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Morphological Variations between Provenances of *Vachellia abyssinica* Hochst. ex Benth. and *Cordia africana* Lam. from Different Seed Sources in Ethiopia

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Abstract: Plants exhibit phenotypic plasticity, which means they can adapt their morphology in response to changes in the environment. The objective of this study was to assess the extent of morphological variations among different seed sources of *Vachellia abyssinica* and *Cordia africana* in Ethiopia. A total of 26 populations from natural vegetation for *V. abyssinica* and 58 populations for *C. africana* from farmlands were surveyed between 2019 and 2020. We computed hierarchical clustering (HC), one-way ANOVA with a significance level of p≤0.05, and redundancy analysis (RDA). The HC analysis resulted in five cluster groups for *V. abyssinica*, based on parameters such as height and DBH. The one-way ANOVA revealed significant differences in mean height and DBH among these cluster groups. The RDA showed that annual mean temperature (Bio01) and mean diurnal range (Bio02) have a highly significant impact on the morphological variability among the populations of *V. abyssinica* (p=0.001***). Similarly, we identified three cluster groups for *C. africana* based on height and DBH parameters. The one-way ANOVA demonstrated significant differences in mean height and DBH among these cluster groups. The RDA indicated that Bio02 had a very significant effect on the morphological variability among the populations of *C. africana* (p=0.01**), followed by Isothermality (Bio03) with a significant effect (p≤0.05*). The findings of this study can be applied to the management and domestication of *V. abyssinica* and *C. africana*.

Key words: *Vachellia abyssinica, Cordia africana*, provenances, morphology, variability.

Plants have a tendency to change their morphological and physiological traits in response to environmental variations or heterogeneity of the habitats they are living in (Sultan, 1995; Robakowski *et al*., 2003; Pescador *et al*., 2015; Qi *et al*., 2020). Hence, plants tend to adjust the expression of morphological traits, rate of growth, functions, and metabolism to maintain their adaptability in diverse environments (Bradshaw, 1965; Berli *et al*., 2013; Gratani, 2014; Qi *et al*., 2020). There is limited knowledge on the genetic variability of provenances in Ethiopia in general and *V. abyssinica* and *C. africana* in particular. *V. abyssinica* and *C. africana* occur in extensive environmental conditions and altitudinal ranges so that the phenotypic and genotypic variability among provenances of *V. abyssinica* and *C. africana* is expected.

Tree seeds are the key input that determines the success of forestry. However, provision of quality tree seed is often in short supply (Mulawarman *et al*., 2003). Therefore, successful forestry relies on a well-functioning tree seed system that integrates the application of quality seed standards, effective mapping and monitoring of seed sources, and the enhancement of seed research systems. Hence, it is vital to ensure that seeds collections are made from sources with high genetic quality and desirable traits (Roshetko *et al*., 2007). So that, planting trees or shrubs in suitable habitat increases tree production by ensuring higher tree survival and improved good growth performance (Wenying *et al*., 2013). Presently, more than 2200 tree seed sources accounting for more than 85 species are identified and described in Ethiopia (unpublished), by the major stakeholders of seed sourcing and documenting, namely, the Ethiopian Forestry Development (EFD), regional tree seed centers (RTSC), and the Provision of Adequate Tree Seed Portfolios (PATSPO) of ICRAF. However, the intra-specific morphological variations among populations of each species were not analyzed to identify sources with high genetic quality (superior populations) and desirable traits.

Vachellia abyssinica Hochst. ex Benth. (synonym: *Acacia abyssinica*), a Fabaceae, is a large flat-topped tree growing up to 20 meters when mature, commonly known as the pod-bearing species (Negash, 2021). In Ethiopia and other African regions, it is widely distributed in the Dry Afromontane areas at altitudes ranging from 1500 to 2800 m a.s.l. (Bekele-Tesemma, 1993 and 2007; Orwa *et al*., 2009; Negash, 2021. *Cordia africana* Lam. (synonym: Cordia abyssinica R.Br.), a member of the Boraginaceae family, is a much-branched deciduous tree with a rounded crown and often a crooked trunk, growing up to 25 meters tall (Bekele-Tesemma, 1993 and 2007; Orwa *et al*., 2009). In Ethiopia, it is widely distributed in the Dry and Moist Afromontane agroclimatic regions, with altitudes ranging from 900 to 2,500 m a.s.l. It is commonly found in Polyscias and Podocarpus forests, as well as in forest remnants in farmlands (Friis, 1992; Bekele-Tesemma, 1993 and 2007; Orwa *et al*, 2009), and have long

