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Research Article

Intraspecific Morphological Variations among the Populations of *Milicia excelsa*, *Pouteria adolfi-friedericii*, and *Prunus africana* in Different Natural Forests of Southwest Ethiopia

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Plants have the ability to change their morphological and physiological traits in response to environmental variations. The objective of this study was to determine the intraspecific morphological variations among the populations of *M. excelsa*, *P. adolfi-friedericii*, and *P. africana* in southwest Ethiopia. Representative forests were systematically selected, and a total of ten transects of 160 m length were randomly laid at 100 m intervals, and 30 quadrats (20 m by 20 m) were laid along each transect line at 50 m intervals. Stem height, DBH, and bole length of trees for each species were measured in each quadrat. The intraspecific morphological variations among populations of each species were computed using hierarchical clustering and principal component analysis (PCA) with R.4.1.3. A total of 55 trees for *M. excelsa* in four forests, 232 trees for *P. adolfi-friedericii* in eight forests, and 184 trees for *P. africana* in five forests were measured. Accordingly, three, five, and three population clusters were identified for *M. excelsa*, *P. adolfi-friedericii,* and *P. africana*, respectively. The analysis of similarity (ANOSIM) indicated the presence of considerable dissimilarity among population clusters for *M. excelsa* and *P. africana* but was not significant at $p \leq 0.05$ ($R = 0.9$, $p = 0.17$). However, ANOSIM indicated the presence of considerable dissimilarity among population clusters of *P. adolfi-freidericii,* which was significant at $p \leq 0.05$ ($R = 0.9$, $p = 0.03$). Overall, there was a visible morphological variability among the populations of *M. excelsa*, *P. adolfi-friedericii,* and *P. africana* each at the different sites. Therefore, it is important to look for conservation strategies, such as domestication, to maintain and improve the variability and genetic quality among the populations in a wider scale of the ecological and social environment.

1. Introduction

Presently, habitat destruction and fragmentation are the major causes of species extinction [1]. The increasing human populations and attendant land-use intensification (e.g., cultivation, grazing, and urban development) resulted in the loss and subdivision of native habitats, increasing species extinction rates, and lowered species diversity within managed ecosystems. Habitat loss has caused a proportional loss of individuals from the landscape resulting from changes in configuration of habitat such as reduction of habitat patch size and isolation of patches [2, 3].

Natural forests in southwestern Ethiopia are the major sources of livelihood (timber and nontimber forest products) for the communities in the surrounding area [4]. These forests are also the potential sources of economically important timber species such as *Prunus africana*, *Milicia excelsa,* and *Pouteria adolfi-friedericii*. However, in recent years, the conversion of the forest ecosystem into agriculture lands for coffee and tea plantations is increasing. As a result, species such as *Milicia excelsa*, *Pouteria adolfi-friedericii,* and *Prunus africana* are under high pressure due to random cutting and deforestation [5, 6].

Plants have the ability to change their morphological and physiological traits in response to environmental variations [7–9]. As a result, plants tend to adjust the expression of traits such as morphology, growth, structure, function, and metabolism to persevere their adaptability in diverse

environments [9, 10]. Hence, plant populations respond to variable environments by becoming phenotypically plastic and genetically variable [11], provided that phenotypic plasticity itself could be under genetic control and subjected to selective pressure [12]. The adaptation of a species to these variations may produce different morphological and physiological characteristics, resulting in the development of ecotypes.

Milicia excelsa (Welw.) C. Berg is a deciduous tree with a height ranging from 30–50 m and a straight clear bole. The seeds can be stored in airtight containers in a cool dry place for a period of up to 2 years with no significant loss of viability [13, 14]. *Prunus africana* (Hook.f.) Kalkm is an evergreen tree, 10–24 (36 max.) m in height, with a stem diameter of 1 m; bark blackish-brown and rugged; branchlets dotted with breathing spots, brown and corky; twigs that are knobbly [14]. *Pouteria adolfi-friedericii* (Engl.) Baehni is a very tall tree, to 50 m, with a clear straight bole to about 16 m, topped by a rather small dense crown, mature trees buttressed at the base [13, 14]. All the three tree species are socioeconomically important ones and are used for timber, firewood, medicine, etc. [13, 14]. Therefore, the aim of this study was to determine the intramorphological variation (stem height, diameter the breast height (DBH), and bole length) of individual trees and populations of *M. excelsa*, *P. adolfi-friedericii,* and *P. africana* in southwest Ethiopia.

2. Materials and Methods

2.1. The Study Area. The study area includes four zones in southwest Ethiopia: Ilu Ababora, Shaka, Bench Maji, and Kaffa (Figure 1). The descriptions of the study area given below are based on WorldClim version 2.1 climate data [15]. The mapping and extraction of bioclimatic data for the study area was made using ArcMap10.8. The study area covers about 61,591 km^2. The area is located between 5.325–8.967°N and 34.183–36.825°E. The elevation ranges from 407 m to 3308 m above sea level. The mean annual temperature of the study area is 21.1°C, while the minimum annual temperature is 10.7°C and the maximum annual temperature is 28.4°C. The mean annual precipitation is 1624.7 mm, while the minimum annual precipitation is 1096 mm and the maximum annual precipitation is 1968 mm.

