

Full Length Research Paper

Diversity of *Vepris heterophylla* (Engl.) Letouzey morphotypes in the Sudano-Sahelian zone of Cameroon

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Vepris heterophylla (Engl.) Letouzey (Rutaceae) is a useful and threatened plant of Sudano-Sahelian zone of Cameroon. However, its description is not clear. This study aims to evaluate morphological variations among ten populations to delimitating the *V. heterophylla* morphotypes in the zone. Two hundred and fifty (250) individuals belonging to 10 populations were assessed by morphological descriptors including eight quantitative parameters (Leaf length, leaf width, petiole length, petiolule length, acumen length, fruit major axis and fruit minor axis). The data underwent a principal components analysis (PCA). Results show that the leaf length, leaf width, the petiole length, the petiolule length and the acumen length varied from 54.4 to 102.29, 18.61 to 30.0, 16.44 to 31.16, 1.18 to 2.8 and 1.71 to 4.3 mm, respectively. The fruit major axis, the fruit minor axis and the peduncule length ranged respectively from 1.13 to 1.34, 0.95 to 1.15 and 0.31 to 0.44 mm. PCA generated from grower general dissimilarity coefficient among all populations showed three distinct populations. Grouping of populations did not reflect bioclimatic patterns, this allows saying that the distribution of population could be influenced by environmental conditions. The divergence between the 10 populations based on morphological traits presented in this study constitutes a fundamental element in the improvement of the delimitation of the species morphotype.

Key words: Morphological descriptors, population, *Vepris heterophylla*, Sudano-Sahelian zone, morphotype.

INTRODUCTION

The genus *Vepris* (Comm. ex A. Juss.) (Rutaceae) consists of about 80 species distributed mostly in African continent. Among them about 30 are endemic to Madagascar and one in India. The kinkéliba of Kita is said to reduce high blood pressure and have antipyretic

properties. The genus has been reported to be useful for conjunctivitis, as diuretics, as antihelminthic. Moreover, decoctions from leaves of Kinkeliba of Kita have been used as beverage each evening fasting in West Africa countries (Keita and Ouattara, 1995); the leaves have

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been traditionally used in crop protection for the reduction of post-harvest losses due to insect pests (Ngamo et al., 2007). The essential oil obtained from leaves of Kinkéliba of Kita contain about 35 compounds mainly alkaloids, triterpenes and flavonoids (Matu, 2011). *Vepris heterophylla* is the most important species. It is a shrub native to Sudano-Sahelian countries and grows wild on rocky mountains. It is a shrub of about 5 m high, low-branched, more or less sarmentous, at rounded crown and dense enough (Arbonnier, 2002). It has a weakly continuous range of distribution in the African continent extending from Mali and Ghana in west towards the Africa centre mountains (Geerling, 1987). In Cameroon, the Sudano-Sahelian mountains represent the easternmost limit of its distribution range (Mziray, 1992; Hawthorne, 1995). This reflects the adaptation of plant to diverse soil and climatic conditions such as drought.

In Cameroon, in spite of the great importance of *V. heterophylla*, only little study has been done on its identification. Kinkéliba of Kita is characterized by a wide geographical distribution (Letouzey, 1985). It has a wide variety of synonyms such as *toddaliopsis heterophylla*, *Tecleopsis heterophylla* and *Teclea heterophylla* (Matu, 2011). However, available reports on its description are insufficient to clarify the situation (Geerling, 1987) leading some authors to make reservations on this species. The existence of this species on the IUCN Red List (Ngamo et al., 2007) as an endangered species reinforces the idea that the revision of its description is necessary. In Cameroon, the distribution and the status of the species population were unknown. Little is known about this species including the morphology description, molecular chemistry, multiplication and ethnobotany. The absence of an accurate taxonomy for *V. heterophylla* poses a problem for the botanical classification. The aim of this work is to evaluate the morphological variations among the ten populations of *V. heterophylla* using numerical method.

