Morphological studies on legume and hilum in seven species of *Vigna Savi* (Fabaceae)

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Received 26 September, 2020; Accepted 19 November, 2020

In this study of different taxa of genus *Vigna* the macro and micromorphological characters were investigated. These taxa collected from different countries (3 cultivated and 8 annual or perennial herbs) representing 7 species of this genus, namely: *Vigna radiate*, *Vigna trilobata*, *Vigna vexillata*, *Vigna caracalla*, *Vigna pubescens*, *Vigna unguiculata*, and *Vigna luteola*. Legume features such as shape, colour, size, surface texture, number of locules, number of legumes per peduncle seed set percentage, orientation. Moreover, by using scanning electron microscopy, additional details were obtained for stomatal shape, size, type, cuticular and wax ornamentation of the surface of the mature pod, including details of the seed such as hilum shape, size, position, ornamentation based on scanning electron microscope evidence. The usefulness of the macro and micromorphological features as criteria for taxonomic identification was emphasized.

Key words: *Vigna*, Fabaceae, morphological characters, pod, hilum, S.E.M., taxonomy.

INTRODUCTION


Economically *Vigna* is a source of plant protein for human food and animal feed in tropical, sub-tropical, arid and semi-arid regions, and also plays an important role in soil fertility by fixing nitrogen (Mbagwu and Endeoga, 2006; Pule-Meulenberg et al., 2010; Sprent et al., 2010; El-Ghamery et al., 2012; El-Gazzar et al., 2013; Popoola et al., 2015). Barthlott and Ehler (1977) maintained that the epidermal features are variable for angiosperm taxa and can be used to evaluate possible relationships. Seed morphological characters are useful in the analysis of of taxonomic distinguish inter and intra relationships in a wide variety of plant families (Esau, 1953; Shetler and Morin, 1986; Takhtajan, 1991; Buss et al., 2001; Zhang et al., 2005; Gontcharova et al., 2009). In addition to
general characters of morphology of seeds, sculpturing details of outer seed coat are quite variable between different species and can be of a systematic importance (Chowdhury and Buth, 1970; Gohary and Mohammed, 2007). The importance of ultrastructural pattern analysis of the seed coat observed under the SEM has been well recognized as a reliable approach for assessing phenetic relationship and identification of species or taxa (Barthlott, 1981; Tobe et al., 1987; Koul et al., 2000; Yoshizaki, 2003; Javadi and Ya).

The micro-morphological characters of the seed and legume surface can be useful for delimiting taxa at various levels (Karcz et al., 2005; Akçin, 2008; Khafagi et al., 2018). Legume colour that is less affected by environmental variations has been used as a marker for the identification of species or varieties (Sangwan and Lodhi, 1998). In Vigna species, many taxonomists have used seed and hilum morphology and micromorphology to differentiate the species (Chandel et al., 1991; Nath and Dasgupta, 2015; Umdale et al., 2017). This work aims to describe the significance of macro and micromorphological characters of legume and hilum using SEM in 11 taxa of the genus Vigna and its implication in the classification of this plant.

### MATERIALS AND METHODS

Seeds of Vigna Savi accessions were obtained from the International Livestock Center for Africa (ILCA), Jazan Region of Saudi Arabia and from different localities in Egypt. Eleven taxa representing 11 species belonging to genus Vigna Savi were investigated. The sources and origins of these taxa are given in Table 1. Seeds were germinated in April 2019, at the Botanic Gardens of the Botany Department, Faculty of Science, New Valley University, Egypt. Flowering began in June 2019. Samples of complete mature plants were collected, including fruits and seeds. Authentic specimens of all taxa were pressed, dried and deposited as herbarium specimens at the Botany Department, Faculty of Science, New Valley University. The morphological characters were examined, which included legume shape, colour, size, texture, apex, shattering, seed per pod, locule per pod, seed set percentages, pod attachment, number of pods per peduncle, and stomata on the surface of a mature pod. Hilum shape, size, colour and position features of different species were also noted. The seed set percentage was calculated using the formula described by Popoola et al. (2015) as follows:

$$\text{Seed set \%} = \frac{\text{No.of seed / pod}}{\text{No.of locule / pod}} \times 100$$

