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Micromorphology of Cleome Gynandra L. (Capparaceae) from the Eastern Cape Province, South Africa

Sowunmi, I.L., Oyedeji, O.F., Amadi, J.O., Adebusuyi, G.A., Dunmade, Y. A.
Forestry Research Institute of Nigeria, P.M.B. 5054
Jericho, Ibadan, Nigeria

Abstract:- Many species in the family Capparaceae are economically important as wild vegetables with medicinal and nutritive values. Among the species is Cleome gynandra commonly known as spider plant. Several medicinal uses of the species have been ascribed to its aerial parts. This study investigated the micro morphological features and quantified the elemental composition of the aerial parts of C. gynandra. Fresh leaves, stems and roots of the plant were examined for their anatomy and micromorphological characteristics with the use of light scanning electron microscope. The chemical constituent of the aerial parts was also done using x- ray spectroscopy dispersing energy. It was observed the asymmetrically shaped epidermal cells having cell wall undulating with four subsidiary cells around the stomata. The leaves have anomocyctic stomata which are more distributed in the abaxial surface than the adaxial surface. The average guard cells' length and width in the abaxial surface are 0.09 ± 0.01 mm and 0.08 ± 0.01 mm respectively while that of adaxial surface are 0.1± 0.01 mm and 0.07 ± 0.01 mm respectively. The EDXS microanalysis of the leaf revealed the presence of phosphorus, manganese, iron, calcium, sodium, magnesium, potassium and zinc as the main composition of the crystals in the stomata pores and the mesophyll. The knowledge of the plant's ultra-morphological characteristics and elemental composition can further help in proper identification to ascertain its use for herbal remedy and consumption.

Keywords:- *Scanning electron microscopy, Cleome gynandra, mineral elements, light microscopy, x-ray spectroscopy.*

I. INTRODUCTION

The resurgence of interest in wild vegetable species, especially in their ethnopharmacological potentials has become noteworthy. Wild vegetables serve as essential constituents of diets, enriching the body with minerals, essential fatty acids and vitamins. In addition, they have medicinal values such as antibacterial, hepatoprotective and anticarcinogenic properties [1]. Based on these, people are becoming aware of the inherent nutritional and medicinal status of wild vegetables, thus, the exploitation of these potentials has increased greatly.

Several studies have reported the significance of foliar micromorphological features for taxonomical delimitations of many plants [2], [3], [4] and [5]. Ultramicromorphological features of plants have become essential tools in correct identification and authentication of several plant species. These features are also useful for herbal products standardization in various indigenous medicinal plants [6]. Micromorphological characters of the leaves that have been used in some studies include epidermal cell types, stomata, trichomes, vascular bundle patterns and arrangements [7] [8] [9].

Many species in the family Capparaceae are economically important as wild vegetables with medicinal and nutritive values. One important species in this family is *Cleome gynandra* L. commonly known as spider plant. *Cleome gynandra* is multi-branched herbs which can growth up to 1.5 meter in height with long tap roots and a few secondary roots. Its stems are densely glandular. The leaves are palmately compound with 3-5 leaflets. This plant is endemic to Africa and commonly found in South Africa extending to the Limpopo, the North-West, Gauteng, Mpumalanga, KwaZulu-Natal, Free State and the Northern Cape Provinces [10]. In addition to its edibility as wild vegetable, the species is used for the treatment of sexual impotence, epileptic fits, gastro intestinal disorders and inflammatory conditions [11] [12]. These pharmacological values have been ascribed to the property of its leaves, roots and flowers. *C. gynandra* occurs 2400 m above sea level and requires warm conditions that are not below 15°C. It thrives well on a wide range of soils from sandy loam to loamy clayed soil especially if the soil is deep and well drained having pH range 5.5-7.0 [13].