2.2. Population Sampling and Measuring Morphometric Parameters. A systematic random quadrat sampling technique was employed, i.e., representative forest habitat areas were identified and systematically selected for sampling purposes (Figure 2). The data were collected between June and August 2019. A total of 10 parallel transect lines (160 m) were laid down across a representative forest habitat area, and 30 quadrats (20 m by 20 m) were laid randomly at 50 m intervals along each transect line (modified from [16–18]). The delimitation of the quadrant numbers to 30 was to satisfy the minimum empirical assumption to run statistical analysis. Then, matured tree population, with DBH ≥2.5 cm and height ≥1.5 m [19], was counted, and three important morphometric parameters (i.e., stem height in meters, diameter at the breast height (DBH) in centimeters, and bole length in meters) were measured. Hence, for trees with DBH ≥2.5 cm and height ≥1.5 m [19], stem height, DBH (at 1.3 m above the ground), and bole length were measured. Moreover, the altitude of each sampling site was recorded. The heights were measured using a clinometer and the DBH was measured using a standard caliper. The altitudes of the sampling sites were recorded using a Garmin eTrex 20 GPS device.

2.3. Data Analysis. The morphological dissimilarities (or diversity) among the different individual trees and populations of the *P. africana*, *M. excelsa,* and *P. adolfi-friedericii* were analyzed using a multivariate data procedure using distance-based agglomerative hierarchical clustering using R.4.1.3. Agglomerative hierarchical clustering is a "bottom-up" approach where each observation starts in its own cluster, and pairs of clusters are merged as one moves up the hierarchy. Moreover, principal component analysis (PCA) ordination was computed using R.4.1.3 based on a correlation matrix to determine the most important morphometric parameters contributing for the variations among the populations of each species among the different forest sites. Data analyses were based on untransformed measurement data, and clustering was done based on the mean value of each morphometric parameter. As stated by Chahal et al. [20], morphometric parameters with high component values on axis 1 (close to 1 or −1) have a high contribution to clustering. Overall, PC1 and PC2 explain greater than 90% of the variance. The greater the value of the main component on the first axis (PC1), the greater the effect of this parameter in clustering. The length of the vectors represents the magnitude of the representation of each variable for each component and the angles between the variables indicate the correlation between them.

3. Results

In this study, a total of 13 forest areas were surveyed for this study (Table 1). *M. excelsa* was recorded in four forest sites, *P. adolfi-friedericii* in eight forest sites, and *P. africana* in five forest sites. Hence, a total of 55 tree stems for *M. excelsa*, 232 tree stems for *P. adolfi-friedericii,* and 184 tree stems for *P. africana* were recorded and measured.

In this study, a total of 13 forest areas were surveyed of which *M. excelsa* was recorded in four forest sites, *P. adolfi-friedericii* in eight forest sites, and *P. africana* in five forest sites. Accordingly, a total of 55 trees for *M. excelsa*, 232 trees for *P. adolfi-friedericii,* and 184 trees for *P. africana* were recorded and measured. Accordingly, three, five, and three population clusters were identified for *M. excelsa*, *P. adolfi-friedericii,* and *P. africana*, respectively (Figures 3–5). The analysis of similarity (ANOSIM) indicated the presence of considerable dissimilarity among population clusters for *M. excelsa* and *P. africana* ($R = 0.9$, $p = 0.17$) which however, was not significant at $p \leq 0.05$. However, ANOSIM indicated the presence of considerable dissimilarity among population

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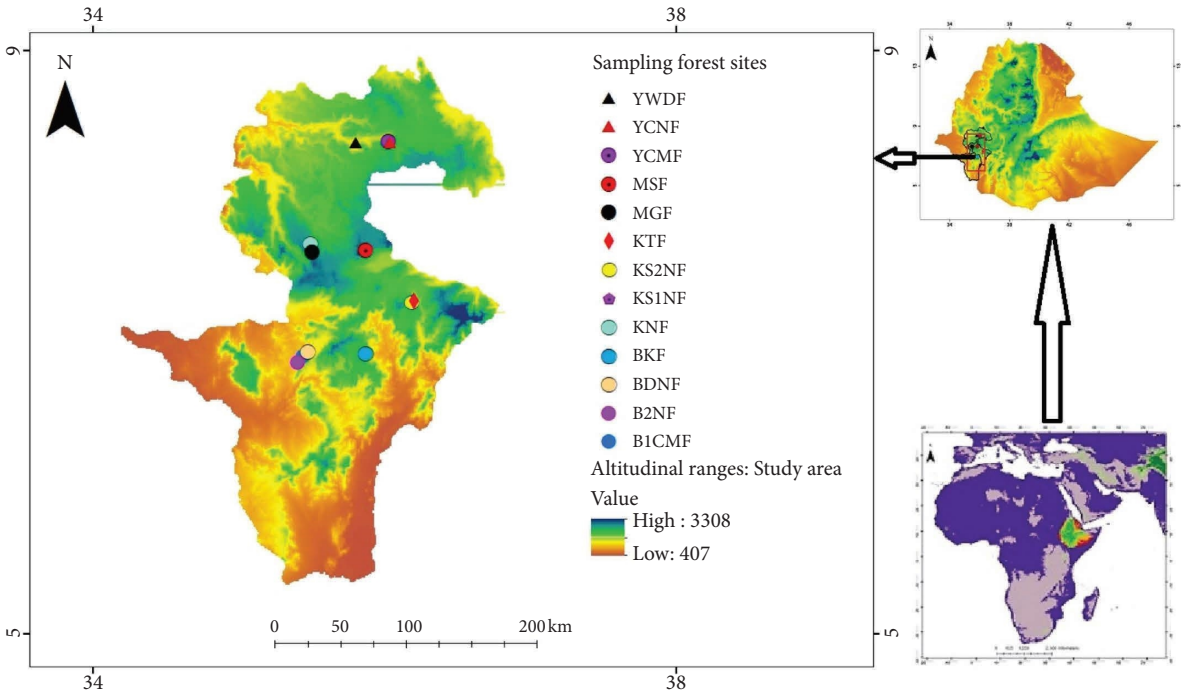