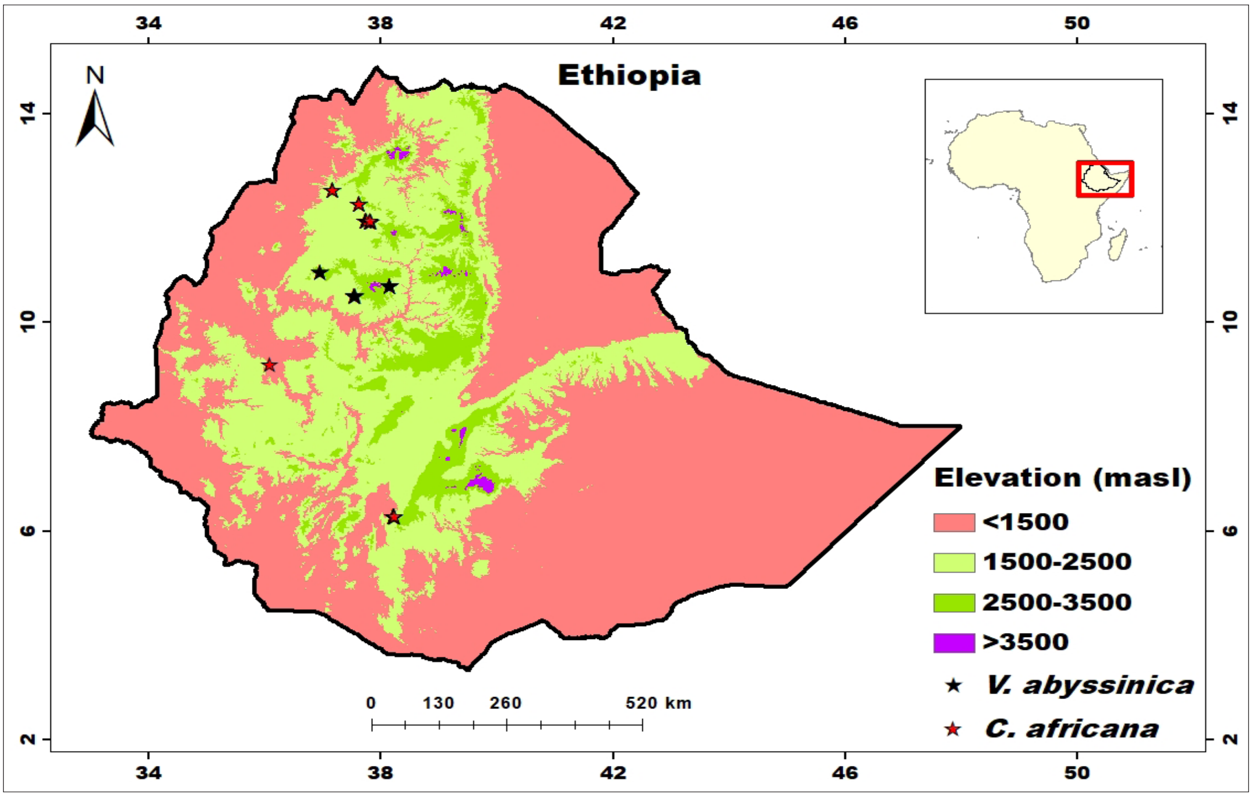

Fig. 1. Map of Ethiopia indicating the sampling sites for V. abyssinica and C. africana.

Table 1. Descriptions of bioclimatic, soil and landscape variables used for ordination analyses.

Label	Variable description	Units
Bio01	Annual mean temperature	Degrees celsius
Bio02	Mean diurnal range [Mean of monthly (max. temp - min. temp)]	Degrees celsius
Bio03	Isothermality (Bio2/Bio7)	Dimensionless
Bio04	Temperature seasonality (Standard deviation)	Degrees celsius
Bio05	Max. temperature of warmest month	Degrees celsius
Bio06	Min. temperature of coldest month	Degrees celsius
Bio07	Temperature annual range (Bio5-Bio6)	Degrees celsius
Bio08	Mean temperature of wettest quarter	Degrees celsius
Bio09	Mean temperature of driest quarter	Degrees celsius
Bio10	Mean temperature of warmest quarter	Degrees celsius
Bio11	Mean temperature of coldest quarter	Degrees celsius
Bio12	Annual precipitation	Milimeters
Bio13	Precipitation of wettest month	Milimeters
Bio14	Precipitation of driest month	Milimeters
Bio15	Precipitation seasonality (Coefficient of variation)	Fraction
Bio16	Precipitation of wettest quarter	Milimeters
Bio17	Precipitation of driest quarter	Milimeters
Bio18	Precipitation of warmest quarter	Milimeters
Bio19	Precipitation of coldest quarter	Milimeters
Elev	Elevation	Meters
Srad	Solar radiation	kJ m^{-2} day^{-1}
Tpi	Topographic index ($\lambda = \ln(\alpha/\tan\beta)$	Unitless

been exploited as a major commercially timber species (Abebe and Holm, 2003).

The objectives of this study were to determine the degree of intra-specific morphological variability, and identifying key environmental factors affecting the intra-specific morphological variations among the populations of *V. abyssinica*, and *C. africana*. Hence, it was hypothesized that *V. abyssinica* and *C. africana* would show intra-specific morphological variations along bioclimatic and physical gradients.

Materials and Methods

Study area: Population sampling was done across selected natural and farmland areas in Ethiopia where *V. abyssinica* and *C. africana* are mainly distributed (Fig. 1).

The study area encompasses the *Acacia-Commiphora* woodland and bushland proper, *Acacia* wooded grassland of the Rift Valley; and *Combretum-Terminalia* woodland and wooded grassland ecosystems of Ethiopia (Friis *et al.*, 2010; Asefa *et al.*, 2020).

Population sampling and measuring morphometric parameters: For this study, a total of 26 natural vegetation populations (provenances) of *V. abyssinica*, and 58 farmlands tree populations (provenances) of *C. africana* were surveyed between 2019 and 2020. So, for each vegetation site (seeds source), ten sampling plots were employed at every 100 m interval randomly. Therefore, one superior (mother) and 3 nearby matured trees with DBH ≥2.5 cm and height ≥1.5m (Tussie, 2004), were sampled for measuring morphometric parameters in each sampling plot. Thus, a total of 40 trees were sampled in each vegetation site. So, two morphometric parameters (*i.e.*, stem height in decimeters (dm), and diameter at the breast height (DBH) in millimeters (mm) were measured.

Extracting environmental variables: The 30s resolution raster data of bioclimatic variables (*i.e.*, Bio01-Bio19), elevation, solar radiation, and topographic index (all averaged for 1970-2000) were downloaded from www.worldclim.com/version2, the Worldclim database (Fick and Hijmans, 2017), and downscaled to the study area (i.e., Ethiopia) using ArcGis 10.8 (Table 1). The values for environmental variables corresponding to each vegetation sites were extracted using the SDM package (Naimi and Araújo, 2016) with the support of other required packages such as Dismo (Hijmans *et al*., 2015), Raster (Hijmans and van Etten, 2012; Hijmans, 2015).

Variables sharing the higher variance (i.e., (>80% total variance) were identified using PCA as recommended by Manly (1994) and Cruz-Cárdenas *et al*. (2014), and less collinear ones ($r \leq 0.6$) were selected (De Marco and Nóbrega 2018).

Data analysis: The intra-specific morphological variations among the populations of *V. abyssinica* and *C. africana* were analyzed using agglomerative hierarchical clustering with "Euclidean" distance and "ward. D2" methods in R 4.1.3. The degree of morphological dissimilarity among population clusters was also determined using box-potting. In box-plotting 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 within and between groups (Clarke and Gorley, 2001). In other words, the closer the R value to 1 showed that more the populations within clusters were similar to each other and dissimilar to the populations in other clusters. Moreover, constrained Redundancy Analysis (RDA) was performed to determine the effects of environmental factors on the morphological variations among the populations of *V. abyssinica* and *C. africana*. Moreover, one-way ANOVA was computed to see the level of significance difference among the clusters for morphological variations at $p \leq 0.05$. Tukey's Honestly Significant Difference (HSD) was computed to assess the significance of difference between pairs of cluster groups at $p \leq 0.05$ (Steel *et al*., 1997), allowing all possible pairwise

