	8	0	0	25.5 ± 5.7*	6 ± 0.3*	0.64
MC-PC-SS	8	2	0	26.3 ± 3.7	7.8 ± 0.9	1.31
FHP-BV-SL	10	0	80	19.5 ± 3.8	6.1 ± 0.5	0.67
FHP-BV-SH	12	3	0	19.8 ± 3.1	6.6 ± 0.4	1.15

*DBHa, average of the DBH ± standard deviation (n-1); THa, average of the TH ± standard deviation (n-1).

species; and natural young seedlings prefer full sunlight (Louppe et al., 2008). The high density of vegetation cover and associated species, as in the case of the Betampona site, tend to compromise survival and the passage of regeneration to more advanced stages of development (regrowth, sapling). In addition, *D. haraka* is an endozoochorous species, and there is a need for intestinal seed transit, after ingestion by an animal, to stimulate seed germination. Louppe et al. (2008) argue that it is the lemurs that eat and disseminate the seeds of *Dupuya haraka*. The considerable decrease in the number of lemurs, especially in the Tampolo protected area, due to poaching and habitat destruction, could have significant effects on the species' reproductive capacity in the natural habitat.

Dalbergia baronii (Fabaceae)

This key species has been found in Pointe à Larrée, Tampolo and Manombo. Whereas in the past, the species was found in the Mahabo Mananivo site with a specific abundance of 34 indiv. ha⁻¹ for EW and ST stems with a DBH ≥ 5cm (Ralambomanana, 2006), currently no individuals have been found. According to Razafintsalama (2016), *D. baronii* is among the highly exploited rosewood species, so that even the seed trees have disappeared, resulting in the absence or rarity of juvenile individuals and regeneration (Table 5).

In addition to the influence of anthropogenic disturbances on the demographic characteristics and reproductive potential of the species, they are also influenced by ecological parameters, such as variability of the habitats throughout its distribution area (Kruskal-Wallis; fruit production: $p < 0.02$; availability in wildings: $p < 0.01$). The fruit and seed production capacity of the ST individuals is low on the coastal plains of the northeastern part ($N_a = 6$ mature fruits/ m²/indiv. ST) compared to those of the southeastern part ($N_a = 13$ fruits/ m²/indiv. ST) situated on the low-elevation humid forest. This is influenced by the phenological shift of seed trees in these two areas: the fruiting and maturing phenophase lasts two months (October - November) in the North and four

months (November - February) in the South. This difference appears to be explained by climatic variability between sites, constituting the external factors of phenological variability including local daytime and nighttime temperatures, precipitation and photoperiodism. Climate is not only variable on a temporal scale but also on a spatial scale that could vary within a region and even at the local level (Rasamimanana, 2011). In the face of this climate variability and the adverse effects of climate change, the species may have adopted a particular form of adaptation. According to Kramer (1995), this adaptation generally takes the form of a compromise between the different needs of the plant by moving forward, backward or reducing the onset and duration of a phenophase. This phenomenon is often found in species that are most vulnerable to rainfall variability, such as those in the sub-humid regions of southern Madagascar: *Acacia bellula*, *Rhigozum madagascariensis*, *Grewia franciscana*, *Commiphora sp.*, *Uncarina grandidieri* and *Terminalia fatrae* (Rasamimanana, 2011). This ecological and phenological variability between sites significantly affects the installation and development of natural regeneration. The natural regeneration rate is zero (NRR = 0%) in littoral forests on the coastal plain of the northeastern part of Madagascar. Similar results were found by Razafintsalama (2016) confirming that *Dalbergia baronii* appears to have a regeneration problem in this region (NRR < 100%). Soil characteristics were identified as another factor of variability conditioning the germination of seeds in their natural state and the availability of natural seedlings. In coastal plains on sandy soils, the thin and highly permeable layer, often low in organic material and humus, is not favorable to seed germination and natural regrowth development ($N_a = 3$ wildings/ m²/indiv. ST). In temporarily flooded areas with hydromorphic soils, the fruit ripening period coincides with the period of heavy rainfall leaving light-weight fruits and seeds washed away by the floods ($N_a = 2$ wildings/ m²/indiv. ST). On the other hand, in the southeastern part, with an average seedling availability under the crown of $N_a = 16$ wildings/ m²/indiv. SM, the natural regeneration capacity, according to the Rothe scale (1964), is good with a

Mean specific density (indiv. ha⁻¹)