Figure 1: Map the study area including Ilu Aba Bora, Sheka, and Bench-Sheko, and Kaffa zones. YCMF = Yayu coffee mixed forest, B1CMF = Bebeka 1-coffee mixed forest, B2NF = Bebeka 2-natural forest, KS1NF = Kaffa-Shera1 natural forest, KNF = Kahoshemeta natural forest, BDNF = Bebeka-Duduka natural forest, YCNF = Yayu-Chach natural forest, BKF = Bebeka-Kebereta forest, KS2NF = Kaffa-Shera 2 natural forest, MGF = Masha-Gorashewi forest, MSF = Masha-Sherach forest, YWDF = Yayu-Wabu Dureni forest, and KTF = Kaffa-Tejadela forest.

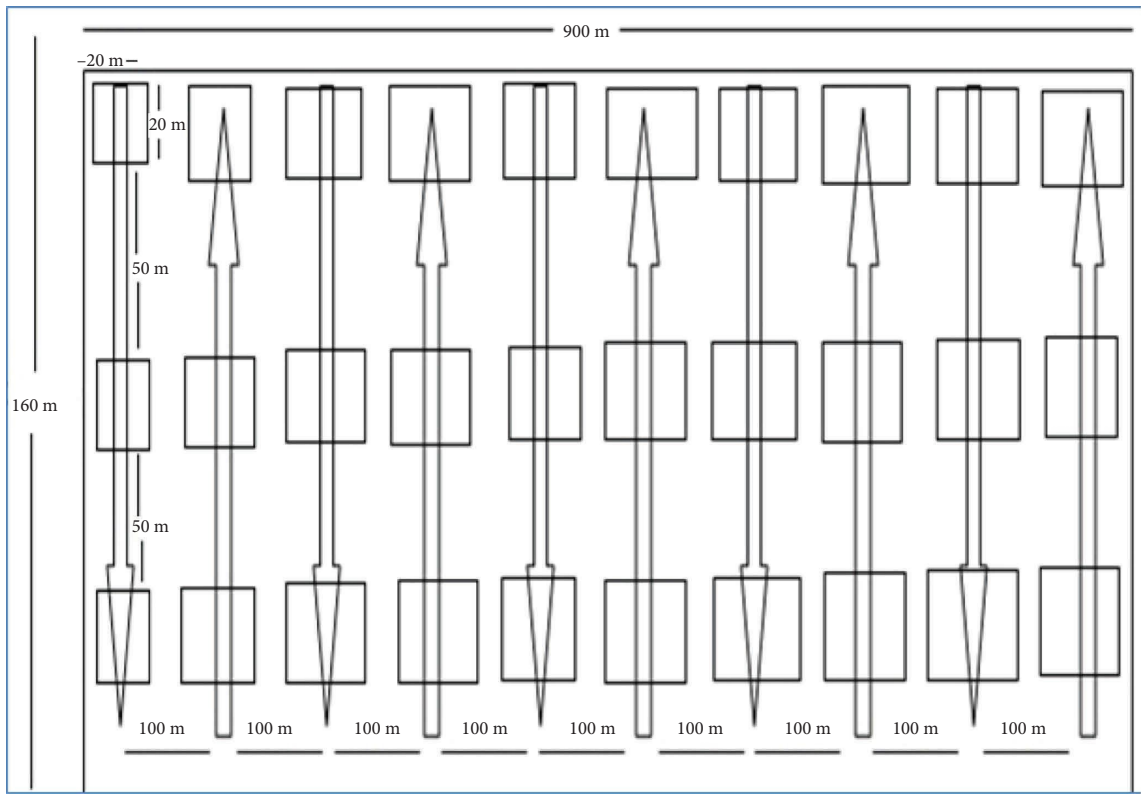

Figure 2: Layout of the sampling plots (transects and quadrats).