For preparation of legume and seeds of each taxon to scan the surface by using the scanning electron microscope (SEM), legumes and seeds from each taxon were divided into two groups. The first group was mounted on an adhesive surface and prepared for SEM investigation. The second group was washed thoroughly using distilled water to remove any impurities on the seed surfaces; then they were dried and soaked in 10% HCl for 6 h to remove the coat enveloping the seed (Ismail and El-Ghazaly, 1990). This was followed by washing the seeds with distilled water and then dried and prepared for SEM examination by mounting these dry legumes and seeds onto clean stubs using double-sided adhesive tape. These clean dry legumes and seeds were gold coated using a JEOL JFC 1100E ion-sputtering device. Then, the coated seeds were viewed and photographed with a JOEL ISM-550LV scanning electron microscope, operated at accelerating voltage of 15 KV at the Scanning Electron Microscopy Unit in the Regional Center for Mycology and Biotechnology, Al-Azhar University. Magnifications of images were denoted with scale bars either in mm or µm as appropriate. Measurements (L x B) of legumes and seeds were taken using a measuring scale. Seeds were uniformly scanned at the hilum, the surface surrounding both sides of the hilum, and the surface pattern was highlighted to observe the cellular and intercellular patterns. The terminology for describing seed coat patterns follows Barthlott (1981, 1990) and Stearn (1996).

### RESULTS

#### Legume

The legume morphological characters of the studied taxa are given in Table 2, represented in Plate 1 (1a, 2a, 3a,
Table 2. Pod morphometric characteristic of the studied taxa.

<table>
<thead>
<tr>
<th>N</th>
<th>Species</th>
<th>Shape</th>
<th>Colour</th>
<th>Size (Length cm)</th>
<th>Width (cm)</th>
<th>Exocarp hairiness</th>
<th>Pod apex</th>
<th>Pod shattering</th>
<th>Seed / Pod</th>
<th>Locules / Pod</th>
<th>Seed set %</th>
<th>Pod orientation</th>
<th>No. Pod / Peduncle</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>V. radiata</td>
<td>Linear - terete</td>
<td>Black</td>
<td>4-5</td>
<td>1.4-1.6</td>
<td>Pubescent</td>
<td>Straight beak</td>
<td>Present</td>
<td>5-6</td>
<td>6-8</td>
<td>75-83</td>
<td>Erect</td>
<td>4-6</td>
</tr>
<tr>
<td>2</td>
<td>V. radiata</td>
<td>Linear - terete</td>
<td>Brown</td>
<td>6-8</td>
<td>1.4-1.5</td>
<td>Scabrous</td>
<td>Straight beak</td>
<td>Absent</td>
<td>8-10</td>
<td>9-12</td>
<td>63-89</td>
<td>Pendent</td>
<td>5-8</td>
</tr>
<tr>
<td>3</td>
<td>V. trifoliate</td>
<td>Linear - terete</td>
<td>Black</td>
<td>2.5-3</td>
<td>0.8-0.9</td>
<td>Glabrous</td>
<td>Straight beak</td>
<td>Present</td>
<td>5-6</td>
<td>6-8</td>
<td>75-83</td>
<td>Erect</td>
<td>6-8</td>
</tr>
<tr>
<td>4</td>
<td>V. vexillata</td>
<td>Linear - terete</td>
<td>Brownish black</td>
<td>11-12</td>
<td>1-1.1</td>
<td>Puberulous</td>
<td>Declined beak</td>
<td>Present</td>
<td>10-12</td>
<td>11-14</td>
<td>86-91</td>
<td>Erect</td>
<td>4-6</td>
</tr>
<tr>
<td>5</td>
<td>V. caracalla</td>
<td>Linear - flat</td>
<td>Gray</td>
<td>7-8</td>
<td>1.9-2</td>
<td>Glabrous</td>
<td>Hooked</td>
<td>Absent</td>
<td>8-10</td>
<td>9-13</td>
<td>76-89</td>
<td>Pendent</td>
<td>5-7</td>
</tr>
<tr>
<td>6</td>
<td>V. pubescens</td>
<td>Linear - terete</td>
<td>Black</td>
<td>7-8</td>
<td>0.9-1</td>
<td>Pubescent</td>
<td>Declined beak</td>
<td>Present</td>
<td>7-9</td>
<td>8-11</td>
<td>82-88</td>
<td>Erect</td>
<td>4-6</td>
</tr>
<tr>
<td>7</td>
<td>V. pubescens</td>
<td>Linear - terete</td>
<td>Creamy</td>
<td>6-7</td>
<td>1-1.1</td>
<td>Pubescent</td>
<td>Declined beak</td>
<td>Present</td>
<td>6-7</td>
<td>8-10</td>
<td>70-75</td>
<td>Pendent</td>
<td>4-6</td>
</tr>
<tr>
<td>8</td>
<td>V. unguiculata</td>
<td>Curved - terete</td>
<td>Creamy</td>
<td>17-23</td>
<td>2.4-2.6</td>
<td>Glabrous</td>
<td>Declined beak</td>
<td>Absent</td>
<td>20-25</td>
<td>22-28</td>
<td>89-91</td>
<td>Pendent</td>
<td>4-6</td>
</tr>
<tr>
<td>9</td>
<td>V. unguiculata</td>
<td>Curved - terete</td>
<td>Creamy</td>
<td>12-15</td>
<td>1.5-1.8</td>
<td>Glabrous</td>
<td>Declined beak</td>
<td>Absent</td>
<td>7-12</td>
<td>9-15</td>
<td>83-87</td>
<td>Pendent</td>
<td>4-6</td>
</tr>
<tr>
<td>10</td>
<td>V. unguiculata</td>
<td>Curved - terete</td>
<td>Creamy</td>
<td>16-17</td>
<td>2.2-2.5</td>
<td>Glabrous</td>
<td>Declined beak</td>
<td>Absent</td>
<td>15-18</td>
<td>16-20</td>
<td>93-94</td>
<td>Pendent</td>
<td>5-7</td>
</tr>
<tr>
<td>11</td>
<td>V. luteola</td>
<td>Linear - terete</td>
<td>Brown</td>
<td>4-5</td>
<td>1.3-1.5</td>
<td>Glabrous</td>
<td>Declined beak</td>
<td>Present</td>
<td>7-8</td>
<td>9-11</td>
<td>73-78</td>
<td>Erect</td>
<td>8-9</td>
</tr>
</tbody>
</table>