MATERIALS AND METHODS

Study sites

The study was conducted at the Sudano-Sahelian zone of Cameroon that is situated between the 8 and 13° of the Northern Latitude. It extends from Adamawa to the banks of the Lake Chad on about 100,000 km², that including more than 1/5 of the surface of Cameroon. It included the following mountains: Kalliao, Bilguim, Ndougour, Kotorba, Mokolo, Tinguilin, Lagdo, Nakalba, Fignolé and Tcholliré which are the different habitats of *V. heterophylla* in Cameroon (Geerling, 1987) (Figure 1). The Sudano-Sahelian climate is characterized by two different seasons: the dry season which lasts for 8 months (ranging from October to May) and the short rainy season which lasts for four months (ranging from June to September). The average (\pm standard deviation (SD)) annual rainfall from 1970 to 2010 was evaluated at 748 \pm 120 mm while the mean annual temperature was 28°C. The characteristic of the flora in the region is that of steppes with thorn-bush made up of shrubby savannahs with a very irregular herbaceous floor cover dominated

by thorn-bushes, strongly degraded as the result of human actions (Letouzey, 1985). The rainfall of the region has created a network of permanent (Benue, Deo Logone) and seasonal (Mayo Tsanaga) rivers that flows toward the Lake Chad (Mayo Tsanaga, the Logone) and toward the Katsena (Deo, Benue).

Measurements

Morphometric studies were carried out on 250 individuals belonging to 10 mountains presented in Figure 1 (25 individuals/mountain). The different parameters were obtained on parts which are not parasitized. In all, eight descriptors were employed for each population and data collected were statistically analyzed and recorded. Measurements were taken for eight selected quantitative major descriptors which are: leaf length, leaf width, petiole length, petiolule length, acumen length, fruit major axis, fruit minor axis and peduncule length.

As *V. heterophylla* leaf being a compound leaf, the leaf width and the acumen length were obtained from the average of the width of the leaflets and the average of the acumen length of the leaflets. All these parameters were measured using calipers. The mean, standard deviation and coefficient of variation were calculated for all eight descriptors. Principal component analysis was also carried out on the eight selected quantitative measurements. The objective was to determine the descriptor that contributed strongly to the delimitation of taxa.

RESULTS

The eight morphological descriptors of the *V. heterophylla* populations in Sudano-Sahelian zone of Cameroon were examined with numerical methods. The morphological features employed for delimitation of the 10 populations with their means, standard deviations and coefficients of variation are shown in Table 1.

To assess the patterns of variance, principal coordinate analysis was run considering all the studied variables. The first three principal components explained 89% of the total morphological variation in *V. heterophylla* populations (Table 2). The traits in the principal components (axis) were identified on the basis of eigenvectors (Table 2 and Figure 2).

The first principal component (axis 1) accounted for 42.01% of inertia explaining the largest portion of the variance (Table 3). It is correlated positively with all parameters. The axis 2 accounted 31.28% of the total inertia and was negatively correlated with leaf length, leaf width and petiole length (Table 3). The population plot in the plan defined by the two-first axes showed the separation of population in three distinct groups coded respectively Gp1, Gp2 and Gp3 independently to their geographical origins (Figure 2). The first group comprises three populations Nakalba, Lagdo and Tinguilin. The second group includes four populations which are Bilguim, Kotorba, Fignolé and Mokolo. Populations of Ndougour, Kalliao and Tcholliré constitute the third group situated at the negative side of axis 1.