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Table 1: Summary of biogeological characteristics of the sampling forest sites. The mean annual temperature and precipitation each forest site are extrapolated from WorldClim version 2.1 climate data [15] using R4.1.3.

Forest name	Forest ID	Location	Altitude (m)	Mean annual temperature (°C)	Mean annual precipitation (mm)	Species recorded
Yayu coffee mixed forest	YCMF	8.37658°N 36.02716°E	1508	20	1744	*M. excelsa* and *P. adolfi-friedericii*
Bebeka 1-coffee mixed forest	B1CMF	6.90377°N 35.44350°E	1070	22	1681	*M. excelsa*
Bebeka 2-natural forest	B2NF	6.85780°N 35.40083°E	1113	32	1685	*P. adolfi-friedericii*
Kaffa-Shera1 natural forest	KS1NF	7.27775°N 36.18269°E	1852	18	1796	*P. adolfi-friedericii*
Kahoshemeta natural forest	KNF	7.672194°N 35.493°E	2354	16	1688	*P. Africana*
Bebeka-Duduka natural forest	BDNF	6.931944°N 35.469528°E	1211	21	1737	*M. excelsa*
Yayu-Chach natural forest	YCNF	8.372833°N 36.035933°E	1479	20	1756	*M. excelsa*
Bebeka-Kebereta forest	BKF	6.918958°N 35.868917°E	1234	21	1855	*P. adolfi-friedericii*
Kaffa-Shera2 natural forest	KS2NF	7.268697°N 36.181056°E	1858	18	1805	*P. adolfi-friedericii* and *P. Africana*
Masha-Gorashewi forest	MGF	7.61835°N 35.50166°E	2440	15	1689	*P. adolfi-friedericii* and *P. Africana*
Masha-Sherach forest	MSF	7.629167°N 35.86941°E	2378	15	1548	*P. adolfi-friedericii* and *P. Africana*
Yayu-Wabu Dureni forest	YWDF	8.369069°N 35.801528°E	1415	20	1668	*P. adolfi-friedericii*
Kaffa-Tejadela forest	KTF	7.27825°N 36.202086°E	1656	19	1838	*P. Africana*

clusters of *P. adolfi-friedericii* ($R = 0.9$, $p = 0.03$) which was significant at $p \leq 0.05$. R value closer to 1 normally indicated that the presence of high dissimilarity among cluster groups, while when closer to 0 implied an even distribution of high and low ranks (or ranges) within and between groups.

Analysis of the principal component analysis (PCA) to determine the effect of morphometric parameters on morphological variations among the populations of *M. excelsa* indicated that DBH has a higher coefficient value (−0.99) in PCA1, and bole length and stem height have coefficient values of 0.80 and 0.58 in PCA2, respectively (Table 2). An ANOVA test of the significance of the effect of parameters showed all morphometric parameters, i.e., DBH ($F = 156.6$, $p \leq 2e - 16$), bole length ($F = 13.34$, $p = 1.82e - 07$) and stem height ($F = 19.27$, $p = 1.2e - 09$) have a highly significant effect on the variation of the populations of *M. excelsa* at $p \leq 0.001^{***}$.

On the other hand, PCA to determine the effect of morphometric parameters on morphological variations among the populations of *P. adolfi-friedericii* revealed that stem height and bole length have higher coefficient values of 0.73 and 0.67 in PCA1, respectively, and DBH has a coefficient value of 0.88 in PCA2 (Table 2). However, an ANOVA test of the significance of the effect of parameters showed that bole length ($F = 109.2$, $p = 7.5e - 05$) has a highly significant effect on the variation of the populations of *P. adolfi-friedericii* at $p \leq 0.001^{***}$. Moreover, PCA to determine the effect of morphometric parameters on morphological variations among the populations of *P. africana* indicated that bole length had a higher coefficient value of −0.99 in PCA1, and stem length with 0.79 coefficient value in PCA2 (Table 2). However, an ANOVA test of the significance of the effect of parameters showed that bole length ($F = 118.7$, $p = 0.008$) has a very significant effect on the variation of the populations of *P. africana* at $p \leq 0.01^{**}$.

Moreover, analysis of the bivariate correlations between the morphometric parameters indicated that the stem height and bole length were positively correlated, while DBH was negatively correlated with both the stem height and DBH for both the populations of *M. excelsa* and *P. africana*. On the contrary, DBH was positively correlated with stem height while negatively correlated with bole length in the population of *P. adolfi-friedericii* (Figure 6).

