Comparative Anatomical Studies on the leaves of family
Loranthaceae parasitic on trees in the Kingdom of Saudi Arabia

Rahmah Jrais

Leaf anatomical characteristic of six species *Plicosepalus acacia*, *Plicosepalus curviflorus*, *Phragmanthera austroarabica*, *Oncocalyx schimperi*, *Oncocalyx glabratus* and *Tapinanthus globiferus* of the family *Loranthaceae* were investigated in this study to ascertain the usefulness of these characters and determine the relationships among the investigated species. The anatomical features of the leaves showed that these species possess useful biosystematic characters that can be used to establish relationships among investigated species. An interesting aspect of this study is the type of leaves which showed the existence of two groups of plants, the first group included *Phragmanthera austroarabica* and *Tapinanthus globiferus* characterized by that the leaves in which the type Isobilateral. While the second group included species *Plicosepalus acacia*, *Plicosepalus curviflorus*, *Oncocalyx schimperi* and *Oncocalyx glabratus* that the leaves characterized by the kind of Isolateral. Also, The presence of crystals in the cells of the mesophyll of plant species studied the importance of possible discrimination by the species *Plicosepalus acacia* presence of crystals in groups, while two species *Phragmanthera austroarabica* and *Tapinanthus globiferus* characterized by single crystals, and absent of crystals in the species *Plicosepalus curviflorus*. The sunken stomata that characterized both species are discussed in relation to their biosystematic significance.

Keywords: Leaf, anatomy, characters, species, Loranthaceae.

INTRODUCTION

Parasitism by means of haustorial connections to a host is widespread in angiosperms, having arisen independently 10 or more times(Wilson and Calvin, 2006). The santalales contains five families (APG, 2003), three of which have aerial parasites known as mistletoes. The mistletoes are parasitic plant that derive all or most of their nutrition from other flowering plant. Although, many parasitic plants contain functional chlorophyll, they depend on their host plants for some of their carbon and other requirement. According to Takht and Zimmera(1996), the mistletoe plant belong to the kingdom Plantae, division Magnoliophyta, class Rosopsida, order Santales and family Loranthaceae.

In Saudi Arabia, Mistletoes uses are primarily for illness. Their general uses include diarrhoeal disease, cough,
arthritus and problems of urinogenital system. The leaves are used for treating skin diseases, fractured limbs and rheumatism. This research is based on the hypothesis that the variation in this leaf anatomy are significant and revealed that leaf anatomy possess many attributes of potential taxonomic importance that are diagnostic at the genus and species levels (Mbagwu and Edeoga, 2006; Nwachukwu and Mbagwu, 2007). Although, the usefulness of utilizing vegetative and anatomical features in the biosystematics consideration of various taxa have been reported by Edeoge and Eboka(2000) and Edeoge and Ikem(2001), there is no specific investigation conducted on the anatomical features of the leaves of family Loranthaceae. Hence this study reports the anatomical characters of the leaves of six species of loranthaceae. It assesses the relevance of and discusses the extent to which leaf anatomical features might be utilized in biosystematic consideration of these loranthaceae species.

MATERIALS AND METHODS

Fresh leaves from the six species of the Loranthaceae were collected from south western, west and north of Saudi Arabia start in 2009. The most healthy roots were collected and fixed in FAA (1:1:18) glacial acetic acid: 40% formaldehyde: 70% ethanol for 48-72 h. The root were washed several times distilled water then with two changes of 30% ethanol and dehydrated in the order 30, 50, 70, and 95% absolute alcohol. To infiltrate wax into the specimens, they were placed for 3 h in each of the following solutions containing a ratio of absolute alcohol to pure chloroform (3:1, 1:1, 1:3) and then pure chloroform. At the stage of pure chloroform, wax pellets at 60°C melting point were added and the wax changed with new ones at intervals. The specimens were left in the oven for 2-7 days to remove the chloroform. To embed in the wax, the contents the vials were transferred into moulds and the specimens kept in place with hot needles. As the wax solidified, it was transferred to a cold water bath for hardening and later stored for 2 days in an refrigerator.