4a and 5a) and Plate 2 (6a, 7a, 8a, 9a, 10a and 11a).

Legume shattering, shape and apex

The results in Table 2 show all studied Vigna species have a shattering (dehiscent) legume, except Vigna unguiculata, Vigna caracalla and Vigna radiata (No.2) collected from Egypt. The time of legume shattering is related to the maturity of the legumes; also all studies species were of the linear-terete legume shape, except V. unguiculata that is slightly curved and V. caracalla that is linearly flat. The apex of legume is straight beak in the subgenus Ceratotropis. There is declined beak in the subgenus Plectrotropis and subgenus Vigna and a hooked beak in the subgenus Sigmidotropis (Table 2, Plate 1 (1a, 2a, 3a, 4a, 5a) and Plate 2 (6a, 7a, 8a, 9a, 10a and 11a).

Colour

Legume colour at maturity varied from black, brown, brownish black, gray to creamy. It was black in V. radiata, Vigna trifoliate and Vigna pubescens collected from Ethiopia, while brown in V. radiata collected from Egypt and Vigna luteola collected from Ethiopia, brownish black in Vigna vexillata, grey in V. caracalla and creamy in Vigna pubescens collected from Saudi Arabia and V. unguiculata (Table 2 and Plate 1 (1a, 2a, 3a, 4a, 5a) and Plate 2 (6a, 7a, 8a, 9a, 10a and 11a).

Surface texture

Surface texture of V. pubescens and the cultivated of V. radiata was pubescent, while in the V. radiata wild scabrous, glabrous in V. trifoliate and V. caracalla, V. unguiculata and V. luteola (Table 2, Plate 1 (1a, 2a, 3a, 4a, 5a) and Plate 2 (6a, 7a, 8a, 9a, 10a and 11a).

Size

Three categories of legume size are recognized the smallest are those of wild species V. trifoliate, V. radiata and V. luteola; while the legume of V. radiata cultivated, V. pubescens and V. caracalla are of medium size. The remaining species have long legume (Table 2, Plate 1 (1a, 2a, 3a, 4a, 5a) and Plate 2 (6a, 7a, 8a, 9a, 10a and 11a).

Number of seeds per legume

The number of seed per legume was recognized into two categories: the first with less than 10 seeds (V. radiata, V. trifoliate, V. caracalla, V. pubescens and V. luteola) and the second with more than 10 seeds (Table 2).