According to Bunanwan *et al*., [14], the morphology and anatomy of the leaf is the most variable plant organ and micromorphological characters such as the trichomes are occasionally specific for species, genera or even families. Today, there is no published literature on the micromorphology of the aerial parts of *C. gynandra*, especially the variety growing in the Eastern Cape Province of South Africa. The study was carried out to examine the micro morphological features of the aerial parts of *C. gynandra* in light scanning electron microscopes. In addition, the chemical constituent of various parts of the species was estimated using Energy Dispersive X-ray (EDX) analysis. EDX is slowly becoming an essential and strong tool in drug and other plant products standardization.

II. MATERIALS AND METHODS

➢ *Plant material*

The leaves, stems and roots of *C. gynandra* were collected from the Research Farm, University of Fort Hare, Alice with latitudes of 32° 47' 0" S and longitudes of 26° 50' 0" E. The identification of plant was done at the Department of Botany; University of Fort Hare and deposited a voucher

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specimen (LinMed 2013/01) in Giffen's herbarium of the University.

➢ *Leaf epidermal studies (Light microscopy)*

Leaf, stem and root samples of 1- 3 cm were sectioned using a razor blade and observation were made under motic light microscope. The microphotographs were taken with the aids of digital camera mounted on the microscope.

➢ *SEM and Energy Dispersive X-ray Spectroscopy (SEM-EDXS)*

Fresh roots, stems and leaves were segmented into 4- 6 mm in length and placed in 6% glutaraldehyde of pH 7.3 for 12h. Following this, 0.05M sodium cacodylate buffer (pH 7.5) was used to rinse the sections. Each sample was rinsed with dehydrated in a graded series of ethanol 10-100% for 20 min per rinse. Then sections were then dried in a Hitachi HCP-2 critical point dryer and mounted on stubs coated with double-sided carbon coated sputter aluminum and gold palladium (Elko IB-3 Ion Coater). The aerial parts of the plant were sectioned and examined at varying magnifications with the use of JEOL (JSM-6390LV) scanning electron microscope (SEM) ran at 10 – 15 kV accelerated voltage.

The chemical analysis was done by energy dispersive x-ray analyzer coupled to scanning electron microscope (SEM), of Thermo Electron Corporation, 6733B-IUUSN, USA manufacturer. The imaging was done with the use of Noran system six software.

III. RESULTS AND DISCUSSION

➢ Light and scanning electron microscopy

Different features were seen in the plant. These include type of stomata, stomata density and crystal deposits. These are significant characters in differentiating the species. Other characters like mean length of guard cell and width of guard cell, number of subsidiary cells, number of epidermal cells per unit area and guard cell index in both abaxial and adaxial surfaces were also captured. The leaves of *C. gynandra* have high distribution of stomata on both sides of the leaf (Fig. 1A & B).

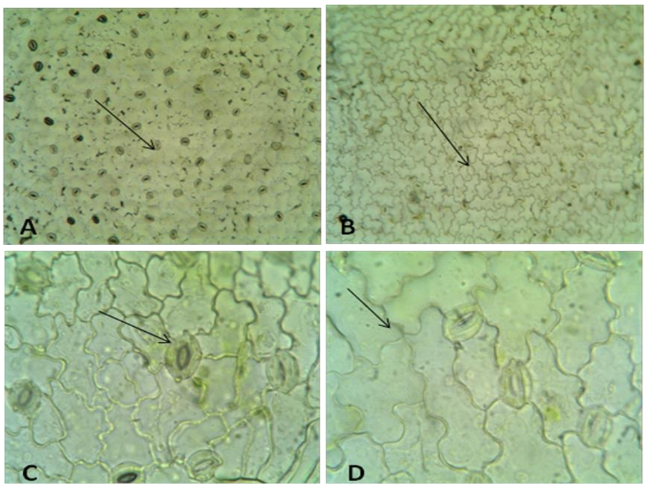

Fig 1:- (A) Shows distribution of stomata on abaxial leaf surface (10X); (B) Shows epidermal cells and stomata distribution on adaxial surface (10X); (C) Shows guard cells and sunken stomata (arrow) in abaxial surface (40X); (D) Shows epidermal cells (arrow) in adaxial surface (40X).