For sectioning, a reichert rotary microtome was used and 7-10 mm thick sections were made. The ribbons were placed on clean slides smeared with a thin film of Haupts albumen, allowed to dry and drops of water added prior to mounting.

The slides were placed on a hot plate at 40°C for few minutes for the ribbons to expand and were stored overnight. The slides were immersed in pure xylene for 2-5 min in a solution of xylene and absolute alcohol with 1:1 ratio for few minutes. The slides were then transferred to another solution of xylene and alcohol in a ratio 1:3 for few minutes, to 95, 90, 70, and 50% alcohol. Drops of alcaine blue were added on the specimens, washed off with water and counter stained with safranin for 2 min, then dehydrated 50% alcohol, 70, 80, 90% xylene/alcohol solution and mounted in canda balsam. The slides were dried on a hot plate at 30°C. Then photomicrographs of the specimens were taken from the permanent slides (Figure 1) using light microscope.

RESULTS

Table (1-2) summarize the quantitative and qualitative data of leaf anatomy. Where we can compare and division of plants to groups based on many characters such as epidermis, mesophyll, main vascular bundle, lateral vascular bundle, crystals, ducts, Schlechny cell in mesophyll and supporting tissue around vascular bundles.

DISCUSSION

For leaves has been marked by two species Tapinanthus globiferus (Figure 1-D) and Phragmanthera austroarabica (Figure 1-F) that cuticle Low thick layer where the average thickness ranging from (µm 0.03 - µm 0.07), while the species Plicosepalus acacia, Oncocalyx schimperi (Figure 1-C) Oncocalyx schimperi (Figure 1-B) and Oncocalyx glabratus (Figure 1-E) the average thick cuticle µm0.12. Plicosepalus curtiflorus (Figure 1-A) the average high-thick cuticle µm0.22. The main characteristics of the important introduction the value of taxonomic to distinguish between plant species, type of leaves which showed the existence of two groups of plants, the group included the first two species Phragmanthera austroarabica and Tapinanthus globiferus characterized by that the leaves in which the type Isobilateral while the second group included species Plicosepalus acacia, Plicosepalus curtiflorus, Oncocalyx schimperi and Oncocalyx glabratus that the leaves characterized by the kind of Isolateral, where Mesophyll is non differentiated. The present study has shown that the average length of mesophyll through the middle of possible discrimination of plant species into two groups, the first group included species Phragmanthera austroarabica, Plicosepalus curtiflorus and Plicosepalus acacia which varies the length of mesophyll between the middle (µm 6.21- µm 6.9) and the second group included species Oncocalyx schimperi, Oncocalyx glabratus and Tapinanthus globiferus ranging between average length of mesophyll (µm 3.93 - µm 3.21). Also varied anatomical structure of the mesophyll where the uniqueness of Plicosepalus curtiflorus the presence of air spaces in high density in the tissue average, while the rest of the species is a dense air spaces. The presence of crystals in the cells of the mesophyll of plant species studied the importance of possible discrimination by the species Plicosepalus acacia presence of crystals in groups, while two species Phragmanthera austroarabica and Tapinanthus globiferus.
Table 1. Leaf anatomical study of six species of family Loranthaceae in Kingdom of Saudi Arabia.