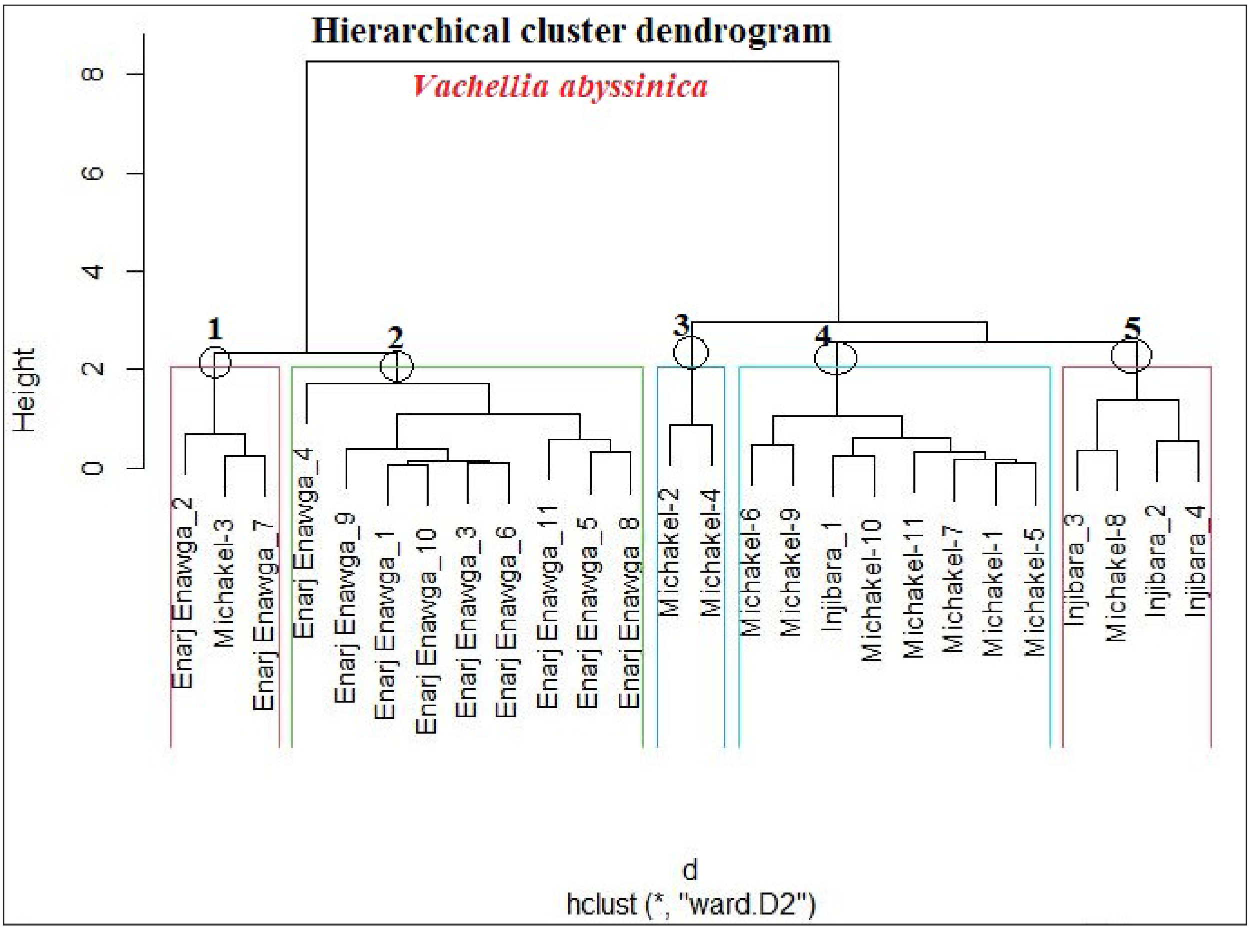

Fig. 2. Hierarchical clustering of the populations of V. abyssinica based on dissimilarity matrix of scaled data using Euclidean distances and Wards.D2 method.

Table 2. One-way ANOVA of the mean significance difference of height (H) and DBH among clusters of the populations of V. abyssinica in the natural vegetations

	Df	Sum sq.	Mean sq.	F-value	Pr(>F)
Height					
Clusters	4	291001	72750	98.43	2.74e-13 ***
Residuals	21	15522	739		
DBH					
Clusters	4	291001	72750	98.43	2.74e-13 ***
Residuals	21	15522	739		

Significance codes: Highly significant '***' if p≤0.001, Very significant '**' if p≤0.01, Significant '*' if p≤0.05.

comparisons while keeping the family-wise error rate low.

Results and Discussion

Morphological variations between and within the species of Vachellia abyssinica

The agglomerative hierarchical cluster analysis for the populations of *V. abyssinica* resulted five conspicuous cluster groups (morphotypes) based on the height (H) and DBH parameters (Fig. 2). The analysis of similarity (ANOSIM) further verified the presence of considerable dissimilarity among cluster groups of *V. abyssinica* ($p=0.001^{***}$, R=0.918) (Fig. 3).

The one-way ANOVA showed that there exists a significant mean difference in height and DBH among cluster groups *V. abyssinica* (Table 2). Tukey's HSD test showed that the largest height and DBH were recorded by cluster-5 (height = 126±*19.0*, DBH = 342±*34.9*), and cluster-4 (height = 318±*32.9*, DBH = 318±*32.9*) that is significant at p≤0.05 (Table 3, Fig. 4).

In other words, except between cluster-4 and cluster-5, there exists a significant mean difference among cluster groups with respect to their height and DBH measurements (Table 3, Fig. 4).

