COMPARATIVE MORPH-ANATOMICAL LEAF CHARACTERS OF NERIUM OLEANDER AND CATHARANTHUS ROSEUS FAMILY (APOCYNACEAE)

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ABSTRACT

The present study reports the morphology and anatomy of the leaves of two well known medicinal plants in Sudan; Nerium oleander and Catharanthus roseus family (Apocynaceae). Morphologically, the two studied species have green leaves with entire margins, acute apex and reticulate venation. Radar shape for leaf length, leaf width, leaf apex angle and leaf base angle showed that these characters are correlated for each species. Anatomically, the two species have anisocytic stomata. N. oleander leaf showed larger number of epidermal hairs and four layers of epidermal cells, calcium oxalate crystals, sunken stomata and isobilateral leaves. Leaves of Catharanthus roseus are of smaller number of epidermal hairs, one layer of epidermal cells and the leaf is dorsiventral. The vascular bundles of the two species are bicollateral.

Keywords: Leaf, morphology, anatomy, Nerium oleander, Catharanthus roseus.

INTRODUCTION

Plants serve as vast source for varied phyto-constituents exhibiting varied pharmacological property. Identifying such potential plants is of significance in medicine, so it becomes necessary to study the pharmacognostic characteristic of the plants before its use in the field of research and also in pharmaceutical formulation. Moreover it also helps in distinction from other allied species and adulterants. Morphological and anatomical characters of leaf are used as taxonomic markers to assist in the correct identification of the plant species.

The foliar epidermis is one of the most noteworthy taxonomic characters from the biosystematics point of view and the taxonomic studies of number of families are made on the basis of leaf epidermis (Baranova, 1972; Bhatia, 1984; Jones 1986). Leaf epidermal anatomical features such as stomata, trichomes and other characters are useful anatomical tools. According to WHO, the morphological (macroscopic) and anatomical (microscopic) descriptions of a medicinal plant is the first step towards establishing its identity and should be carried out. Microscopic evaluation is an indispensable tool for identification of medicinal plants and is one of the essential parameters in modern monograph.

N. oleander L. is an evergreen shrub or small tree in the family Apocynaceae, it grows to 6 m tall, the leaves are in pairs or whorls of three, thick, leathery, dark green narrow lanceolate 5-21cm long and 1-3.5cm broad and with an entire margin (Chaudhary and Kamal, 2014). Venation of the leaf is reticular. The adaxial and abaxial epidermises are covered by a thick layer of cuticle, anticlinal walls of cells of both the adaxial and abaxial epidermis are almost straight. The adaxial epidermis has scattered unicellular trichomes which are also present.
on the abaxial epidermises where they are located in deep crypts. The leaf is hypostomatic. The mesophyll is composed of two or three layers of dense palisade cells on both leaf surfaces, cells of the spongy parenchyma are located in the narrow central part of the leaf. The powder of the leaves is used as a snuff for treating epilepsy, (Chaudhary and kamal, 2014). The leaves of Catharanthus roseus (L.) G.Don. are oval to oblong 2.5-9cm long and 1-3.5cm broad, glossy green, hairless with a pale midrib and a short petiole about 1-1.8cm long and they are arranged in opposite pairs (Sain and Sharma, 2013).

In India, the juice of leaves is used as application to bee sting, wasp sting. In Africa the leaves of C. rosea are used for menorrhagia and rheumatism ( Sain and Sharma, 2013).

The objectives of this study is to identify the morphological and anatomical characteristics of the leaves of N. oleander L. and Catharanthus roseus (L.) G.Don. for the purpose of proper identification of medicinally important species, identify the leaf epidermal markers that can be utilized for the solution of existing taxonomic problems of plant species which overlap in most of their morphological characters, to evaluate the relationship of the critical taxa and to interpret the numerical data of biometric measurements of some leaf characters (such as leaf length, leaf width, leaf apex angle, leaf base angle).

MATERIALS AND METHODS
The plant materials
The plant materials for this study Nerium oleander and Catharanthus roseus family (Apocynaceae) were collected from Khartoum state (Central Sudan) in 2014, taxonomically identified at the Botany Department, Faculty of Science and Technology Omdurman Islamic University. Voucher specimens were deposited at the herbarium as reference Materials.

Morphological studies
Leaf shape, margin, apex, base, venation and color were examined.

Radar shape of leaf characters (leaf length, leaf width, leaf apex angle and leaf base angle) in studied plant species were carried out using Microsoft Excel (2010).

Anatomical Study
Epidermal Study

Fresh leaves of Nerium oleander and Catharanthus roseus were peeled with razor blade, the epidermal peels obtained were stained with alcoholic safranin for one minute and excess safranin stain was drained. The prepared epidermal peels were examined under the microscope and the type of stomata and epidermal hairs were determined Sass (1958).

Transverse Sections of the leaves

The method of double staining technique was adopted Johansen, (1940) segments from fresh mature leaves were fixed in formalin: glacial acetic acid: 70% alcohol (5:5:90) Sass,(1958). Transverse sections were prepared by using a rotatory microtome (Leitz 1512-West Germany) and
Stained with safranin and fast green stains. A light microscope was used to view the slides and adjusted to finest resolution. The sections were photographed using digital camera fitted with the microscope.

