MORPHO-ANATOMICAL AND PHYSIOLOGICAL STUDIES OF MORPHOLOGICAL FORMS OF *ERIOSPERMUM ABYSSINICUM* BAKER (ASPARAGACEAE)

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ABSTRACT

Morphology and physiology of the available samples of Eriospermum abyssinicum Bak. in Ilorin, Nigeria were studied using both live and herbarium specimens. Floral and vegetative morphology, as well as the anatomy and stomatal features of the main organs of the plants and transpiration rates were investigated. Some variations were observed from the features studied and therefore, two morphological forms were recognized and identified.

KEY WORDS: Eriospermum abyssinicum, morphology, physiology, taxonomic status

INTRODUCTION

Eriospermum Jacq. ex Willd. is a genus of tuberous flowering plants. It contains about 80-100 species, native to sub-Sahara Africa. In the APG III classification system, the genus is placed in the family Asparagaceae, subfamily Nolinoideae (formerly of the family Ruscaceae) (Chase et al. 2009) and order Asparagales. It was formerly placed in its own family, Eriospermaceae (Perry 1994). Some other botanists placed the genus in the family Liliaceae. The genus consists of popular species like E. abyssinicum, E. capense, E. cooperi, E. corymbosum, E. degree, E. erinum, E. paradoxum, E. Villosum and E. zeyheri. Eriospermum abyssinicum is probably the most common and widespread species of Eriospermum, occurring in both tropical and subtropical Africa. Summarily, the distribution of the plant in Africa goes thus, the East Tropical Africa: Kenya, Tanzania and Uganda; Northeast Tropical Africa: Chad, Ethiopia and Sudan; South Tropical Africa: Angola, Malawi, Mozambique, Zambia and Zimbabwe; Southern Africa: Botswana, Cape Provinces, Free State, KwaZulu-Natal, Namibia, Northern Provinces, Swaziland; West Tropical Africa Benin, Burkina, Ghana, Guinea, Ivory Coast, Mali, Nigeria, Senegal and Togo; and West-Central Tropical Africa: Burundi, Cameroon, Central African Republic, Equatorial Guinea, Rwanda and Zaire (Hepper 1968).

According to the information on the e-monocot website (http://e-monocot.org/), as may be expected of a genus with such a widespread species,

considerable variability exists, especially regarding the length of peduncle and pedicels, stoutness and straightness of contemporary leaf sheath and size of flowers. However, *E. abyssinicum* appears to be the only yellow-flowered species. Due to the appearance of a single leaf after the inflorescence, many herbarium specimens are incomplete and could make identification of specimen difficult. Also, the depresso-globose, white-fleshed tuber with apical growing point and the single, narrowly lanceolate leaf with prominent veins are characteristic. Meanwhile, the lax inflorescence with unusually long pedicels in flower or fruit is the main feature for identification of samples.

The Ilorin population of the *E. abyssinicum* has two distinct morphological forms which need to be studied to confirm their taxonomic status. The species *E. abyssinicum* is commonly called cottonseed lily. It is the most widespread species and highly variable of all the species of the genus *Eriopsermum*. Dormant structures are in the form of plump tubers, which can have white, yellow, pink or red mucilage. Tubers remain underground. The leaves vary in forms such as filiform, bottlebrush, or large, thick blades in various shades of green and gray. Flowers are often inconspicuous and plants produced seeds that are coated in a thick layer of hair. As a result of this there many local variants, this upon further study may deserve taxonomic ranking.

Eriospermum abyssinicum is medicinally useful as molluscicides (Adewumi & Sofowora 1980). *Eriospermum* is thus the only genus of the tribe in West Tropical Africa. West Africa as a whole and Nigeria in particular, only a few genera of the family Asparagaceae have been subjected to analytical studies. One of the reasons for this might be the fact that, very few genera have appreciable representatives in West Africa or Nigeria (Adeyemi 1981). This accounts for the little or no information on *Erispermum*. Thus, there were scanty literatures on it. *Eriospermum abyssinicum* has an east-west distribution across Nigeria. Herbarium records show that it is found in Ilorin, Nupe, Zaria, Mambila Plateau and Yola. In West Africa, it occurs in Northern Nigeria, Ghana, Upper Volta and Guinea (Hepper 1968). However, its north-south distribution range is quite narrow, occurring within latitude 8° 30°N and 9°N. This study is, therefore, to elucidate the morphological and physiological features of available samples or collections of *E. abyssinicum* in Ilorin, Nigeria.