Similarly, the agglomerative hierarchical cluster analysis for the populations of *C. africana* resulted three cluster groups (morphotypes) based on the height and DBH parameters (Fig. 5). The analysis of similarity (ANOSIM) also

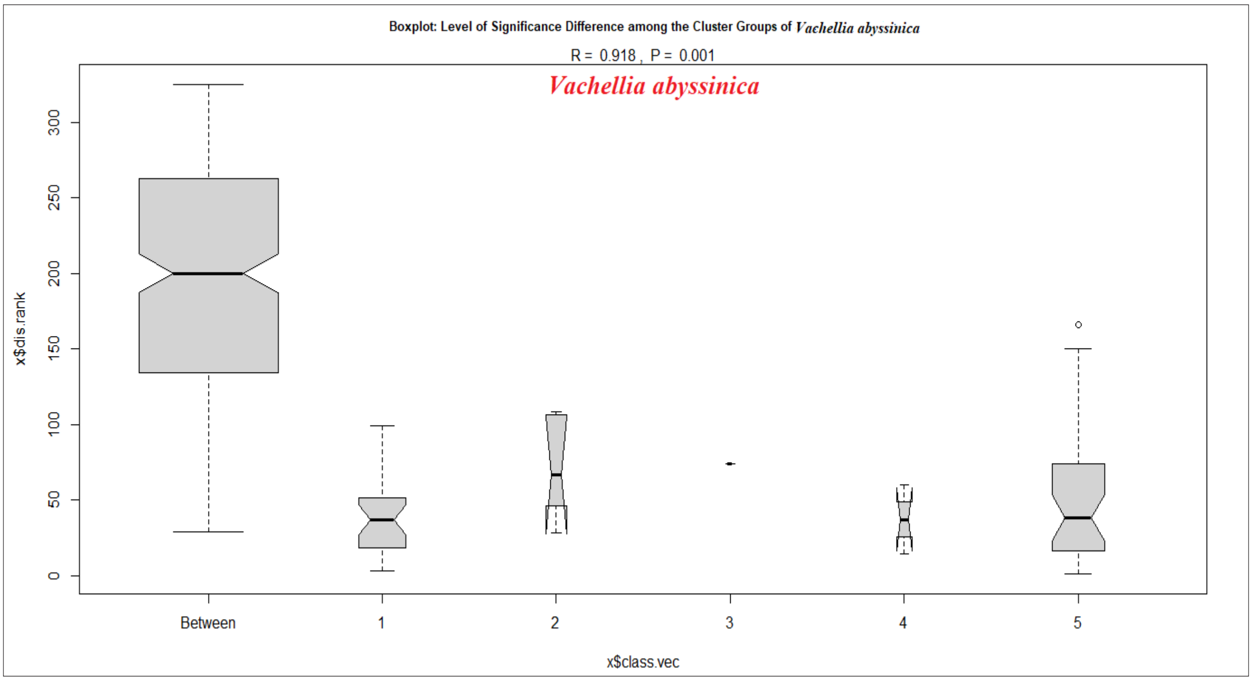

Fig. 3. A boxplot showing the dissimilarity ranks between and within classes in V. abyssinica populations (Number of permutations=999, p=.001).

Table 3. Summary of mean (x̄), standard deviation (SD) and standard error (SE) of height and DBH for cluster groups of V. abyssinica. The significance difference between pairs of treatments at p≤0.05 is calculated using Tukey's HSD test. The values denoted with the same alphabet (s) along the vertical column of each germination parameter are not significantly different at p≤0.05, and vice versa

	x̄±SD	SE
Height		
Cluster-1	55±8.1[a]	4.7
Cluster-2	65±18.4[b]	6.1
Cluster-3	96±48.8[c]	34.5
Cluster-4	105±7.9[d]	2.8
Cluster-5	126±19.0[d]	9.5
DBH		
Cluster-1	264±31.0[a]	17.9
Cluster-2	144±17.1[b]	5.7
Cluster-3	504±4.2[c]	3.0
Cluster-4	318±32.9[d]	11.6
Cluster-5	342±34.9[d]	17.5

verified the presence of noticeable dissimilarity among cluster groups of *C. africana* (p=0.001***, R=0.685) (Fig. 6). The one-way ANOVA showed that there exists a significant mean difference in height and DBH among cluster groups of *C. africana* (Table 4). Tukey's HSD test also showed that the largest height and DBH were recorded by cluster-2 (height = 178±*20.2*, DBH = 312±*49.7*), that is significant at *p*≤0.05 (Table 5, Fig. 7, Fig. 8), followed by cluster-3 (height = 95***±19.0***, DBH = 294±*60.2*).

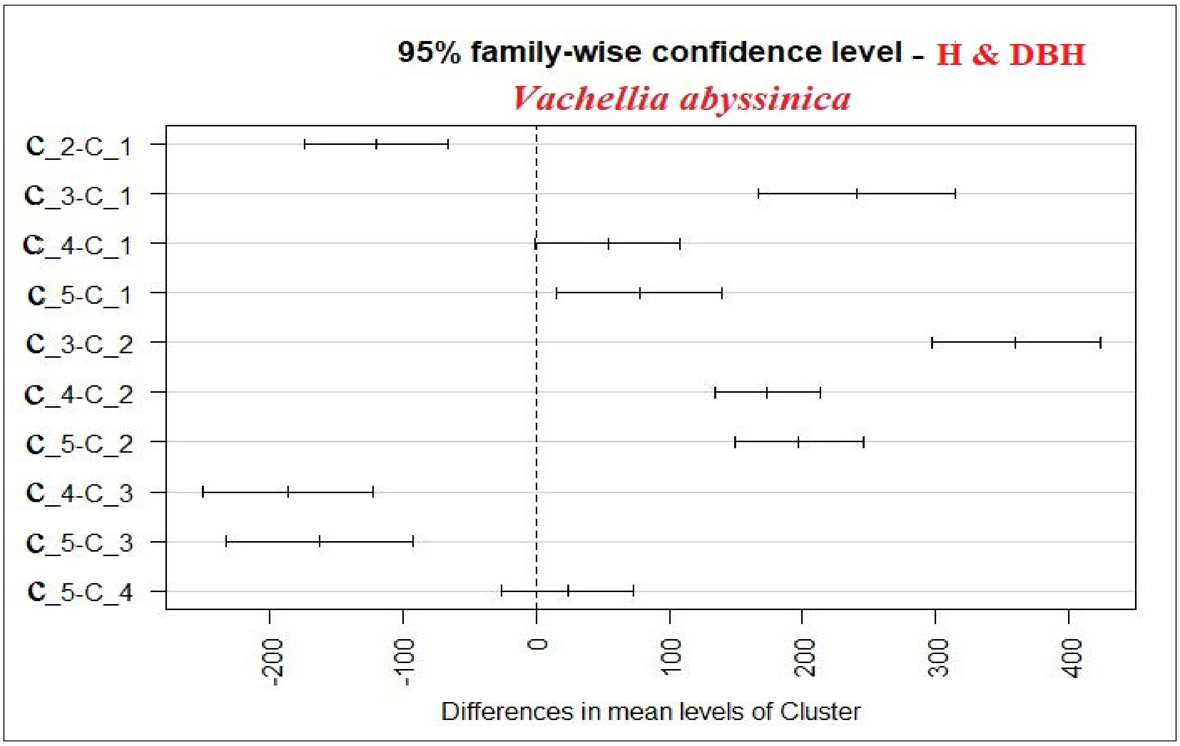

Fig. 4. Tukey's HSD test graph for pair-wise comparison of cluster groups in reference to height and DBH measurements for the populations of V. abyssinica in the natural vegetations. Any confidence intervals that do not contain 0 provide evidence of a difference in the groups (C = cluster, and H= height).