RESULTS AND DISCUSSION

Morphology

The morphological characters (shape, margin, apex, base, vention and color) are outlined and presented on table (1). The leaf margins, apex, venation and color of the two species are identical whereas the leaf shapes and bases are different.

<table>
<thead>
<tr>
<th>Characters</th>
<th>Nerium oleander</th>
<th>Catharanthus roseus</th>
</tr>
</thead>
<tbody>
<tr>
<td>shape</td>
<td>lanceolate-linear</td>
<td>ovate or oblong</td>
</tr>
<tr>
<td>margin</td>
<td>entire (smooth)</td>
<td>entire</td>
</tr>
<tr>
<td>apex</td>
<td>acute</td>
<td>acute</td>
</tr>
<tr>
<td>base</td>
<td>Cuneat</td>
<td>rounded</td>
</tr>
<tr>
<td>venation</td>
<td>pinnate</td>
<td>pinnate</td>
</tr>
<tr>
<td>color</td>
<td>green</td>
<td>green</td>
</tr>
</tbody>
</table>

Four characters; length, width, apex, base angle, leaf apex angle, venation and color of the leaves of *N. oleander* and *C. roseus* are shown on table (2). The radar shape showing that correlations between these characters of the two species is drawn (fig 1). The radar shape shows that the leaf characters of each species are identical and correlated.

Table 1: Leaf characters

<table>
<thead>
<tr>
<th>Character</th>
<th>Nerium oleander</th>
<th>Catharanthus roseus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leaf length (cm)</td>
<td>12</td>
<td>5</td>
</tr>
<tr>
<td>Leaf width (cm)</td>
<td>1.7</td>
<td>3.9</td>
</tr>
<tr>
<td>Leaf base angle</td>
<td>28</td>
<td>47</td>
</tr>
<tr>
<td>Leaf apex angle</td>
<td>15</td>
<td>40</td>
</tr>
<tr>
<td>Leaf length X Leaf width</td>
<td>20.4</td>
<td>19.5</td>
</tr>
<tr>
<td>Leaf width/Leaf Length</td>
<td>0.14</td>
<td>0.78</td>
</tr>
</tbody>
</table>

![Fig. 1. Radar shape of four leaf characters in *N. oleander* and *C. roseus*](image-url)
Anatomy
Epidermal Study

In the two studied species, Anisocytic stomata are found within the small polygonal epidermal cells. Large numbers of epidermal hairs are found. The number of epidermal hairs are found to be larger in *N. oleander*.

Transverse Sections

Transverse sections of *N. oleander* leaf is shown in Plate 1 (A and B). The transverse section of *Catharanthus roseus* leaf is shown in Plate 2. The upper epidermis is formed of 4 layers of small lignified parenchyma cells in *N. oleander* where as in *Catharanthus roseus* the upper epidermis is formed of one layer of small epidermal cells of larger sizes than those of *N. oleander*. There are many simple, non-glandular trichomes their numbers are larger in *N. oleander*. The leaf shows an isobilateral structure in *N. oleander*, the leaf is dorsiventral in *Catharanthus roseus*. In *N. oleander* The leaf consists of 2 layers of palisade cells, they are compact, long and filled with plastids. The spongy parenchyma is formed of loosely arranged parenchyma cells (with large intercellular spaces), there are some druses of calcium oxalate. *C. roseus* mesophyll is formed of one layer of long palisade cells and wide area of small spongy parenchyma cells. Lateral veins appeared in the region between the palisade and spongy parenchyma. They are formed of phloem and xylem vessels. In *N. oleander* the lower epidermis is formed of 3-4 layers of small lignified parenchyma covered with thick cuticle. There are many sunken stomata and large number of hairs. The number of hairs in the lower epidermis is larger than the upper and their cell sizes are smaller than the upper ones, there are three types of trichomes unicellular –simple hairs and non-glandular, cuticle is thinner than the upper, epidermis. In *C. roseus* the lower epidermis is formed of one layer of small lignified cells covered with thin cuticle no epidermal hairs appeared.

In the midrib region, the upper epidermis is followed by 7-8 layers of collenchyma and 8 layers of parenchyma. A well-developed crescent-shaped bicollateral vascular bundle is seen in the midrib region, outer and inner phloems are formed of sieve tube cells, companion cells and phloem parenchyma. Lignified xylem is formed of radial vessels the meta xylem in lower region, proto xylem in the upper region separated by xylem parenchyma. Crystals appeared in the midrib region, above the lower epidermis 4-5 layers of collenchyma followed by 10-11 layers of parenchyma, the cell sizes are larger in *C. roseus*. Branislava et al 2007 reported the same anatomical characters for *N. oleander*. *N. oleander* leaf showed larger number of epidermal hairs and four layers of epidermal cells, calcium oxalate crystals, sunken stomata and isobilateral leaves these characters are xeromorphic and indicate that this plant found in dry places compared to *C. roseus* which does not show these characters.
Plate 1. The leaf of *Nerium oleander* (A) Diagrammatic T.S in the midrib at the middle portion (40 X). (B) Detailed T.S in part of lamina at the middle portion (100X).
Plate 2. The leaf of *Catharanthus roseus*: Diagrammatic T.S in the midrib at the middle portion (100X).

REFERENCES