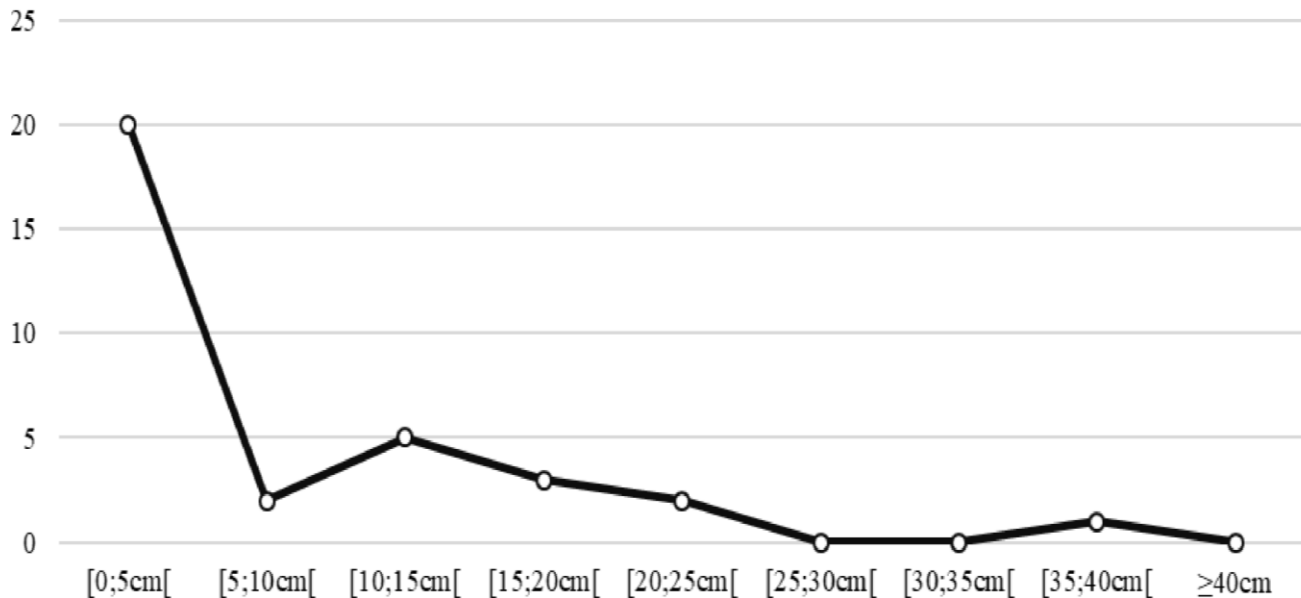


Figure 10 . Distribution of the mean specific density (indiv. ha⁻¹) of *Dalbergia baronii* according to diametric classes (cm).

NRR = 133%. In this region, natural regenerations and juvenile individuals adopt a particular preference and form an isolated group at lowland humid forest types on bottom slopes (FHP-BV-SL) where the germination potentiality of seeds in the natural state, young seedlings development and growth are favored by lateritic soils characterized by a very rapid podzolization phenomenon (Figure 7).

As for the phenotypic characteristics of the species, they are little affected by the variation in the ecological parameters of the habitat throughout its natural range (ANOVA; DHP: $p = 0.77$; HT: $p = 0.5$; Vi: $p = 0.08$); so one could assume that they are determined by their genome. The inventoried plants are generally of poor conformation, characterized by a strongly asymmetrical, open crown, with an irregular, tortuous, partly defective shaft. The bell-shaped, spread-out appearance of the stem distribution curve depending on diametric classes (Figure 10), according to Rollet (1969), gives the species a heliophilous temperament. This statement is consistent with the characteristics of the species according to Blaser et al. (1993): *Dalbergia baronii* has a nomadic temperament and is used in silvicultural enrichment.

***Dalbergia chapelieri* (Fabaceae)**

D. chapelieri was met in Pointe à Larrée, Manombo and Mahabo. Considered to be a widespread species (Du Puy et al., 2002), variation in ecological types has little effect on the following phenotypic characteristics: crown