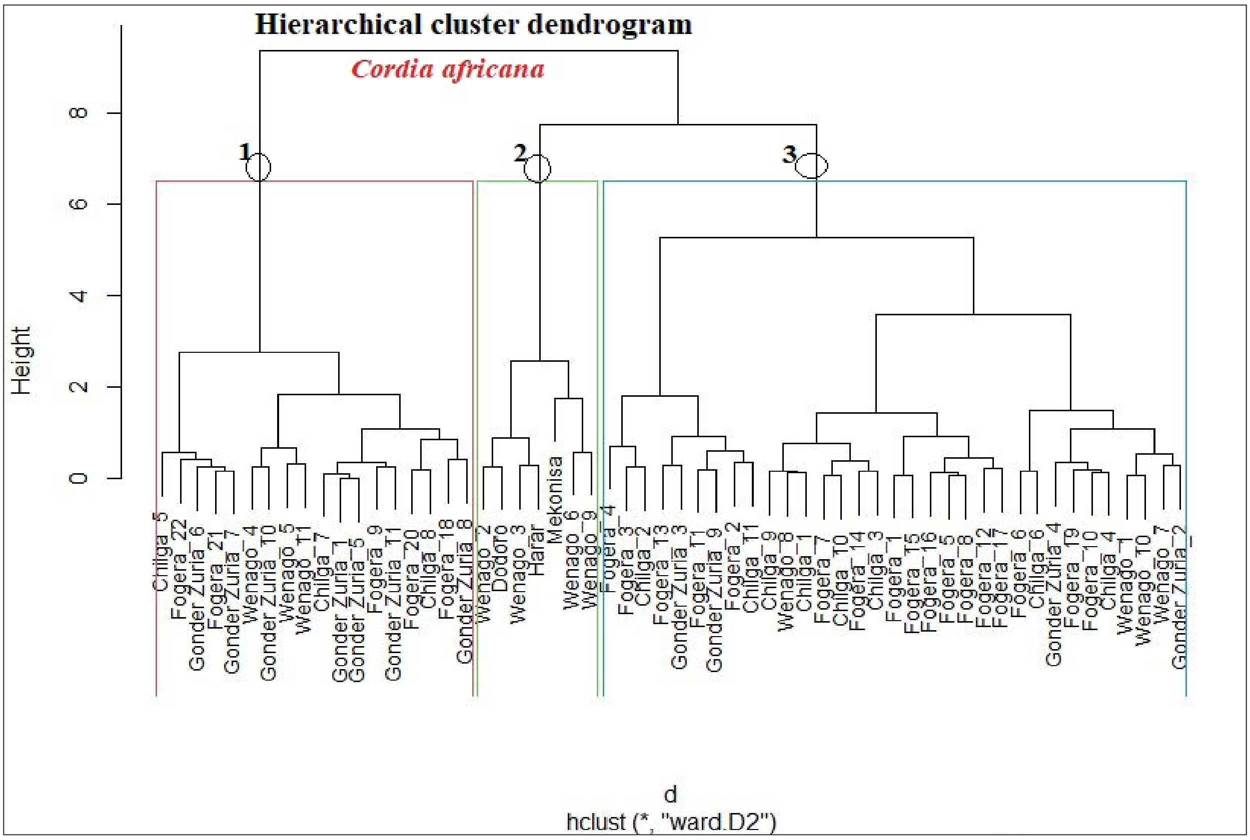

Fig. 5. Hierarchical clustering of the populations of C. africana based on dissimilarity matrix of scaled data using Euclidean distances and Wards.D2 method.

RDA of environmental variables over morphological variations : In this study, RDA showed that the morphological variability within *V. abyssinica* is constrained and affected by mainly Bio01 and Bio02 which are appeared to have highly significant effects on the variability among the populations at p=0.001 (Table 6, Fig. 9).

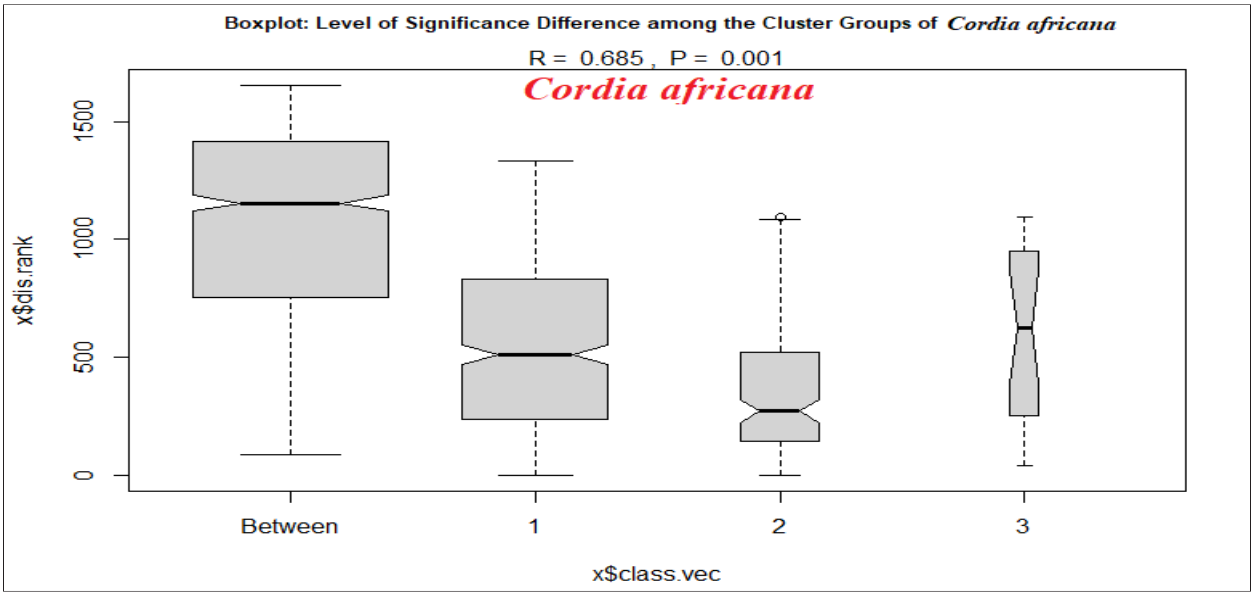

Fig. 6. A boxplot showing the dissimilarity ranks between and within classes in C. africana populations (Number of permutations=999, p=0.001).