MATERIALS AND METHODS

Fresh mature samples of *E. abyssinicum* (Fig. 1) were harvested from Oyun River bank, at the University of Ilorin, Main Campus, Ilorin, Kwara State, Nigeria as well as from herbarium specimens (Fig. 2) collected from Federal University of Technology, Yola Campus, Yola, Adamawa State, Nigeria.

Qualitative features were studied by visual observation and/or physical touch. Features studied are: leaf shape, venation, apex, base, margin, colour, petiole colour, flower colour, type of inflorescence, corm surface texture, nature of the corm, nature of reproductive shoot, seed colour, seed shape, and hairiness of seed coats. While the quantitative feature were measured using a 30 cm rule graduated in millimetres. The features examined are leaf length, leaf width, leaf area, petiole length, pedicel length, size of corm, number of leaves per corm, height of reproductive shoots and average number of flowers on the raceme. A measurement was taken in the laboratory during the practical work and collection after corms were exhumed from the soil. The measurements of each feature were pooled together and averages calculated. Observations were recorded with tables and photographs where appropriate.

A leaf segment of an area of 1cm square from each specimen was cut and immersed in a concentrated solution of nitric acid or trioxonitrate (v) acid for maceration. The upper (adaxial) and lower (abaxial) surfaces were separated with dissecting needle and forceps, and rinsed with clean water.

A portion of each macerated cuticle was taken for microscopic studies. It was stained in 1% aqueous solution of safranin for about 3-5 min. Excess stain was rinsed off with clean water. The stained cuticle was mounted in glycerin. Observations were made on the microscope to determine: stomatal complex types and their frequencies, stomatal size, stomatal density and stomatal index.

Using 35 fields of view at X40 objective as quadrats, the number of subsidiary cells per stoma was noted to determine the frequency of the different complex types present in each specimen. The frequency of each complex type was expressed as the percentage occurrence of such complex type based on all occurrences.

The stomatal density (SD) was determined as the number of stomata per square millimeter (Ianovici, 2011; Abdulrahaman et al, 2016).

 $SD = Number of stomata in 0.152mm^2 field of view / 0.152.$

Stomatal index (SI) was determined as follows:

 $SI = S/E+S \times 100$

Where: SI = stomatal index

S = number of stomatal per square millimeter

E = number of ordinary epidermal cells per square millimeter.

The mean stomatal size or area of a sample was determined by measuring length and breadth using a micrometer of a sample of 35 stomata using eyepiece micrometer.

A cobalt chloride paper method as described by Oyeleke *et al.* (2004) was used to determine the transpiration rate of each specimen. Strips of filter paper of 2cm x 6cm dimension were cut and immersed in 20% cobalt chloride solution. The strips were thoroughly dried in an oven. The property of cobalt paper is that they are deep blue when dried, but in contact with moisture they turn pink. The dried strips were placed in a sealed, airtight polythene bag and weighed (W1) using mettler balance. It was transferred quickly to the plastic containers and affixed with a string to the marked small branch (of the plant) with leaves. Two dried cobalt papers were placed on the

leaf, one on the upper and the other one on the lower surface of a thick, healthy leaf, and were covered completely with glass slides (this is to determine transpiration from the two surfaces of a dorsiventral leaf). The time (in seconds) taken for the strips to turn pink was noted. Once turned pink, the bag was quickly untied and sealed again, and transferred to the laboratory and weighed (W2). Weight of water transpired was determined as W2 minus W1. The surface area of leaves used was measured (i.e. as described in the mean leaf area above), taking note if the leaf is amphistomatic, when the upper and lower surfaces of the leaf was measured. Transpiration rate was expressed as $mol/m^{-2}/sec^{-1}$.