4. Discussion

This pilot survey analyzed the populations of *M. excelsa*, *P. adolfi-friedericii*, and *P. africana* in 13 natural forests in southwest Ethiopia. A total of 55 trees of *M. excelsa* from four forest sites, 232 trees of *P. adolfi-friedericii* from eight forest sites, and 184 trees of *P. africana* from five forest sites were randomly sampled and measured for their stem height, DBH, and bole length (Table 1). Analysis of the AHC of the

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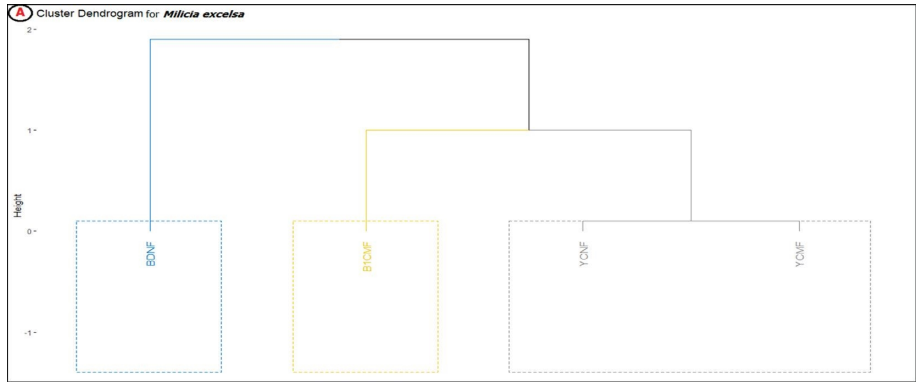

FIGURE 3: Hierarchical clustering of the populations of *M. excelsa* based on one scaled morphometric data using "Euclidean" distance and "Wards.D2" method. YCMF = Yayu coffee mixed forest, B1CMF = Bebeka 1-coffee mixed forest, BDNF = Bebeka-Duduka natural forest, YCNF = Yayu-Chach natural forest.

population of *M. excelsa* showed three possible population clusters (Figure 3). The analysis of similarity (ANOSIM) indicated the presence of considerable dissimilarity among population clusters for *M. excelsa*, but was not significant at $p \leq 0.05$ ($R = 0.9$, $p = 0.17$). The R value of 1 showed that more of the populations within clusters were similar to each other and dissimilar to the populations in other clusters [21]. A study by Ouinsavi and Sokpon [22] also identified four distinct population clusters of *M. excelsa* across the different bio-geographical zones in Benin based on eight morphometric parameters. Nevertheless, the type and the number of parameters included in a study determine the number of possible population clusters in a given bio-geographical area. Moreover, the number of populations of a given species depends on the heterogeneity of biogeography where the scope of the study is focused. On the other hand, ANOVA tests of significance for the relative contributions of morphometric parameters in clustering the population of *M. excelsa* showed that DBH, stem height, and bole length was all important with highly significant effects at $p \leq 0.001^{***}$. In other study, Ouinsavi and Sokpon [22] reported that stem height was found to show significant morphological variation in the *Milicia excelsa* populations across the different biogeographical zones in Benin.

In this study, analysis of the patterns of morphological variation among the populations of *M. excelsa* along the altitudinal gradient indicated that stem height and DBH tends to show contrasting patterns of dynamics along the altitudinal gradient (Figure 7). In this study, larger sizes of DBH and shorter stem heights were recorded between 1050 m and 1210 m above sea level. Moreover, shorter stem heights were also recorded above 1310 m above sea level. In contrast, longer stem heights were observed in the mid-altitude between 1210–1290 m above sea level. Overall, both parameters have appeared to have nearly a contrasting distribution along the altitudinal gradient. Overall, the stem height smoothly increased with increasing altitude and later decreased with increasing altitude making a unimodal normal distribution. However, Christelle et al. [23] also reported that tree height decreased significantly with increasing altitude in their study focusing on tree species diversity on a small montane of Atlantic Central Africa. Furthermore, our results also indicated that DBH tends to decrease with increasing altitude. In the contrary, Pokhrel, and Sherpa [24] reported that DBH showed a small increase with increasing elevation in their study of the tree species diversity on the Chitwan-Annapurna landscape in Nepal. Therefore, our study revealed that DBH and stem height were negatively correlated in the populations of *M. excelsa* (Figure 6). In the contrary, Buba [25] reported that DBH was positively correlated with the tree height while Sumida et al. [26] reported that DBH was negatively correlated with the stem height. These findings suggested that the variability of correlations between DBH and stem height was likely species and habitat-specific. Overall, many studies have indicated that morphological variation is apparently the result of an adaptive response to the environment. For instance, variations in growth and phonological traits are associated with altitudinal ranges [27, 28] or with contrasting climatic conditions [29].

Similarly, AHC of the population of *P. adolfi-friedericii* showed that five population clusters were possible (Figure 4). However, ANOSIM indicated the presence of considerable dissimilarity among population clusters of *P. adolfi-friedericii,* which was significant at $p \leq 0.05$ ($R = 0.9$, $p = 0.03$). On the other hand, an ANOVA test for the relative contributions of parameters in clustering the population of *P. adolfi-friedericii* showed that the effect of bole length is highly significant on the clustering of populations at $p \leq 0.001***$. Moreover, the analysis of the patterns of morphological variation among the populations of *P. adolfi-friedericii* along an altitudinal gradient indicated that the stem height and DBH appeared to show similar patterns of dynamics along altitudinal gradient with bimodal distributions (Figure 8). Here, the maximum measurement scores in both parameters were recorded around an altitude of 1250 m above sea level, while the minimum measurement scores were observed around 1120 m above sea level. A study by Christelle et al. [23] on tree species diversity on a small montane of Atlantic Central Africa showed that the tree height decreased significantly with increasing altitude. However, in this study,