Similarity matrix based on correlation of *V. heterophylla* populations presented as Table 4 shows that close

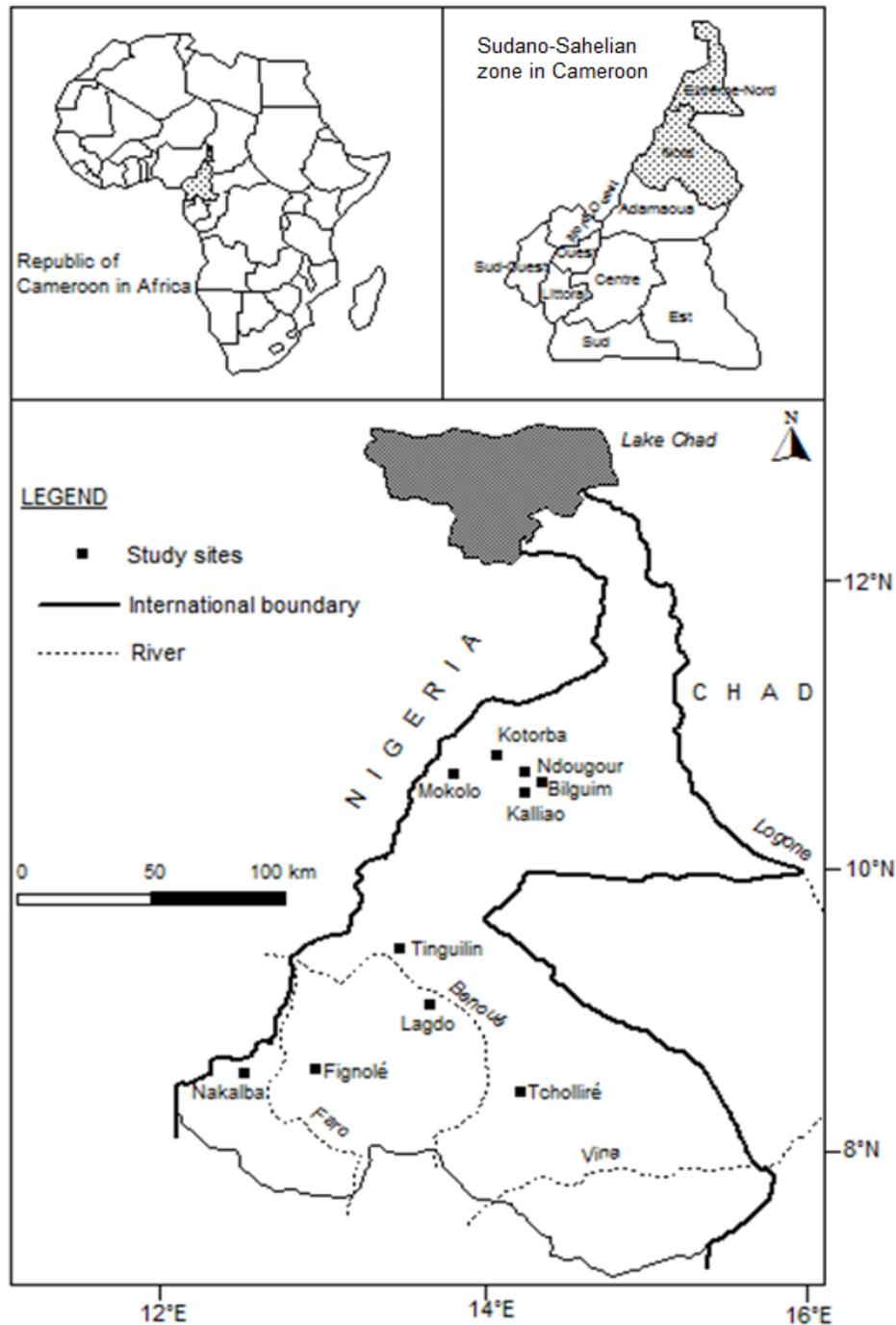


Figure 1. Localization of the study sites.

resemblance of populations could be observed when certain characters are employed. For example, when leaf length was correlated with leaf width, the degree of affinity was 0.965 and 0.117 when correlated with fruit major axis. Similarly when the petiole length was correlated with leaf width, the degree of resemblance was 0.951. It was 0.331 when correlated against peduncule length. Thus, it is shown that there is significant

correlation between leaf length and leaf width, leaf length and fruit major axis, petiole length and leaf width, petiole length and peduncule length. The principal coordinates analysis on morphological descriptors determined on 10 populations shown in Table 2 also revealed that some characters carry more weight in the variation than others indicating the importance of certain characters in the delimitation of the population. It gives the figurative

Table 1. The means, standard deviations of morphometric descriptors used. The coefficients of variation are in parenthesis.