The average stomata densities on the abaxial and adaxial surfaces were 68.14 ± 0.34 and 43.68 ± 0.21 per mm^2 respectively. The amphistomatic features reveal guard cells surrounding sunken stomata. The epidermal cells are assymetrically shaped having cell walls undulating on both surfaces (Fig 1C & D). The epidermal cells density were 163.1± 0.26 and 124.6 ± 0.32 per mm^2 respectively in lower and upper surfaces. The stomata apertures are elliptical (Fig. 2A). The stomas are anomocytic, surrounded by the epidermal layer having four subsidiary cells surrounding each stoma (Fig. 1C & D). The epidermal cells and stomata are more in number at the abaxial surface than the adaxial surface.

The guard cells are seen as banana shaped cells having a thick inner and thin outer wall which are vertically embedded to the subsidiary cells (Fig. 2B). The guard cell index was 5653.7μm^2 and 5496.7μm^2 respectively in the abaxial and adaxial surfaces. The average length and width of the guard cells were 0.09 ± 0.01 mm and 0.08 ± 0.01 mm respectively on the abaxial surface, and 0.1± 0.01 mm and 0.07 ±0.01mm respectively on the adaxial surface.

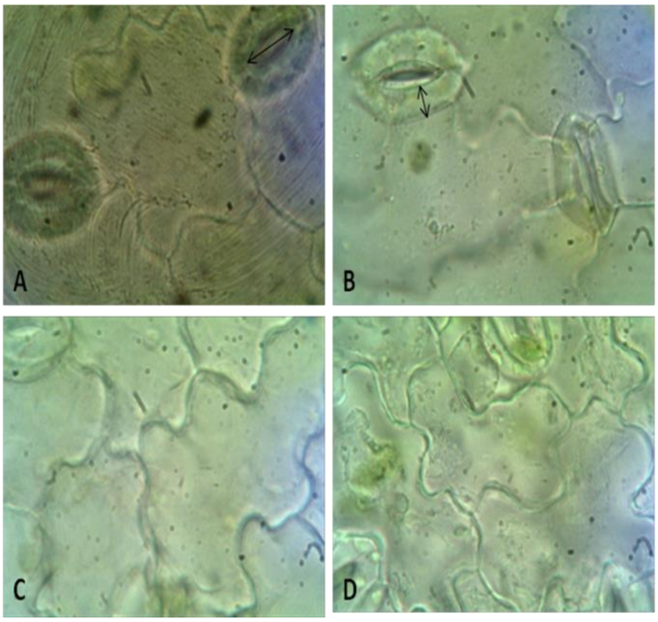

Fig 2:- (A) shows distribution of stomata on the abaxial surface (100X); (B) shows distribution of stomata on the adaxial surface (100X); (C) shows epidermal cells on the adaxial surface (100X); (D) epidermal cells on abaxial surface (100X).

The micromorphology of *C. gynandra* leaves as observed under SEM is presented in Figures 3 A- D, showing the distinguished sunken stomata in the presence of mineral crystals positioned in the stomata pores and in the intercellular spaces on the plant surface.

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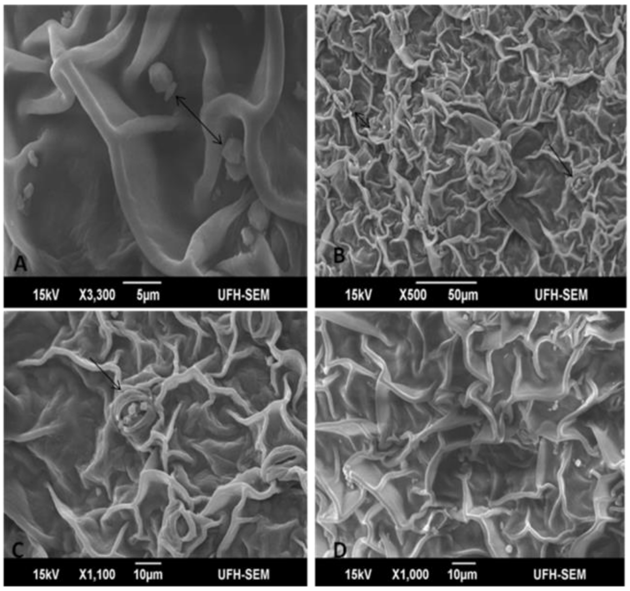