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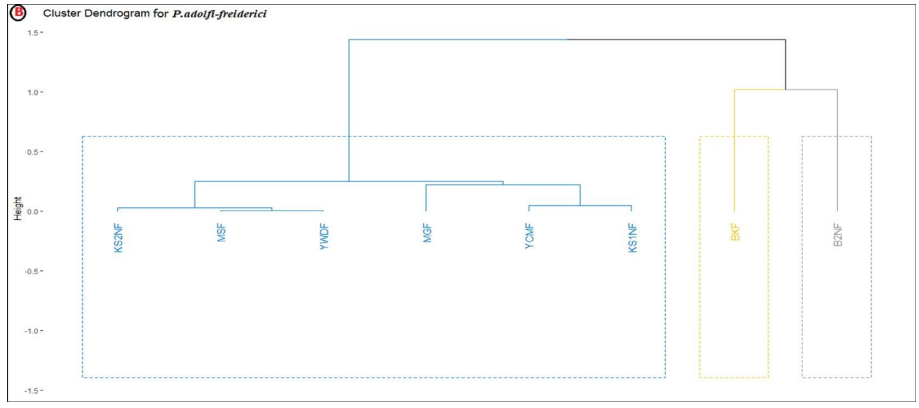

Figure 4: Hierarchical clustering of the populations of *P. adolfi-freiderici* based on one scaled morphometric data using "Euclidean" distance and "wards D2" method. YCMF = Yayu coffee mixed forest, KS1NF = Kaffa-Shera 1 natural forest, KS2NF = Kaffa-Shera 2 natural forest, MGF = Masha-Gorashewi forest, MSF = Masha-Sherach forest, YWDF = Yayu-Wabu Dureni forest, BKF = Bebeka-Kebereta forest, and B2NF = Bebeka 2-natural forest.

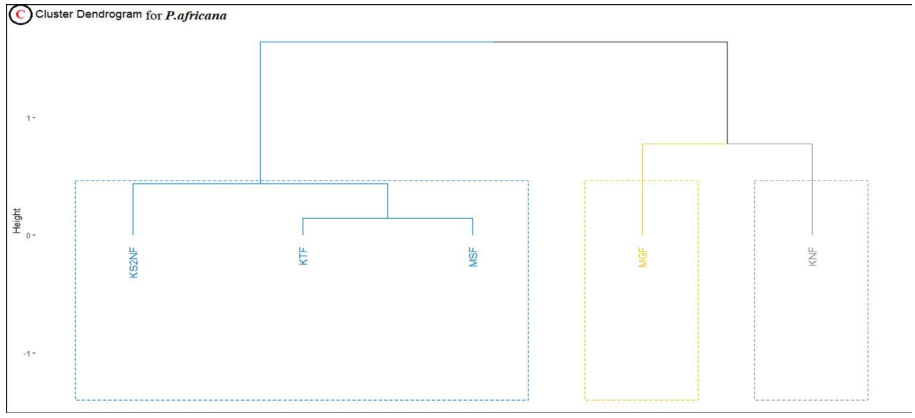

Figure 5: Hierarchical clustering of the populations of *P. africana* based on one scaled morphometric data using "Euclidean" distance and "wards D2" method. KNF = Kahoshemeta natural forest, KS2NF = Kaffa–Shera 2 natural forest, MGF = Masha-Gorashewi forest, MSF = Masha-Sherach forest, and KTF = Kaffa-Tejadela forest.

Table 2: Principal component analysis (PCA) showing the effect of morphometric in clustering the populations of *M. excelsa, P. adolfi-friedericii,* and *P. africana* measured at the different forests.

	M. excelsa		*P. adolfi-friedericii*		*P. africana*	
	PC1	PC2	PC1	PC2	PC1	PC2
Stem height	−0.16	0.58	0.73	0.23	−0.06	0.79
DBH	−0.99	−0.13	0.12	0.88	0.07	−0.59
Bole length	−0.04	0.80	0.67	−0.41	−0.99	−0.09

ANOVA test of the significance of the effect of parameters showed all morphometric parameters, i.e., DBH (F = 156.6, p ≤ 2e − 16), bole length (F = 13.34, p = 1.82e − 07), and stem height (F = 19.27, p = 1.2e − 09) have highly significant effect on the variation of the populations of M. excelsa at $^{***}P \leq 0.001$, i.e., DBH @PCA1, stem height and bole length both @PCA2. However, only bole length (F = 109.2, p = 7.5e − 05) has highly significant effect on the variation of the populations of P. adolfi-friedericii at $^{***}P \leq 0.001$, and similarly, only bole length (F = 118.7, p = 0.008) has a very significant effect on the variation of the populations of P. africana at $^{**}P \leq 0.01$.

the relationship between morphological parameter and altitude is not linear but rather observed to bimodal. Generally, our study showed that DBH and stem height were positively correlated in the population of *P. adolfi-friedericii* (Figure 6). Similarly, Buba [25] also reported that DBH was positively correlated with the tree height, while Sumida et al. [26] reported that DBH was negatively correlated with the stem height.