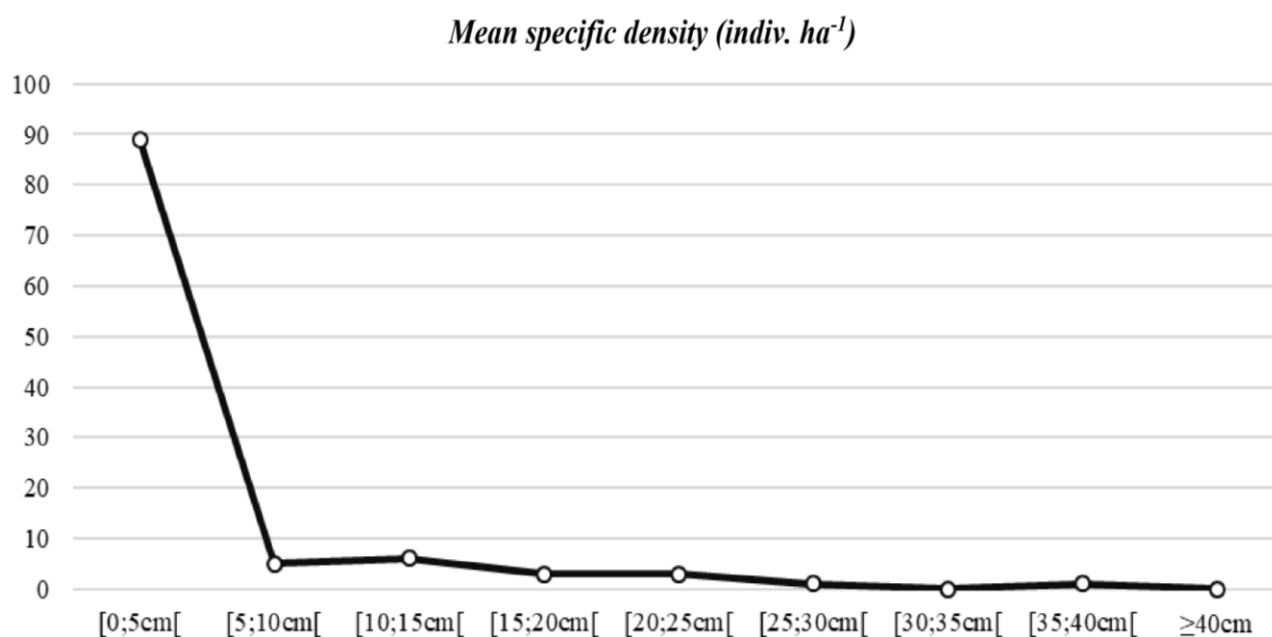
and bole shape, DBH and adult tree biovolume. In relation to these variables, all individuals share common characteristics across their range: a tolerable crown shape (more or less circular in plan with some deficiencies of symmetry or with some dead branches), a poorly shaped bole (irregular, tortuous, partly defective) with a DHPa = 14.2 cm. On the other hand, in relation to the variable total height TH, the tree found in low and medium altitude humid forests have higher heights (THa = 7.8 m) than those inventoried on coastal forests (THa = 6.5 m) (ANOVA; $p < 0.035$). These variations can be explained by various factors, including species temperament, density and structure of associated vegetation, and competition for light (Rakotondrasoa et al., 2013) (Table 6).

Like *D. baronii*, the demographic characteristics and natural reproductive potentiality of the target species are highly dependent on the ecological variability of the habitat. A phenological shift between the seed-tree ST in the northeast and southeast has also been observed: the fruiting and maturing phenophase lasts two months (December - January) in the north and up to three months (November - January) in the south. This made it possible to identify two significantly different groups in terms of fruit and seed production capacity (Kruskal-Wallis; $p < 0.001$). On the one hand, individuals characterized by a relatively low average production of $N_a = 11$ mature fruits/m²/indiv. ST were found in the North. And on the other hand, in the South, trees were found with an abundant fruit production capacity of up to $N_a = 30$ mature fruits/m²/indiv. SM; even in agricultural

Table 6. Demographic and dendrometric characteristics of the key species *Dalbergia chapelieri*.

Ecological types code	Specific density (indiv. ha ⁻¹)			Dendrometric characteristics (ST individuals)		
	ST	EW	RG	DBHa (cm)	THa (m)	Vi (m ³ ha ⁻¹)
FLD-PC-SS	15	10	80	15.6 ± 11.2*	7.1 ± 1.7*	1.40
MC-PC-SS	13	0	50	13.8 ± 2.1	5.9 ± 0.8	0.26
FHP-MV-SL	5	3	10	14 ± 4.1	8.7 ± 1	0.41
FHP-BV-SL	10	0	30	14.7 ± 4.8	7.9 ± 0.8	0.44
FHP-BV-SH	18	10	280	12.4 ± 5.9	6.8 ± 1.1	1.20
FLP-PC-SH	20	3	80	14.6 ± 7	6.5 ± 1.6	1.22

*DBHa, average of the DBH ± standard deviation (n-1); THa, average of the TH ± standard deviation (n-1).

**Figure 11.** Distribution of the mean specific density (indiv. ha⁻¹) of *Dalbergia chapelieri* according to diametric classes (cm).

wooded fields. But unlike *Dalbergia baronii*, no significant difference was observed between the availability of wildings under the crown of selected parent plants across its natural distribution area (Kruskal-Wallis; $p = 0.103$). The germination capacity of the species in its natural state is average, with an availability of $N_a = 20$ wildings/m²/ indiv. ST. However, the installation and development of young regrowth in the upper stages (thicket, saplings, young pole stands) varies according to habitat types. Soil type has been identified as the main differentiating factor. The installation of natural regenerations is more favored in the ecological type of low and medium altitude humid evergreen forests, on temporarily flooded alluvial soils on which natural regenerations are relatively good reaching a NRR of 255% on the Rothe scale (1964). In the sandy

coastal plains of littoral forests, the density of natural regenerations is low, especially in disturbed environments and agricultural fields (NRR < 80%). This also seems to be linked to the semi-sciaphilous builder type temperament of the species, resulting from the negative exponential aspect of the curve (Figure 11) of stem distribution by diametric class (Rollet, 1969), where a significant opening of the canopy linked to the destruction of its habitat compromises the development and installation of young seedlings.