RESULTS AND DISCUSSIONS

Morphological (Tables 1 and 2), anatomical features including stomata (Fig. 3; Tables 3 and 4) and transpiration rates (Table 4) were investigated in the samples of the plant, *E. abyssinicum*. For taxonomical purpose, the physical and numerical data obtained from the morphological studies, anatomical studies, stomatal complex types and transpiration rates of the two samples were compiled in Tables 1 - 4. These characters were used to separate the plants into morphological forms or samples A (Figs. 1a, b and c) and B (Figs. 1f, g and h). Living plant samples collected from their natural habitats are shown in Figs. 1a, b, f and g while Figs. 1c and h showed the herbarium collections of the two samples. Sample A is longer in length than sample B.

The relationship of observations to each other and to the plants as a whole has long been and still is one of the fundamental concerns of plant morphology. Anatomical studies show that the leaf of the two species is amphistomatic i.e. having stomata on both upper and lower surfaces. The two morphological groups possessed the large epidermal cell size in terms of length and breadth. Based on the stomatal complex types, it was observed that the plant with long leaves have a higher number of stomatal size and stomatal density while the plant with shorter leaves have smaller numbers and when compared with the work done by Metcalfe & Chalk (1988) which says that stomatal size is often correlated with stomatal density such that small stomata give high density and large ones give low density. Based on this work, there is no degree of any kind, which can be due to the fact that all plants are not the same even when grown in the same environment. Stomatal index varied from species to species on both abaxial and adaxial surfaces in all the species. The highest stomatal index was found in the abaxial surface and lowest on the adaxial surface in the two groups. For further details, the two morphological groups were examined externally and measurement were also taken, it was observed that the groups show the same degree of similarities with differences in the measurement as the long group give higher value and the short group give lower value. In terms of transpiration, there is high rate of transpiration in the longer species than the shorter species. In this connection, the principal question is whether plant organs differ essentially from one another or whether they are modifications of one basic type of structure (Esau 1969).



FIGURE 1: Living plants of *Eriospermum abyssinicum* (sample A: a and b) and (sample B: f and g) collected from the natural habitats, herbarium specimens (sample A: c) and (sample B: h), paracytic, diacytic and anisocytic stomata on the leaf surfaces of samples A (d and e) and B (i and j)

Characters/features	Sample A	Sample B
Leaf surface texture	glabrous (smooth)	glabrous (smooth)
Leaf shape	lanceolate	lanceolate-linear
Leaf apex	cuspidate	cuspidate
Leaf base	cuneate	cuneate
Leaf margin	entire	entire
Leaf colour	greenish	greenish
Petiole colour	whitish	whitish
Root system	fibrous root	fibrous root
Types of reproductive shoot	stouter and taller with more longer fruit pedicel	shorter and wiry with fewer fruit pedicel
Corm surface texture	coarse (more scaly)	less coarse (less scaly)
Inflorescence	raceme	raceme
Fruit type	trigonous capsule	trigonous capsule
Flower colour	yellow	yellow-mauve
Seed	black, oblong and covered with a tangle of fluffy hair	black, oblong and covered with a tangle of fluffy hair
lature of vascular system	collateral	collateral
Habit	herbaceous	herbaceous

TABLE 2: Quantitative morphological features in two morphological samples of Eriospermum abyssinicum