Table 4. One-way ANOVA of the mean significance difference of height (H) and DBH among clusters of the populations of C. africana in the natural vegetations

	Df	Sum sq.	Mean sq.	F-value	Pr(>F)
Height					
Clusters	2	50513	25256	51.85	1.81e-13 ***
Residuals	56	27280	487		
DBH					
Clusters	2	164954	82477	26.07	9.97e-09 ***
Residuals	56	177200	3164		

Significance codes: Highly significant '***' if p≤0.001, Very significant '**' if p≤0.01, Significant '*' if p≤0.05.

Table 5. Summary of mean (x̄), standard deviation (SD) and standard error (SE) of height and DBH for cluster groups of C. africana. The significance difference between pairs of treatments at p≤0.05 is calculated using Tukey's HSD test. The values denoted with the same alphabet (s) along the vertical column of each germination parameter are not significantly different at p≤0.05, and vice versa

	x̄±SD	SE
Height		
Cluster-1	80±27.2[a]	6.2
Cluster-2	178±20.2[b]	7.6
Cluster-3	95±19.0[a]	3.3
DBH		
Cluster-1	185±50.8[a]	11.7
Cluster-2	312±49.7[b]	18.8
Cluster-3	294±60.2[b]	10.5

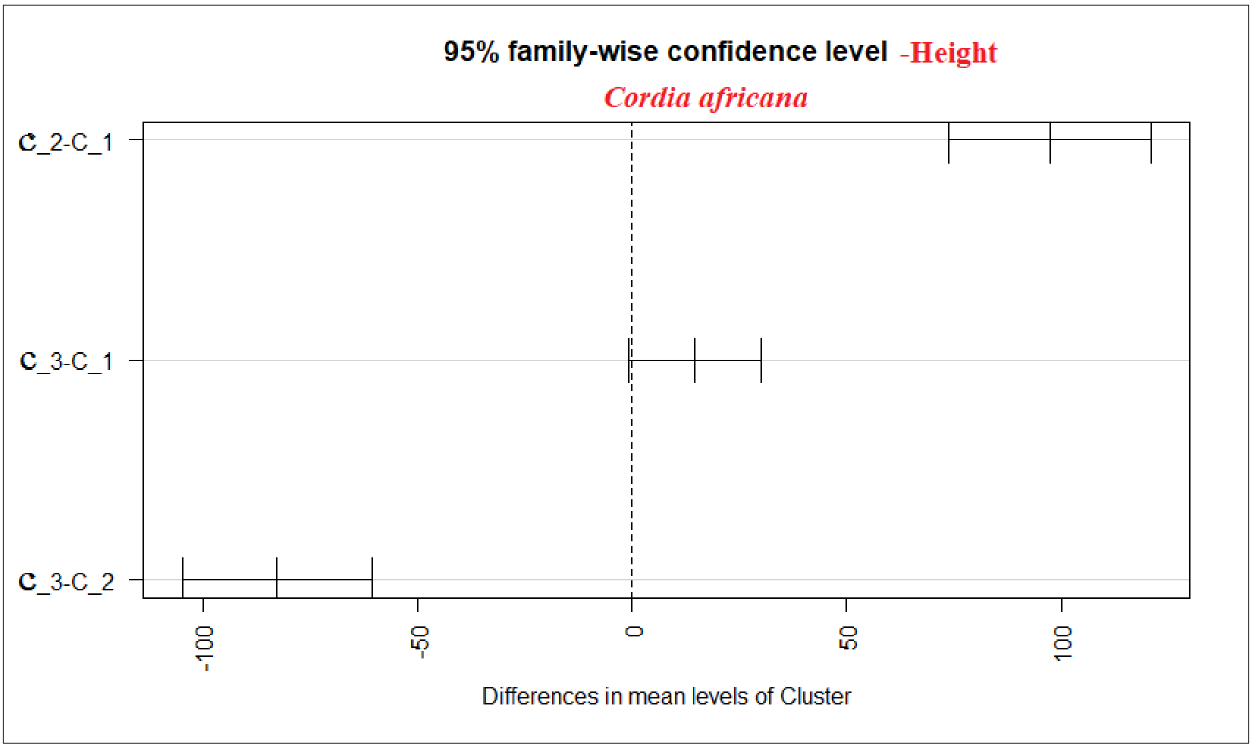

Fig. 7. Tukey's HSD test graph for pair-wise comparison of cluster groups in reference to height measurements for the populations of C. africana in the natural vegetations. Any confidence intervals that do not contain 0 provide evidence of a difference in the groups (C= cluster).