***Faucherea tampoloensis* (Sapotaceae)**

F. tampoloensis was found at two sites in the northeastern

Table 7. Demographic and dendrometric characteristics of the key species *Faucherea tampoloensis*.

Ecological types code	Specific density (indiv. ha ⁻¹)			Dendrometric characteristics (ST individuals)		
	ST	EW	RG	DBHa (cm)	THa (m)	Vi (m ³ ha ⁻¹)
FLP-PC-SS	15	3	40	14.6 ± 4.6*	7.1 ± 1.8*	0.77
FLP-PC-SH	10	0	30	15 ± 1.4	5.6 ± 0.6	0.33
FLD-PC-SS	13	0	20	15.1 ± 1.5	5.7 ± 0.6	0.40

*DBHa, average of the DBH ± standard deviation (n-1); THa, average of the TH ± standard deviation (n-1).

part: Pointe à Larrée and Tampolo. Throughout the species' natural distribution area, the variation in phenotypic characteristics of the inventoried trees, particularly the total height and timber potential, is strongly influenced by ecological types (ANOVA; TH: $p < 0.003$; Vi: $p < 0.04$). Two significantly different groups were identified with respect to these two dendrometric variables. The stems inventoried at Pointe à Larée have a higher total height (TH) and wood volume (Vi) (THa = 8.2 m; Vi = 0.88 m³ ha⁻¹) than those found at Tampolo (THa = 5.6 m; Vi = 0.38 m³ ha⁻¹). In contrast, the DBH does not vary according to habitat types with an average of DBHa = 14.9 cm over its entire distribution area (ANOVA; DBH: $p = 0.78$). Similar cases have been observed in the target species *D. chapelieri*, and other endemic species such as *Uapaca bojeri*, where trees with the same DBH have significantly different total heights TH (Rakotondraso et al., 2013). However, it should be noted that the state of habitat degradation is more significant in Tampolo than in Pointe à Larrée, which is the main factor differentiating the timber potential between these two sites.

The demographic behaviors of the target species are characterized by a small number of individuals in all diameter classes (Table 7). Among the main factors that influence this distribution is the phenological behavior of the species during flowering and fruiting phenophases. They are spread over about 4 months and are marked by the abundance of flowers. Aubreville (1974) had described that flowering in this species occurs from January to April; while phenological monitoring work at study sites showed that flower bud formation begins in October, followed by fruit bud appearance from December. This phenological lag could be considered as a form of adaptation of the species to climate change phenomena (Kramer, 1995). The species has advanced the onset of flowering and fruiting phenophases to meet its various needs (Kramer, 1995). However, this time lag coincided the period of evolution of floral buds to the stage of fruit bud formation, with the cyclonic periods when Madagascar's eastern coastal forests are subjected to repeated passage (Lisan, 2015). The flowers and fruits, small and very slight, do not resist strong winds. Very few bud fruits reach the ripening stage. The

potential for fruit and seed production is, therefore, low throughout its natural range. The distance and location of the seed plants from the sea has a negative influence on their ability to produce fruit, and varies significantly according to the environment (Kruskal-Wallis; $p < 0.006$). Individuals located along the seaside are directly exposed to strong winds during cyclonic periods and have a low production capacity of Na = 5 mature fruits/m²/ indiv. SM. On the other hand, trees further inland, at transect level, have a higher production potential of Na = 15 mature fruits/m²/ indiv. SM. But, this variation does not affect germination capacity in the natural state where the availability of natural seedlings under the crown of the parent plants remains low throughout the species' range (Nm = 4 wildings/m²/ indiv. SM). Consequently, over its entire range, the species has a low germination and regeneration capacity in its natural state (NRR < 80%). This low potentiality is particularly marked in the types of degraded forest formations (NRR = 40%). This is partly explained by the semi-sciaphilous character temperament ("sciaphilous builder type") (Figure 12) of the species, where the large opening of the canopy tends to compromise seed germination, survival and development of natural seedlings to higher stages.

Tina thouarsiana (Sapindaceae)

The species has been found in three sites: Pointe à Larrée, Tampolo and Mahabo Mananivo. The ecological environment types have very little effect on the variation of the following phenotypic characteristics: crown and bole shape, DBH and biovolume (ANOVA; DHP: $p = 0.43$; Vi: $p = 0.08$). With an average DBH of 15.5 cm and a biovolume of 0.67 m³ ha⁻¹ over its entire natural range, adult trees generally have a good crown shape: free from above, circular in plan, symmetrical and extended. As for the shape of the bole, it is tolerable: partially straight, conical and without serious defects. However, three significantly different groups were identified with regard to the total height TH variable (ANOVA; $p < 0.02$). In other words, as in the case of the *D. chapelieri* and *F. tampoloensis* species, for the same diameter, some inventoried trees are significantly higher than others