	Sample A				Sample B		
Characters	Range	Х	S.D	Range	Х	S.D	
Leaf length (cm)	154 – 10.1a	12.86	1.60	13.4 - 8.1b	10.80	1.82	
Leaf width (cm)	1.2 - 0.4a	0.06	0.24	0.8 - 0.2b	0.46	0.06	
Leaf area (cm ²)	7.76 – 2.04a	4.98	1.33	0.8 - 0.2b	3.19	1.38	
Petiole length (cm)	10.1 - 4.0a	7.30	2.03	5.96 – 1.28b	4.64	1.48	
Pedicel length (cm)	12.9 – 6.2a	8.90	1.84	7.8 - 2.4b	5.93	1.87	
Height of reproductive shoot (cm)	28.4 – 21.1a	24.62	2.02	9.2 - 2.6b	18.95	3.21	
Average number of leaf per corm	1 - 2a	1.4	0.32	1 - 3a	1.7	0.51	
Number of corm	1 - 2a	1.4	0.32	1 - 3a	1.7	0.51	
Number of pedicel	1 – 9a	13.0	28.84	1 - 3b	12.0	3.69	
Floral formula			$P_{3+3}A_6G(2)$				

Means with the same letters along the columns are not significantly different

Table 3: Anatomical features in t	two morpho	logical samp	les of Erios	permum al	byssinicum
		a 1			

	Sample A			Sample B			
Characters	Range	Х	S.D.	Range	Х	S.D.	
Epidermal cell length (µm)	102.2-156.8a	139.37	11.08	102.2-149.8a	135.76	13.39	
Epidermal cell width (µm)	9.8-15.4a	12.80	2.04	5.6-12.6b	8.96	2.69	
Epidermal cell size (µm)	1536.64-2350.04a	1943.43	575.16	838.88-1869.84b	1354.36	515.48	
Stomatal frequency in adaxial epidermis	0-23b	11.67	10.20	16-19a	11.67	10.2	
Stomatal frequency in abaxial epidermis	6-18a	12.00	8.48	9-17a	13.00	5.65	

Means with the same letters along the columns are not significantly different

Sample	Surface	Stomatal	Stomatal	Stomatal index	Stomatal	Transpiration rate $(ma/mm^2/aaa)$
-		complex types	density (mm-)	(%)	size (µm)	(ing/initi-/sec)
Sample A	Abaxial	Diacytic	29.4a	88.82a	643.95a	1.73 x 10 ⁻⁵ a
-		Paracytic				
		Anisocytic				
	Adaxial	Paracytic	27.1b	88.85a		
		Diacytic				
Sample B	Abaxial	Paracytic	25.5c	87.93a	638.795a	7.41 x 10 ⁻⁶ b
		Diacytic				
		Anisocytic				
	Adaxial	Diacytic	22.6d	85.95b		
		Anisocytic				

TABLE 4: Stomatal features and transpiration rate of two morphological samples of Eriospermum. abyssinicum

Means with the same letters along the rows are not significantly different

However, the description of the vegetative and floral parts of angiosperm is one of the most significant aspects in plant identification and this is because the recognition of an individual based principally on shoot, number of leaves and so on has been shown to be more variable and often correlated with the habitat or density differences (Palmbald 1968). Therefore, it is clear that the classification of the observed stands of *Eriospermum* into two groups, external and anatomical observations are justifiable in correlation with the Tables 1 - 3. It is generally believed that the morphological features play a prominent role in orthodox taxonomy that is because of the fact that first impression one notices on any plant is its external appearances. Based on this, it is therefore easier to classify plants into groups as regards to their similarities and differences as observed morphologically. However, in modern taxonomy, morphological characteristics alone are not enough in the classification of plants. Nowadays, other areas have to be considered such as ecological criteria, chemical compositions and chromosomes as an important role in the classification of plants.

Meanwhile, an attempt to resolve the issue of morphological variations in the population, in a related study of the taxon, involving chromosome structure by Adeigbe *et al.* (2013), it was found that *E. abyssinicum* (Ilorin population) could have evolved through changes in the chromosome structure or through natural hybridization between closely related populations. In the present study, even though there are differences between these two groups or samples A and B, in external morphology, anatomical observations and transpiration rate, splitting the species is not yet substantial. Hence, there is a need to look for other correlations with the differences in morphological observations identified between the two groups before any taxonomic conclusion can be made on the plant, *E. abyssinicum*. Further work could be done in molecular, phytochemistry and cytology as well as another aspect of the biology of the plant that may be used as criteria to put the two morphological groups into different taxonomic status.

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