<table>
<thead>
<tr>
<th>Species</th>
<th>Cuticle shape</th>
<th>Cuticle thickness µm</th>
<th>Isobilateral</th>
<th>Elongate-well</th>
<th>Elongate-embedded well</th>
<th>Spongy</th>
<th>Mesophyll length µm</th>
<th>Number</th>
<th>Size Length x width µm</th>
<th>Bundle sheath</th>
<th>Lateral vascular bundle</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Plicosepalus acacia</em></td>
<td>Corrugated</td>
<td>0.12</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>6.12</td>
<td>1</td>
<td>x 4.05 4.26</td>
<td>-</td>
<td>2.46-2.16</td>
</tr>
<tr>
<td><em>Plicosepalus curviflorus</em></td>
<td>Corrugated</td>
<td>0.22</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>6.9</td>
<td>1</td>
<td>x 2.45 2.79</td>
<td>-</td>
<td>1.41 x 1.38</td>
</tr>
<tr>
<td><em>Phragmanthera austroarabica</em></td>
<td>Straight</td>
<td>0.07</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>6.21</td>
<td>1</td>
<td>2.77 x 2.65</td>
<td>+</td>
<td>1.79 x 1.56</td>
</tr>
<tr>
<td><em>Oncocalyx schimperi</em></td>
<td>Corrugated</td>
<td>0.12</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>3.21</td>
<td>7</td>
<td>2.31 x 1.56</td>
<td>-</td>
<td>0.86 x 0.54</td>
</tr>
<tr>
<td><em>Oncocalyx glabratus</em></td>
<td>Corrugated</td>
<td>0.12</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>3.93</td>
<td>3</td>
<td>1.74 x 2.19</td>
<td>-</td>
<td>0.88 x 0.74</td>
</tr>
<tr>
<td><em>Tapinanthus globiferus</em></td>
<td>Straight</td>
<td>0.03</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>3.40</td>
<td>1</td>
<td>x 2.35 2.89</td>
<td>-</td>
<td>0.65 x 0.59</td>
</tr>
</tbody>
</table>

Table 2. Leaf anatomical study of six species of family Loranthaceae in Kingdom of Saudi Arabia.

<table>
<thead>
<tr>
<th>Species</th>
<th>Crystals</th>
<th>Ducts</th>
<th>Schlemchyma cell in mesophyll</th>
<th>Supporting tissue</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>In group</td>
<td>Solitary</td>
<td>Present</td>
<td>Absent</td>
</tr>
<tr>
<td><em>Plicosepalus acacia</em></td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><em>Plicosepalus curviflorus</em></td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><em>Phragmanthera austroarabica</em></td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Oncocalyx schimperi</em></td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><em>Oncocalyx glabratus</em></td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><em>Tapinanthus globiferus</em></td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>
A = Plicosepalus curviflorus

B = Oncocalyx schimperi
C = Plicosepalus acacia

D = Tapinanthus globiferus
characterized by single crystals, and absent of crystals in the species *Plicosepalus curviflorus*.

For the cells in the tissue Schlemchyma cell average recorded its presence in the current study *Plicosepalus acacia*, *Oncocalyx schimperi* and *Oncocalyx glabratus* while there are not present in *Tapinanthus globiferus*, *Plicosepalus curviflorus* and *Phragmanthera australis*. According to the supporting tissue of vascular bundle we can divided of plant species into three groups, the first group included species *Plicosepalus curviflorus* and *Plicosepalus acacia*, which Cholenchyma tissue rounded of vascular, the second group included species *Oncocalyx schimperi* and *Phragmanthera australis* which Cholenchyma tissue in base of vascular and the three
group included species *Oncocalyx glabratus* and *Tapinanthus globiferus* which Cholenchyma tissue in upper and lower of vascular. The difference in these species are enough to differentiation between the similarities in morphological characters of leaf in the same family *loranthaceae*. The overall findings support the principles, relationships and generalizations of other scientists that leaf anatomical features are useful tools in systematic botany.

**Artificial key according to leaf anatomical characters:**

I isobilateral leaf
- Supporting collenchyma tissue at the base of vascular bundles
  — *Phragmanthera austroarabica*.
- Supporting collenchyma tissue at the top and bottom of vascular bundles
  — *Tapinanthus globiferus*.

II isolateral leaf
- One main vascular bundle.
  - Crystals presence in the leaf mesophyll
    — *Plicosepalus acacia*.
  - Crystals presence in the leaf mesophyll
    — *Plicosepalus curviflorus*.
- More than one main vascular bundle.
  - Supporting collenchyma tissue at the base of vascular bundles
    — *Oncocalyx schimperi*.
  - Supporting collenchyma tissue at the top and bottom of vascular bundles
    — *Oncocalyx glabratus*.

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**REFERENCES**