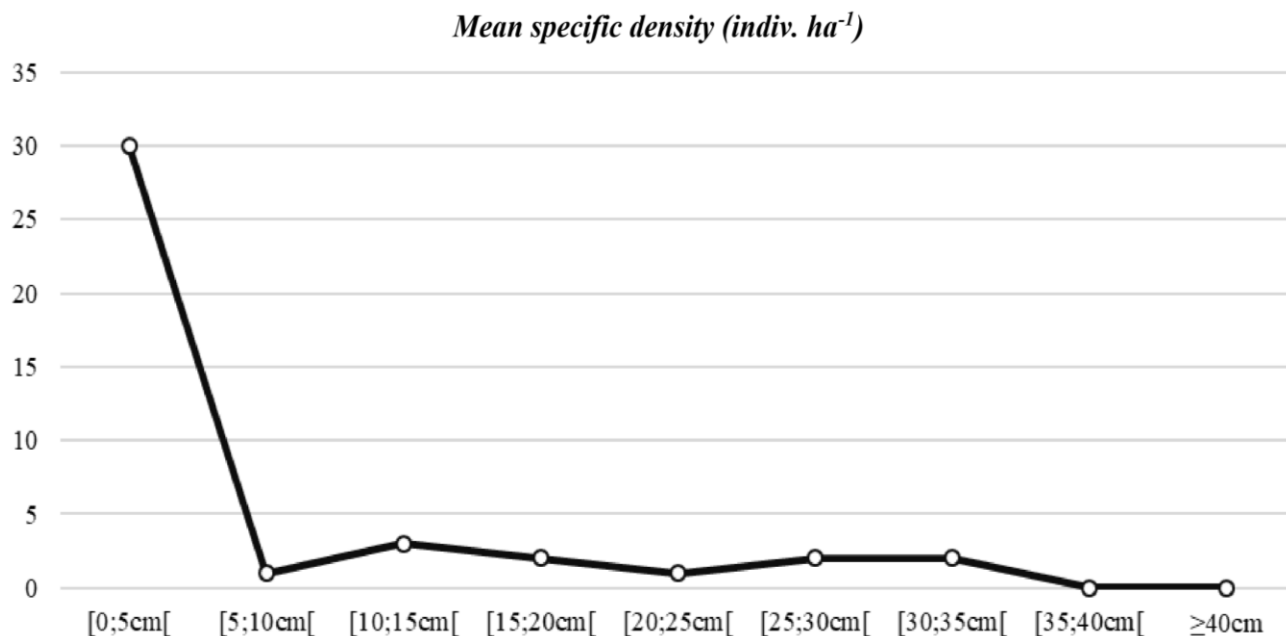


Figure 12. Distribution of the mean specific density (indiv. ha⁻¹) of *Faucherea tampoloensis* according to diametric classes (cm).

Table 8. Demographic and dendrometric characteristics of the key species *Tina thouarsiana*.

Ecological types code	Specific density (indiv. ha ⁻¹)			Dendrometric characteristics (ST individuals)		
	ST	EW	RG	DBHa (cm)	THa (m)	Vi (m ³ ha ⁻¹)
FLP-PC-SS	10	11	40	16.8 ± 4.9*	7.4 ± 1.6*	1.04
FLP-PC-SH	8	2	90	15.6 ± 3.1	6.6 ± 0.7	0.52
FLD-PC-SS	10	0	30	14.3 ± 3.8	5.9 ± 0.5	0.45

*DBHa, average of the DBH ± standard deviation (n-1); THa, average of the TH ± standard deviation (n-1).

(Table 8).

The state of degradation of forest formation types has been identified as a differentiating factor. Individuals in disturbed environments are smaller in size compared to those found in less-disturbed formations. This is related to the heliophilous character temperament of the species (bell-shaped appearance according to the Rollet distribution curve) (Figure 13); where trees in undisturbed areas, with a density of vegetation that is much higher, seek to prioritize growth in height for light competition.

The flowering and fruiting phenophases of the target species are spread throughout the year, with a fruit ripening frequency of 3 to 4 times per year. During fruit ripening periods, the production capacity depends on ecological parameter variations (Kruskal-Wallis; $p < 0.04$). Production is abundant for seed trees in littoral forests with little disturbance, on sandy soils (FLP-PC-

SS) with an average density of $N_a = 46$ mature fruits/ m²/ indiv. ST. It is average for the other ecological types with an abundance of $N_a = 14$ mature fruits/ m²/ indiv. ST. The availability of wildings under the crown of the seed trees, on the other hand, does not depend on the ecological types (Kruskal-Wallis; $p = 0.06$). Over its entire range, it appears that the species has good germination capacity with a high abundance of natural seedlings of $N_a = 20$ wildings/ m²/ indiv. ST. But, the installation and development of these young regrowths are conditioned by the ecological conditions of their habitat. The installation of natural regeneration is favored by the types of alluvial and temporarily flooded soils with a good NRR of 225%. In contrast, many natural seedlings (regrowth) fail to survive and develop to the next stages (thicket and saplings) on sandy soils; the natural germination rate is low (NRR < 100%). Also, the degree of degradation of