Population	Morphometric character (mm)							
	L. length	L. width	P. length	Ple length	A. length	D	d	Pe. length
Kalliao	54.4 ± 8.3 (15.3)	19.3 ± 3 (18.1)	17 ± 2.7 (16)	1.18 ± 0.9 (53.5)	2.56 ± 1.1 (42.57)	1.31 ± 0.07 (5,1)	1.12 ± 0.05 (4.9)	0.36 ± 0.01 (1.7)
Bilguim	80.3 ± 11.2 (13.9)	25.7 ± 3 (13.6)	23 ± 4.6 (20.1)	1.44 ± 0.5 (36.1)	1.76 ± 0.5 (27.84)	1.15 ± 0.1 (4.5)	0.95 ± 0.2 (19.8)	0.32 ± 0.03 (10)
Ndougou	52.9 ± 7.4 (14)	18.6 ± 3 (18.8)	16.4 ± 2.7 (16.6)	1.38 ± 0.78(63)	2.44 ± 0.9 (37.29)	1.13 ± 0.1 (6.2)	0.95 ± 0.2 (18.9)	0.31 ± 0.03 (9.84)
Kotorba	79.9 ± 12.8 (15)	25.3 ± 4 (14.6)	21.44 ± 5 (24)	1.92 ± 1.4 (71.1)	1.71 ± 0.6 (33.33)	1.23 ± 0.1 (8.)	1.06 ± 0.2 (20.63)	0.37 ± 0.04 (11.3)
Mokolo	75.9 ± 7.4 (13.3)	24.9 ± 3 (15.7)	21.3 ± 2 (11.4)	1.91 ± 1 (34.9)	1.78 ± 1.1 (40.80)	1.34 ± 0.03 (2.5)	1.15 ± 0.04 (3.86)	0.44 ± 0.2 (46.93)
Tinguilin	90.6 ± 8.2 (8.9)	27.9 ± 2.4 (8.5)	30.6 ± 3.4 (11.8)	1.34 ± 1 (62.7)	2.96 ± 0.7 (23.98)	1.31 ± 0.1 (4.7)	1.12 ± 0.06 (4.96)	0.40 ± 0.1 (14.3)
Lagdo	101.1 ± 8.2 (8.9)	29.9 ± 2 (6.5)	30.8 ± 4 (11.5)	1.34 ± 1 (37.3)	2.96 ± 0.5 (16.38)	1.34 ± 0.03 (2.4)	1.15 ± 0.04 (3.86)	0.40 ± 0.4 (98.7)
Nakalba	102.4 ± 6.2 (6.1)	30 ± 2.4 (7.9)	31.16 ± 3 (9.3)	1.82 ± 1 (39.43)	2.98 ± 0.5 (16.77)	1.17 ± 0.1 (4.1)	0.95 ± 0.9 (98.53)	0.35 ± 0.04 (10.8)
Figolé	90.6 ± 16.5 (21.9)	24.5 ± 4.2 (16.9)	22.2 ± 5 (24.7)	1.83 ± 1.4 (58.1)	1.74 ± 0.5 (16.77)	1.31 ± 0.1 (5.1)	1.12 ± 0.06 (5.0)	0.36 ± 0.03 (8.6)
Tcholliré	64 ± 105.6 (109.3)	21.8 ± 4 (18.4)	19 ± 4.6 (20.8)	2.8 ± 1 (53.5)	4.3 ± 0.8 (39.8)	1.31 ± 0.1 (4.7)	1.14 ± 0.06 (5.06)	0.37 ± 0.05 (13.1)
F	2.29	5.80	43.01	3.37	3.83	604.205	625.775	615.748
P-Value	0.0038	0.0000	0.0000	0.0000	0.0000	< 0.0001	< 0.0001	< 0.0001

L. length: Leaf length; L. width: Leaf width; P. length: Petiole length; Ple length: Petiolule length; A. length: Acumen length; D: big axis of fruit; d: minor axis of fruit; Pe: length: Peduncle length; F: test F; each value is the mean ± standard deviation

Table 2. Principal coordinates analysis (PCO) on morphological descriptors determined on 10 *Vepris heterophylla* populations.