Fig 3:- (A) shows deposit of crystals on the adaxial surface of *C. gynandra* leaves (B) shows sunken stomata having crystal deposits on the abaxial leaf surface (C) shows crystals concentrated in the stomata of the abaxial surface (D) shows fragmented crystals scattered on the abaxial surface.

The micro morphology of *C. gynandra* root is presented in Figure 4A showing the cross section of the xylem tissue vascular bundle in ringed shape. The vessel that transports water, dissolved minerals and fibres that supports the plant is called xylem tissue. The shoot structures showed droplets of mineral sediments on the phloem tissue which are translocated to all parts of the plant (Fig. 4B).

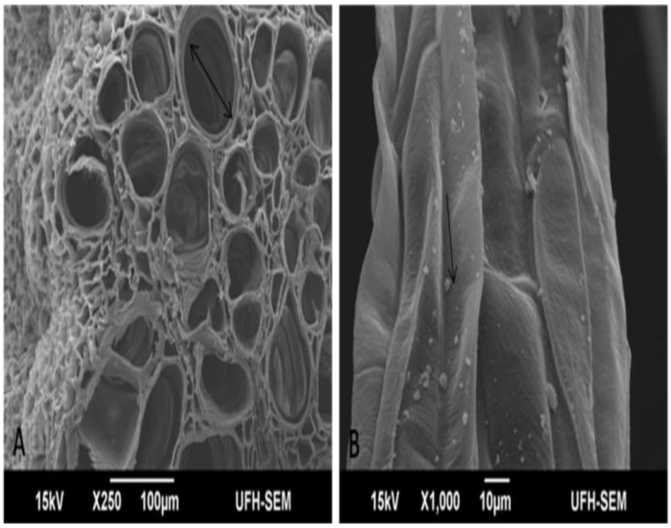

Fig 4:- (A) Cross sections of *C. gynandra* root showing the xylem tissue's inner ring and (B) shoot with mineral deposits on the phleom tissue.

The *C. gynandra* leaf X-ray microanalysis generated a spectra of micro and macro mineral elements such as carbon (C), oxygen (O_2), nitrogen (N), iron (Fe), sodium (Na), magnesium (Mg), phosphorus (P), calcium (Ca), potassium (K), manganese (Mn), zinc (Zn), silicon (Si), sulphur (S) and aluminium (Al) (Fig. 5). Gold (Au) likely come from the spur coater. The mineral composition of this species informed its nutritional and ethnopharmacological importance. For instance, the high carbon, oxygen, nitrogen, phosphorus, manganese, iron and calcium show the abundance amount of these elements while sodium, magnesium, potassium and zinc were in moderate amounts. Aluminium, silicon and sulphur were found in trace amounts. These mineral elements are of great importance in metabolic activities in human development. For example, calcium reduces the risk of contacting various non-communicable diseases and maintenance strength of bones, teeth and muscles in the body [15] [16]. Potassium regulates blood pressure and acid –base balance [17]. Magnesium acts as enzymes co factor involved in energy metabolism; protein production and maintaining the electrical potential of nervous tissues and cell membranes [18]. Iron prevents anemia, increases packed cell volume and boosts the immune system [19]. Also, sulphur is of a vital important in synthesis of protein, regeneration of cells and cleansing of blood [20]. The availability of these elements in plants informs the therapeutic uses of *C. gynandra* in gastro-intestinal disorders, anthelmintic, inflammatory, epileptic fits, aches and pruritis [10].