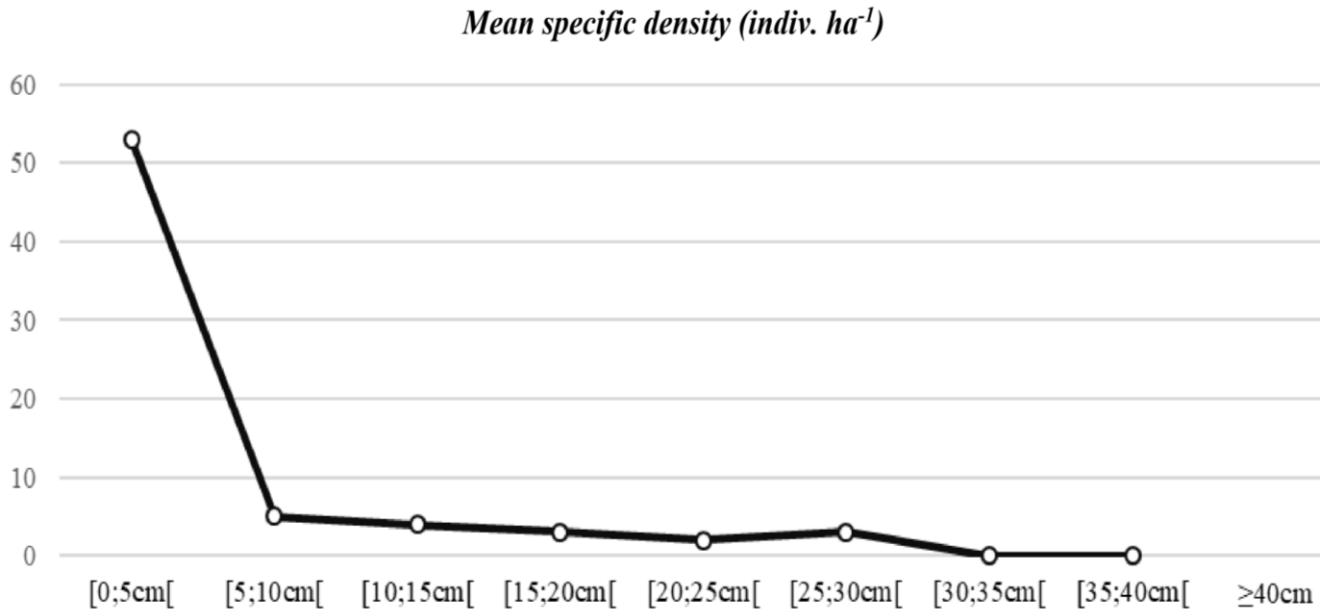


Figure 13. Distribution of the mean specific density (indiv. ha⁻¹) of *Tina thouarsiana* according to diametric classes (cm).

the habitat has a significant influence on the conditions under which natural regenerations are installed. On the same type of soil (sandy), the NRR% is higher in degraded habitat types (NRR = 75%) than in low-disturbed areas (NRR = 50%). This variability is explained by the heliophilous character temperament of the species requiring the opening of the canopy and a less dense vegetation cover to promote optimal conditions for seed germination and the survival of young regrowth.

Actions for the conservation and sustainable use of key species

Several factors were taken into account in the elaboration of conservation and sustainable use models for the 6 key target species (Figure 14). The conservation status of these species according to the IUCN and CITES lists is one of the central elements allowing a hierarchization in regards to their vulnerability, from which the priorities for conservation actions have mainly been proposed.

The categories "Critically Endangered (CR), Endangered (EN) and Vulnerable (VU)" include so-called "threatened" species, which are at high risk of extinction in the wild and deserve special attention (Barneix and Gigot, 2013). *A. amblyocarpa* (CR), *T. thouarsiana* (EN), *D. haraka* (VU), *D. baronii* (VU) and *D. chapelieri* (VU) belong to this category. The two species of the genus *Dalbergia* are also listed in Appendix II of CITES, the trade in specimens of which must be regulated and all forms of exploitation must comply with the laws in force

and use methods that are not detrimental to the survival of the species.

The taxon in the category of "Insufficient Data (DD)" may include unknown species that would be classified as threatened if a minimum of information on the status of their populations were available. *F. tampoloensis* belongs to this category. However, the IUCN and CITES statutes alone are not sufficient to define conservation and sustainable use priorities. They require the integration of complementary or associated factors. For the 6 key species, their demographic characteristics and the variability of their biological behaviors were chosen as additional factors to provide more supplementary information needed to propose and prioritize conservation measures. The sites most representative of the variability of these factors have been chosen as high priorities for conservation actions in order to better preserve the genetic resources of the species.