Parameter	Axis 1	Axis 2	Axis 3
Eigen values	3.361	2.502	1.268
Percentage	42.014	31.280	15.853
Cumulative percentage	42.014	73.293	89.146
Descriptors	Correlations coefficients		
L. length	0.782	-0.575	0.053
L. width	0.800	-0.576	0.093
P. length	0.781	-0.579	0.129
Ple Length	0.090	0.428	0.696
A. length	0.183	0.296	0.794
D	0.679	0.684	-0.199
D	0.603	0.757	-0.184
Pe. Length	0.796	0.439	-0.225

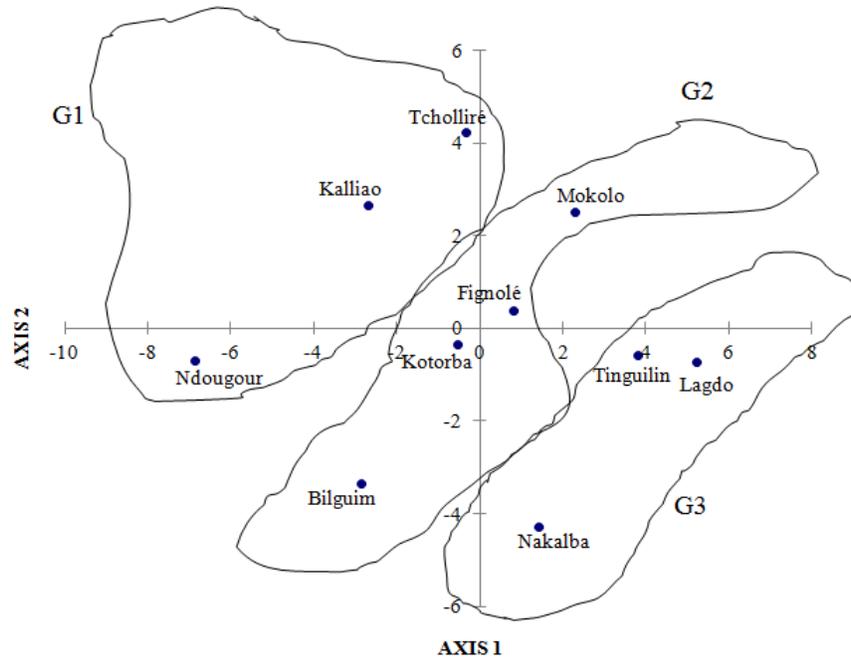


Figure 2: Principal component analysis (PCA) on morphological descriptors data based gower general dissimilarity coefficient determinate on 10 populations of *V. heterophylla* in Sudano-sahelian zone of Cameroon.

Table 3. Similarities matrix based on correlation coefficient of *Vepris heterophylla* populations.

Variable	L. length	L. width	P. length	Ple Length	A. length	D	d	Pe. length
L. length	1							
L. width	0.965	1						
P. length	0.918	0.951	1					
Ple Length	-0.052	-0.057	-0.189	1				
A. length	-0.064	0.004	0.163	0.413	1			
D	0.139	0.117	0.119	0.166	0.207	1		
D	0.040	0.021	0.016	0.216	0.214	0.987	1	
Pe. length	0.319	0.381	0.331	0.162	0.054	0.828	0.797	1

representation of the weight of the characters employed. Those above +1 eigen values are very much stronger than those below. The higher values of cumulative percentage of these characters explain that it weigh higher than the rest five characters employed in the analysis, with eigen values greater than 1.