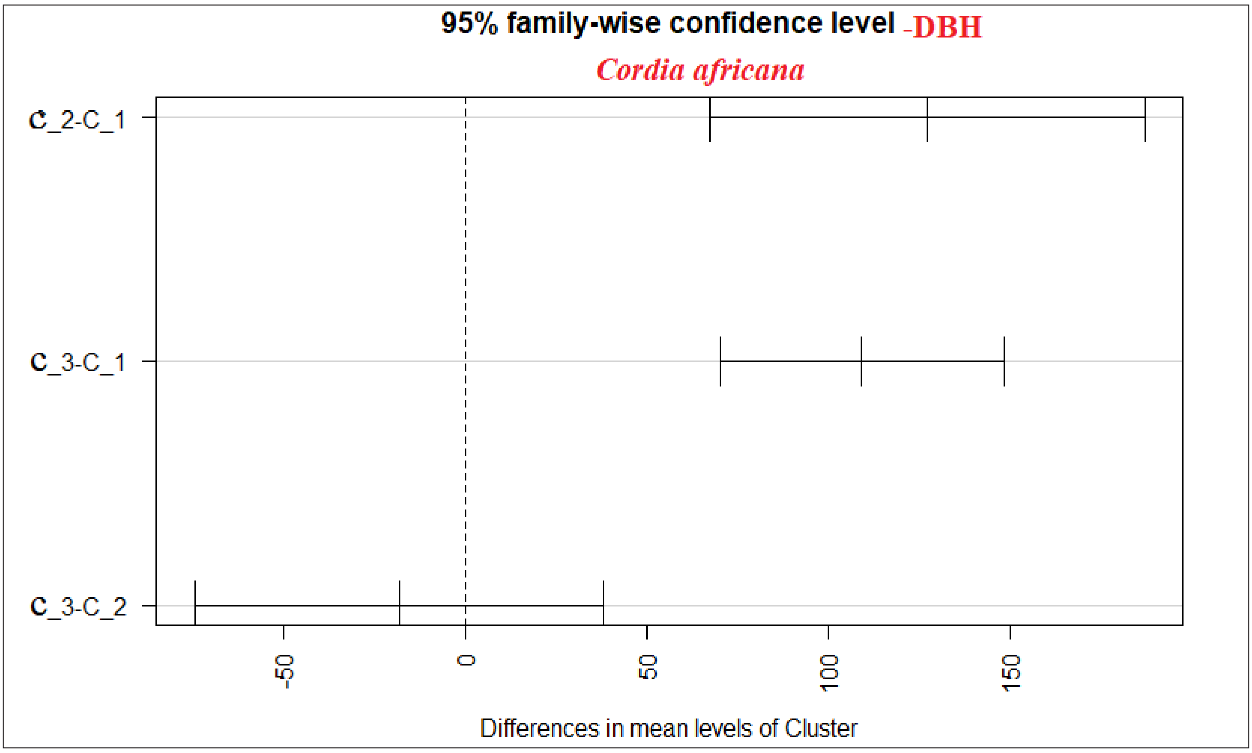

Fig. 8. Tukey's HSD test graph for pair-wise comparison of cluster groups in reference to DBH measurements for the populations of C. africana in the natural vegetations. Any confidence intervals that do not contain 0 provide evidence of a difference in the groups (C= cluster).

Similarly, RDA showed that the morphemically variability within *C. africana* is mainly affected by Bio02 with very significant effects on the variability among the populations at p=0.01, followed by Bio03 that has significant effects on the variability among the populations at p≤0.05 (Table 7, Fig. 10).

Table 6. RDA for important environmental variables effecting the morphological variations among the populations of V. abyssinica (permutations=999, p≤0.05)

	Df	Sums of sq.	Mean sq.	F-model	R^2
Bio01	1	0.3	0.3	25.4	0.001^{***}
Bio02	1	0.5	0.5	44.6	0.001^{***}
Bio03	1	0.0	0.0	1.2	0.299
Residual	22	0.2	0.2		
Total	25	0.9	1.0		

Table 7. RDA for important environmental variables effecting the morphological variations among the populations of C. africana (permutations=999, p≤0.05)

	Df	Sums of sq.	Mean sq.	F-model	R^2
Bio02	1	0.103075	0.086945	7.247929	0.006^{**}
Bio03	1	0.072587	0.061228	5.104096	0.016^{*}
Bio04	1	0.00079	0.000667	0.055574	0.905
Bio06	1	0.005839	0.004926	0.410613	0.636
Bio10	1	0.043387	0.036597	3.05084	0.064
Bio13	1	0.022655	0.01911	1.593041	0.18
Residual	44	0.62574	0.527815		
Total	57	1.185529	1.0		

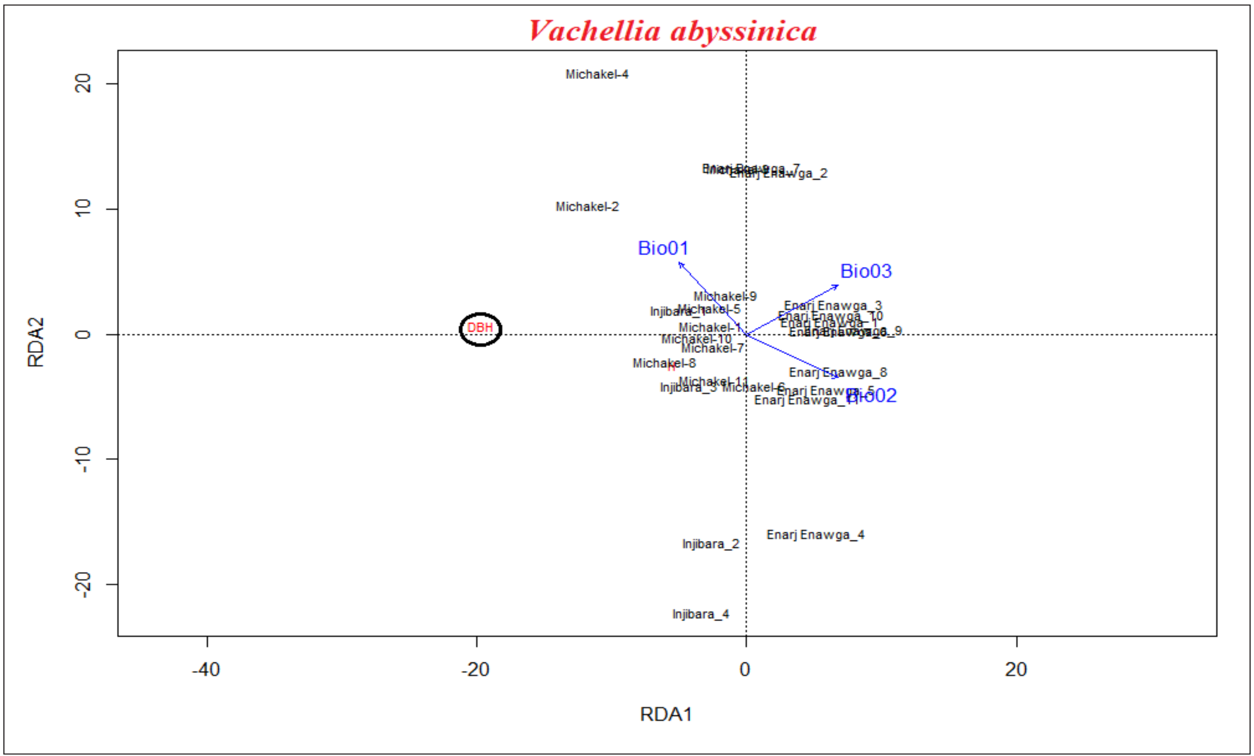

Fig. 9. A constrained RDA triplot of the important environmental variables, morphological parameters, and the population clusters of V. abyssinica.