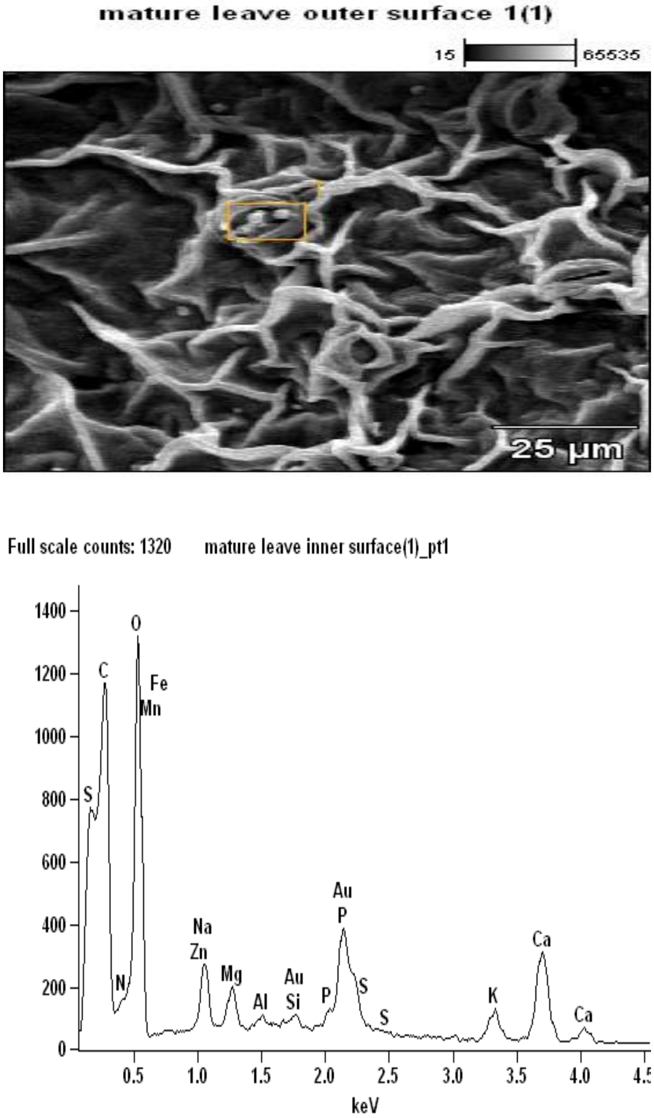

Fig 5:- Showing crystal deposits energy dispersive X-ray analysis in the stomata pore of *C. gynandra*; micrograph shows the peak of the electron beam.

Element	Amount (%)
Carbon	21.36 ± 0.49
Nitrogen	8.01 ± 1.71
Oxygen	50.08 ± 0.93
Sodium	3.69 ± 0.11
Magnesium	1.89 ± 0.08
Calcium	14.98 ± 0.25
Phosphorus	13.53 ± 0.23
Potassium	2.36 ± 0.10
Aluminium	0.49 ± 0.12
Silicon	0.47 ± 0.07
Sulphur	0.49 ± 0.12
Zinc	3.33 ± 0.11
Manganese	11.24 ± 0.17
Iron	15.78 ± 0.21

Table 1:- showing crystal deposits' percentage elemental composition on *C. gynandra* leaf

IV. CONCLUSION

This study examined the micromorphological characters of the aerial parts of *C. gynandra* which includes anomocytic stomata, amphistomatic epidermal surfaces, asymmetric epidermal cells, crystal deposits and the subsidiary cells surrounding the stomata. It found out the availability of micro and macro mineral elements likely informs its high nutritional value and ethnopharmacological importance. The knowledge of this micromorpholgical characters and elemental compositions will help in proper identification of this plant in order to ascertain its uses for herbal remedy and consumption.

V. ACKNOWLEDGMENTS

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