In relation to all these factors, species with the same characteristics have been grouped together (Figure 14) as listed in the following groups: *high priority species* (1), characterized by low fruit and seed production potential, low natural germination capacity and difficult natural regeneration; *medium priority species* (2), with relatively abundant fruit and seed production, good germination capacity in the natural state, but difficult natural regeneration; and *low priority species* (3) characterized by abundant fruit and seed, good germination and natural regeneration capacity. Each group thus formed was the subject of proposals for specific conservation and sustainable use measures in order of priority (grouping by

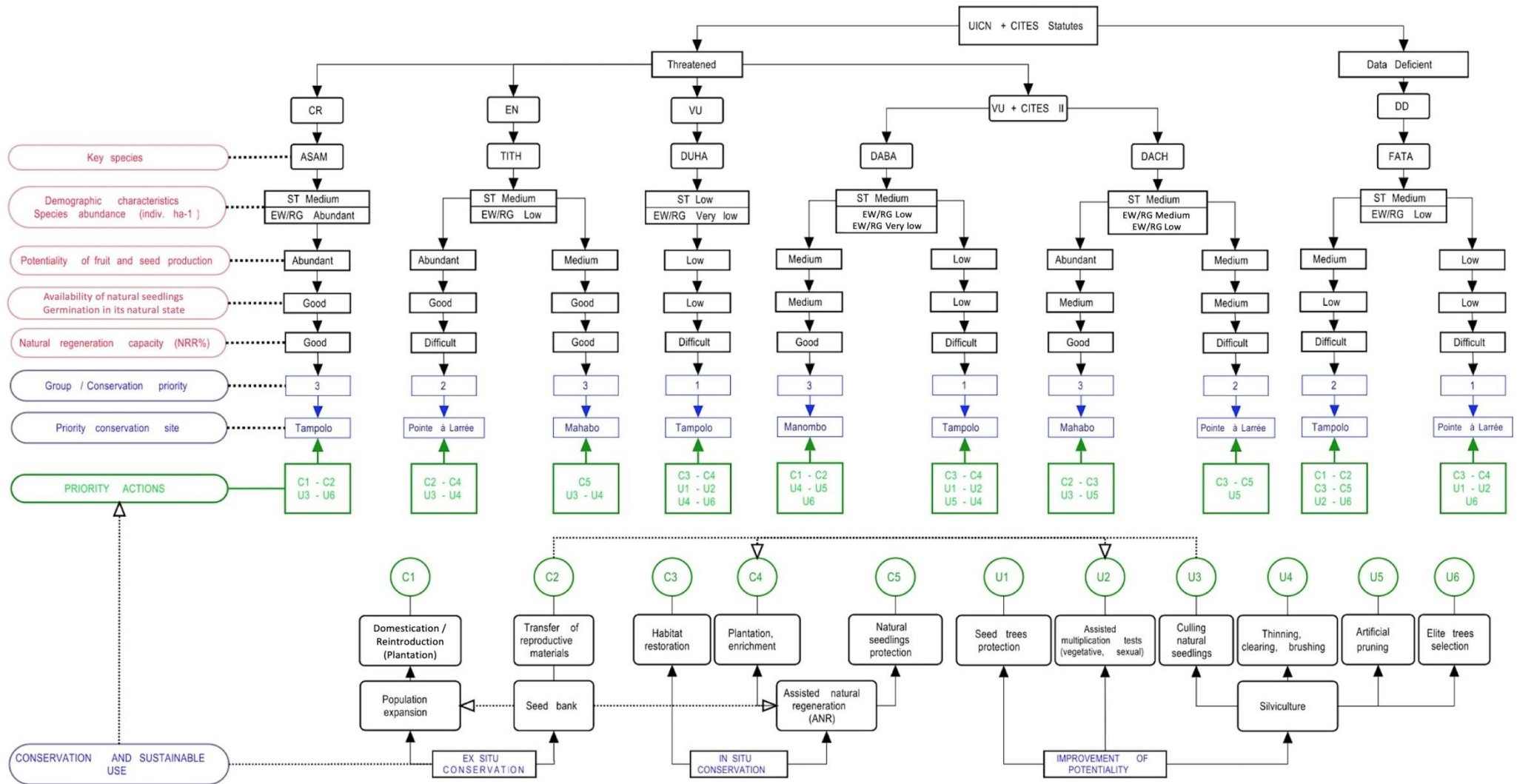


Figure 14. Model for the conservation and sustainable use of key species.

multi-species approaches).

Species conservation actions are focused on two types of measures: "*in-situ conservation*", which aims to maintain the survival, increase and protect the population of target species against extinction by restoring their habitats and conducting natural regeneration or reproduction; and "*ex-situ conservation*", which focuses on the reproduction of species outside their natural habitats. As for sustainable use actions, they consist in improving the potential of key species in terms of production (wood and non-wood forest products) and reproduction.