Furthermore, AHC of the population of *P. africana* showed three possible population clusters (Figure 5). ANOSIM indicated the presence of considerable dissimilarity among population clusters for *P. africana*, but was not significant at $p \leq 0.05$ ($R = 0.9$, $p = 0.17$). A study by Kadu et al. [30] on the multivariate relationships of 25 populations of *P. africana* in the African highlands using nuclear DNA has also identified three distinct (distant) clusters and two overlapping clusters. On the other hand, the analysis of the relative contributions of morphometric parameters in clustering the population of *P. africana* showed that bole length and stem height are important parameters. However, an ANOVA test of the significance of the effect of morphometric parameters showed that bole length has a very significant effect on the clustering of populations at $p \leq 0.01^{**}$. The patterns of morphological variation among the populations of *P. africana* along the

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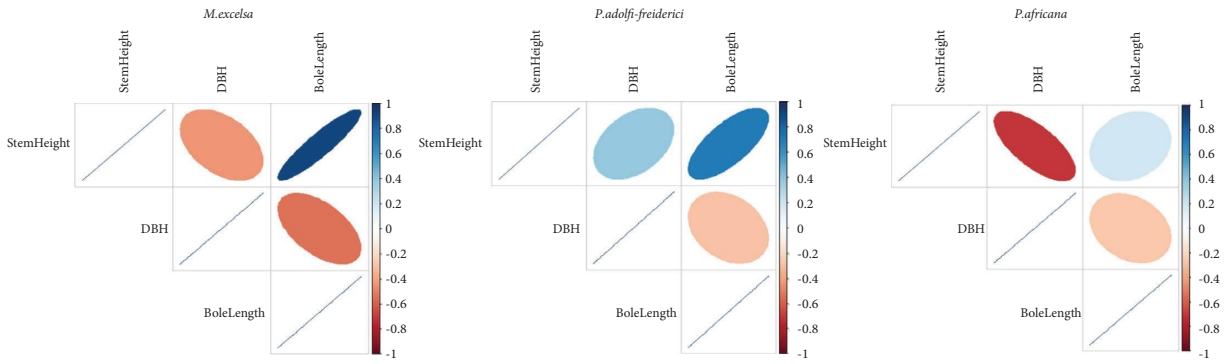

Figure 6: Plot of bivariate correlation ($r = -1$ to 1) between morphometric parameters clustering the populations of *M.excelsa*, *P.adolfi-freidericii,* and *P.africana*. The correlations are presented by colored circles (positive correlation in blue and negative correlation in red).

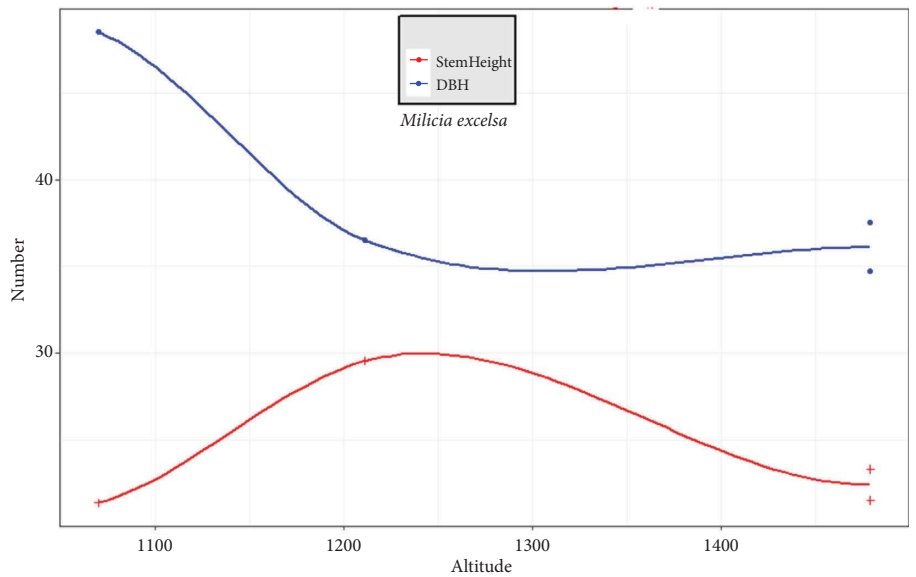

Figure 7: Distribution of stem height and dbh of *M. excelsa* along altitudinal gradients.