DISCUSSION

The methods of numerical taxonomy have been used in classifying many plants as well as interpreting results of taxonomic studies (Sonibare et al., 2004; Soladoye et al., 2008). Morphometric analysis is commonly performed on organisms and is particularly useful in analyzing the fossil

record. Morphometrics add a quantitative element to descriptions, allowing more rigorous comparisons. Botanists have usually established differences within species using several morphological traits. These latter are the first step of species identification before the use of molecular markers and other morphological characteristics such as floral traits. In the numerical analysis of 10 populations of *V. heterophylla* utilizing eight quantitative characters, present results confirm that variations in the leaves and fruits characters among the 10 populations are important diagnostic and could be used taxonomically in their delimitation. According to Terzopoulos et al. (2003), some quantitative traits were very important in the evaluation and characterization of broad beans. Equally, Awatef et al. (2013) used the

morphological descriptors for delimiting three groups of *Capparis spinosa* in Tunisia. Significant correlation existing between leaf length and leaf width, petiole length and leaf width, fruit major axis and fruit minor axis shows that these characters were important in the overall analyses. Of the eight parameters used in the analyses, leaf length, Leaf width, petiole length, fruit major axis, fruit minor axis and peduncle length have higher value (above 0.5) than the remaining two characters; affirming their usefulness for delimitation purpose. Comparable deduction had been made in earlier studies reported by Stern (2000) and Soladoye et al. (2010) in the morphological features in taxonomic classification of plants. The quantitative characters used in this study showed significant coefficients of variation in the Sudano-Sahelian zone (Table 1) indicating a multitude of morphotypes of *V. heterophylla* in the zone. The large number of morphotype was supported by the higher coefficient of variation existing within the population of *V. heterophylla*. For these reasons, several authors reported the usefulness of quantitative characters in taxonomic classification of plants (Nwachukwu, 1997; Stern, 2000; Soladoye et al., 2010).

Morphological variation studies are useful to reveal the genetic diversity collections of *V. heterophylla*. Then, various studies are necessary to find the "hidden treasure among the collection that could both allow to the scientific community to answer definitively to the species description problems. For this, the morphological variation seems to be the first step in the description and the classification of population of this endangered species based on statistical methods as principal components analyses.

The PCA analyses showed clearly distinction between populations independently of their geographical origins. Some authors explain this situation by the influence of environmental conditions that express variations such as mutations, hybridization, natural selection (Szamosi et al., 2009). However knowledge of the existing variations between various morphological characters is vital for any plant taxonomic practice. Marquiasfével et al. (2008) in their study of eight Neotropical species of *indifora* noted that gland types and distribution differed between species and that these gland distribution patterns can be used as diagnostic characters. There are many documents on the influence of the environment on phenotypic variation (Abd El-Ghani and Marei, 2007). However, in this present study, the morphological variations would be useful to distinguish three populations. The diversity of morphotypes of *V. heterophylla* in the Sudano-Sahelian zone could be probably also related to an adaptation to environmental conditions. However it is difficult to claim that this adaptation is enough to make distinction between populations. Awatef et al. (2013) had the same difficulties to determine the population of *Capparis spinosa* in Tunisia. In view of this difficulty, it would be interesting to analyze genetic or molecular materials that

can give precise differentiation within the population of *V. heterophylla*. Qualitative observations showed that there are different forms of leaves confirming the high values of the coefficients of variation within the population of *V. heterophylla*.

Conclusion

This study is the first step to the morphological characterization of *V. heterophylla* populations in Sudano-Sahelian zone of Cameroon. Numerical taxonomy with its quantitative feature provided greater discrimination along the spectrum of the taxonomic difference and was also more sensitive in the delimitation of the taxa. In addition to demonstrating the relative value of morphometric methods in the taxonomy of the taxa, the study presents greater and more detailed information on the level of relationship within the genus. It is important to note that, morphometric analysis is not enough in delimiting taxa though it has benefited systematic. Others methods which include anatomical, palynological, cytotoxic and chemotaxonomic differences should be investigated together with morphometric in order to confirm or change the existing classification based on morphology in the genus.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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