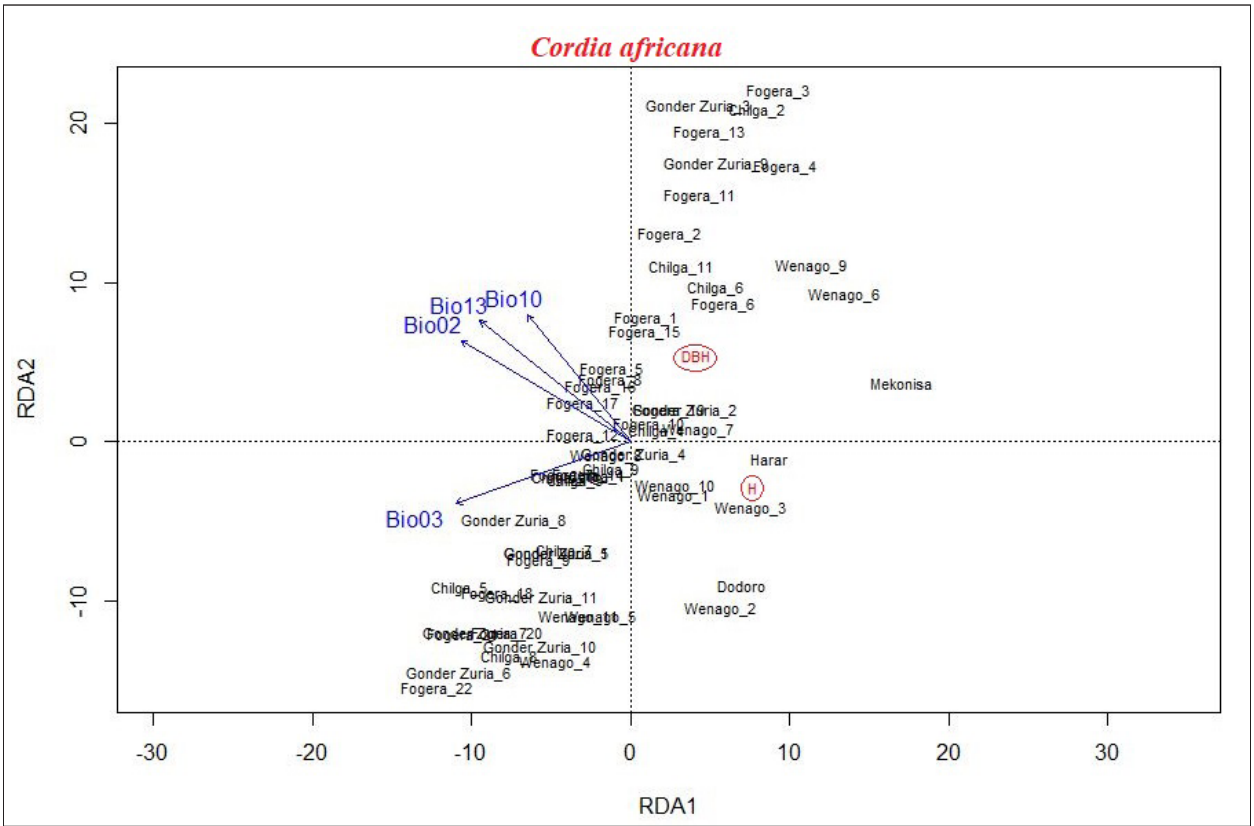

Fig. 10. A constrained RDA triplot of the important environmental variables, morphological parameters, and the population clusters of C. africana.

The majority of populations of *V. abyssinica* studied from seed sources in Michakel district of East Gojjam zone of Amhara region (cluster-5), and Injibara town administration of Awi zone of Amhara region (Cluster-4) are characterized by apparently high morphological superiority presumably with high genetic quality. Similarly, the majority of farmland populations of *C. africana* from Wenago, Dodoro, and Mekonisa districts of Geteo zones in south Ethiopia region, as well as farmlands from Harar city areas show high morphological superiority. A study by Guyassa *et al*. (2013) also indicated the presence of significant morphological variation among six provenances of *C. africana* studied from northern Ethiopia (i.e., Mekelle, Kemisie, Zegie, Finote-selam, Dejen and Tikil-dingay). Therefore, seed collections and establishment of plantation forestry is advised to get sourced from these sites as described by Mulawarman *et al*. (2003) and Roshetko *et al*. (2007), with appropriate selection of site-matching (Lillesø *et al.,* 2011; Nyoka *et al*., 2011). Therefore, plantation forestry of tree species in appropriate site matching would remarkably increase production by ensuring high tree survival rate and good growth successes (Wenying *et al*., 2013).

This study indicated that the morphological variations of *V. abyssinica* is mainly limited by temperature-related bioclimatic variables mainly Bio01 (annual mean temperature) and Bio02 (mean diurnal range), while that of *C. africana* is determined by temperature-related bioclimatic variables, namely, Bio02 and Bio03 (isothermality). These findings have pertinent implications for identifying the most suitable habitat (i.e., site-matching) for plantation and establishment of seed sources for the subject tree species as recommended by Lillesø *et al*. (2011) and Nyoka *et al*. (2011).

It is apparent that natural vegetations show high diversity compared to farmland areas that is also verified in this study where high tree diversity is observed by *V. abyssinica* compared to the *C. africana* studied from the farmland areas. A study by Guyassa *et al*. (2013) also showed that provenances of *C. africana* form natural vegetations are generally superior in their total height and bole length. This is likely related to the selective removal of morphologically superior trees in farmlands (i.e., remnants of natural vegetations) for domestic utilizations. The selective thinning and exploitation of wood products converting natural vegetations into agricultural areas ultimately decrease the tree diversity. This study also indicated that phenotypically and genotypically high-quality trees are present in natural vegetations and protected areas where external pressures are fairly limited.

Conclusions

This study identified the best populations (provenances) of *V. abyssinica* and *C. africana* that can be sourced for seeds of theses tree species for large-scale timber production in Ethiopia. Planting tree in their suitable habitat increases tree growth performance and production forestry. Hence, vegetation areas identified to have high-quality and superior population (provenances) of *V. abyssinica* and *C. africana* can be delineated, managed and maintained as potential seed sources in Ethiopia. Moreover, further field surveys should be done in order to identify additional potential tree seed sources for *V. abyssinica* and *C. africana* in broader landscapes of Ethiopia. Overall, it is also important to carryout provenance trial with appropriate managements to further identify the high-quality provenances through domestication process and different management approaches.

Declaration of funding

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Acknowledgment

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Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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