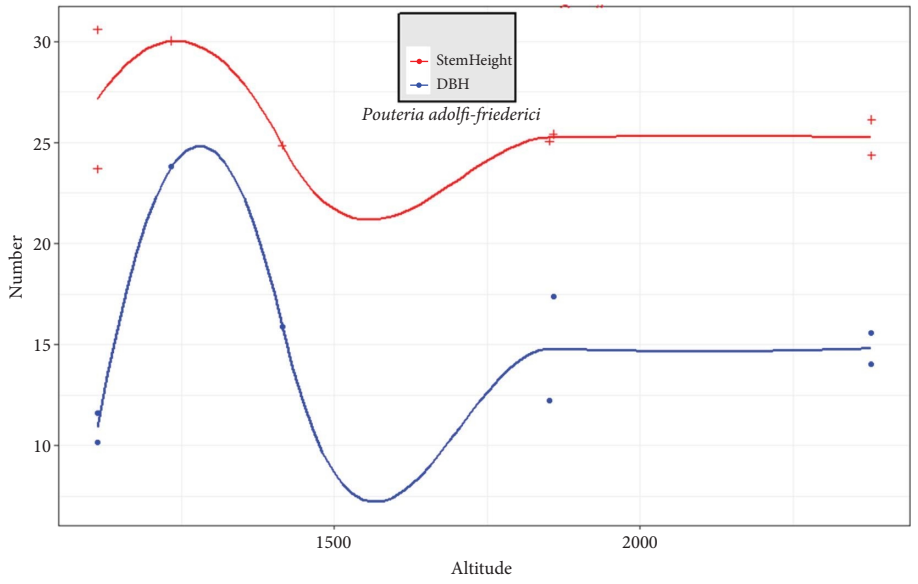

Figure 8: Distribution of the stem height and DBH of *P. adolfi-friedericii* along altitudinal gradients.

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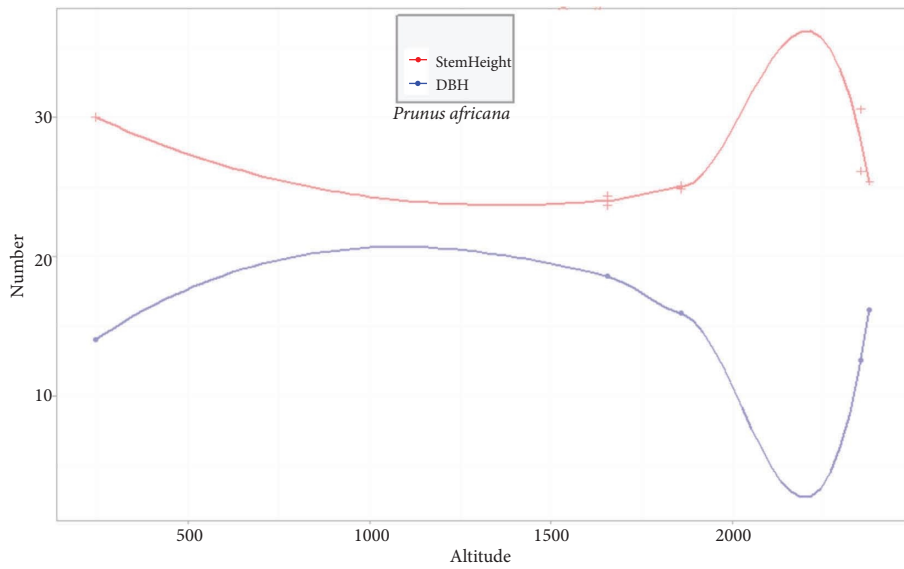

Figure 9: Distribution of stem height and DBH of *P. africana* along altitudinal gradients.

altitudinal gradient showed that stem height and DBH showed contrasting patterns of dynamics along the altitudinal gradient (Figure 9). Generally, our study revealed that the DBH and stem height were negatively correlated in the populations of *P. africana* (Figure 6). In the contrary, Buba [25] reported that DBH was positively correlated with tree height, while Sumida et al. [26] reported that DBH was negatively correlated with stem height. Similar to the other species in this study, the relationship between morphological parameters and altitude is not linear when it comes to *P. africana* as well. For stem height, the maximum measurement score was observed at around 2500 m above sea level.

5. Conclusions

This study analyzed the intraspecific morphological variation among the different populations of *M. excelsa*, *P. adolfi-friederici,* and *P. africana* in different natural forests of southwest Ethiopia. Overall, there was a visible morphological variability among the populations of *M. excelsa*, *P. adolfi-friederici,* and *P. africana* each at the different forest sites. The morphological variability among the populations of *M. excelsa*, *P. adolfi-friederici,* and *P. africana* across the different forest locations has great implications for selection and breeding strategies in the future. Therefore, it is important to look for conservation strategies, such as domestication, to maintain and improve the variability and genetic quality among the populations in a wider scale of the ecological and social environment. These vital indigenous tree species are naturally confined in southwest natural forest in Ethiopia. The species are also found as a remnant of isolated and scattered populations, which are still under severe pressure from human impacts. Hence, the development of management strategies to protect and conserve the remaining wild populations of this valuable species should be effected. In other words, conservation efforts should focus on maintaining large populations to counteract the potential negative effects of drift and promote the maintenance of genetic diversity in these populations.

Data Availability

The data used to support the findings of this study will be made available on reasonable request to the corresponding author.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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