Conservation actions

With a conservation status of "CR", the populations of *Asteropeia amblyocarpa* are characterized by a very restricted distribution throughout its natural range and have only been found in one site at the NPA Tampolo. In order to ensure the conservation and sustainability of the species, the extension and translocation of existing populations to an additional area of potential habitat is recommended, the installation conditions of which should meet the ecological requirements of the species. The same applies for the restoration of population size and distribution of species at risk to their historical level. If *D. baronii* has previously been encountered in Mahabo, it is essential to restore this species by establishing a new population. However, the recovery of species at their historic stations should not be considered as a replacement for the protection and management of existing stations.

Thus, priority conservation measures essentially consist in maintaining existing populations through "*in-situ*" actions. The first step is to restore the current and future habitats of key species, but also of fauna species that play an important role in their natural reproduction, through enrichment and forest restoration actions by well-adapted and valuable species; then to maintain the current populations of the target species through assisted natural regeneration (ANR) techniques. It consists in materializing and protecting young seedlings from natural formations, and in complementing natural regeneration with artificial regeneration. These actions concern in particular highly (1) and medium (2) priority species.

To this end, the production of seedlings in nurseries meets these various objectives, as a support for "*in-situ*" conservation, but also for the strengthening of "*ex-situ*" populations. However, the establishment of nurseries and plant production activities require the supply of seeds and vegetative fragments, and should take into account the propagation and reproduction techniques, sexual or vegetative, adapted for each target species. Seed germination tests in experimental devices are

recommended for species with low natural germination capacity, namely *D. haraka*, *D. baronii* and *F. tampoloensis*. At the same time, vegetative propagation tests (cuttings, layering) are also recommended for these groups of species, but especially those with low fruit and seed production potential. On the other hand, for groups of species where fruit and grain production are abundant (*A. amblyocarpa*, *D. chapelieri*, *T. thouarsiana*), the "*ex-situ*" conservation of reproductive material in seed banks is one of the best ways to prevent the loss of genetic biodiversity and thus guarantee a future for threatened species (Bacchetta et al., 2006).

For example, not only they make it possible to save seeds of species with delayed germination and whose germination requires a conservation period to emerge, such as *D. haraka*, but they also provide and preserve breeding material from potential new sites for population expansion through domestication and reintroduction. The aim is, therefore, not only to keep a large number of seeds in the bank; but to know the plant material in various aspects in order to guarantee the conservation of a site's biodiversity. However, transfer and relocation should only be considered to increase the size of existing stations, create new populations or restore the areas of occurrence and occupation of target species. In such cases, donor populations should be closely monitored to ensure that they do not decline as a result of the harvesting of individuals or propagating material.

Sustainable use actions

Concerning the priority actions to improve the production and reproduction potential of the target species, specific measures are recommended for parent trees and juveniles. But first, the habitat of these key species must be maintained to satisfy and promote their installation conditions. For this purpose, passive and active restoration actions are recommended. The main techniques of so-called "*passive restoration*" aim to promote natural self-restoration and regeneration through individual silvicultural treatments, and by reducing pressures and habitat degradation factors (e.g., firewalls, protection of species' areas of occurrence, and traditional social convention). As for "*active restoration*", it involves planting and/or enrichment actions in the strict sense of the term, with the aim of restoring the ecological connectivity of forests or enriching fallows with species of interest that are often overexploited by populations.

Different actions may be necessary: temporarily defending the areas of occurrence of seed trees against various biological and anthropogenic threats; promote seed germination and seedling survival of target species and reduce competition through depressing natural seedlings (wildings and/or natural regeneration), selective

thinning, cutting trees or shrubs, brushing work for the benefit of target species with light-demanding character such as *D. haraka*, *D. baronii* and *T. thouarsiana*; and through enrichment planting for shade-tolerant species like *A. amblyocarpa*, *D. chapelieri* and *F. tampoloensis*, in order to make the associated vegetation more dense. They also consist in choosing objective trees (“*elite trees*”) which are individuals of good phenotypic quality, vigorous, in good health and dominant on which is concentrated the production of quality and sufficient production (reproductive material, wood and non-wood forest products). The aim is to select a limited number of trees early among the best subjects in a stand and to carry out silvicultural operations for their benefit. These silvicultural operations, such as artificial pruning, should optimize the use of resources by improving the phenotypic quality of juvenile and adult trees in order to enhance the quality of the wood. The forest technique to be implemented is also part of a dynamic tree silviculture, ensuring that the selected trees are placed in optimal growing conditions. For example, sufficient early thinning is needed for the objective-trees to allow priority removal of dominant bothersome associated trees.

Conclusion

The development and exploitation of scientific knowledge on target species through the analysis of ecological and biological variability across their natural range is of crucial importance in the development and proposal of strategies for the conservation and sustainable use of species. The results obtained through this study made it possible to verify and confirm the three hypotheses initially put forward: the viability of the species varies significantly according to the ecological conditions of their habitats; in the face of this variation, the species adopt particular behaviors; the reproductive biology of the species can be strengthened by assisted conservation measures. To ensure the effective conservation of important, key, threatened and economically valuable species, the complementarity of the species-based approach with the ecosystem-based approach is essential.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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