Number of locules per legume

The number of locules per legume 6-8 in V. radiata and V. trifoliate, 9-11, 9-12, 9-13 in V. luteola, V. radiata were collected from Egypt and V. caracalla respectively, while 8-10, 8-11 in V. pubescens, 22-28, 9-15, 16-20 in V. unguiculata respectively (Table 2).
Table 3. Exocarp features (stomata characters and Cuticular ornamentation) of the pod in the studied taxa.

<table>
<thead>
<tr>
<th>Taxa</th>
<th>Level</th>
<th>Shape</th>
<th>Size L (µm)</th>
<th>Size W (µm)</th>
<th>Rim</th>
<th>Peristomatal rim</th>
<th>Cuticular ornamentation</th>
</tr>
</thead>
<tbody>
<tr>
<td>V. radiata</td>
<td>Semidepressed</td>
<td>Elliptical</td>
<td>18-19</td>
<td>8-10</td>
<td>Raised</td>
<td>Present</td>
<td>Favularite</td>
</tr>
<tr>
<td>V. radiata</td>
<td>Semidepressed</td>
<td>Elliptical</td>
<td>15-18</td>
<td>7-9</td>
<td>Raised</td>
<td>Present</td>
<td>Favularite</td>
</tr>
<tr>
<td>V. trilobata</td>
<td>Semidepressed</td>
<td>Elliptical</td>
<td>21-23</td>
<td>9-12</td>
<td>Raised</td>
<td>Present</td>
<td>Favularite</td>
</tr>
<tr>
<td>V. vexillata</td>
<td>Superficial</td>
<td>Broad-Elliptical</td>
<td>30-33</td>
<td>17-20</td>
<td>Raised</td>
<td>Present</td>
<td>Ruminate</td>
</tr>
<tr>
<td>V. caracalla</td>
<td>Semidepressed</td>
<td>Narrow-Elliptical</td>
<td>16-18</td>
<td>4-6</td>
<td>Raised</td>
<td>Present</td>
<td>Rugose</td>
</tr>
<tr>
<td>V. pubescens</td>
<td>At level</td>
<td>Elliptical</td>
<td>18-20</td>
<td>8-10</td>
<td>Raised</td>
<td>Present</td>
<td>Rugose</td>
</tr>
<tr>
<td>V. pubescens</td>
<td>At level</td>
<td>Elliptical</td>
<td>18-20</td>
<td>8-10</td>
<td>Raised</td>
<td>Present</td>
<td>Reticulate</td>
</tr>
<tr>
<td>V. unguiculata</td>
<td>At level</td>
<td>Elliptical</td>
<td>23-26</td>
<td>10-14</td>
<td>Raised</td>
<td>Present</td>
<td>Reticulate</td>
</tr>
<tr>
<td>V. unguiculata</td>
<td>At level</td>
<td>Elliptical</td>
<td>20-23</td>
<td>8-12</td>
<td>Raised</td>
<td>Present</td>
<td>Reticulate</td>
</tr>
<tr>
<td>V. unguiculata</td>
<td>At level</td>
<td>Elliptical</td>
<td>20-24</td>
<td>8-12</td>
<td>Raised</td>
<td>Present</td>
<td>Reticulate</td>
</tr>
<tr>
<td>V. luteola</td>
<td>At level</td>
<td>Elliptical</td>
<td>10-14</td>
<td>5-7</td>
<td>Raised</td>
<td>Present</td>
<td>Reticulate</td>
</tr>
</tbody>
</table>

Orientation

Two types of pod orientation were observed as erect and pendent. In V. radiata, collected from Egypt, and in V. unguiculata and V. caracalla the legume is pendent; while the legume is erect in V. radiata, V. trilobata, V. vexillata and V. luteola.

Seed set percentage

Seed set percentage was reported as two categories: the first less than 75% in wild V. pubescens while the second more than 75% in the remainder of the studied species (Table 2).

Stomata on the surface of mature legume

Most of the studied taxa possess barely sunken elliptical stomata with raised stomatal rims and long narrow aperture; the stomatal leveling ranged between superficial, at a level and semi-depressed. It is superficial only in V. vexillata, semidepressed in V. radiata V. trilobata and V. caracalla and at the level in the remainder of the studied taxa.

The shape

Shape of stomata is broadly to narrowly elliptical in V. vexillata and V. caracalla (Table 3; Plate 1, (4b and 5b); while elliptical shaped in the remainder of the studied taxa (Table 3; Plate 1 (1b, 2b, 3b) and Plate 2 (6b, 7b, 8b, 9b, 10b and 11b).

The size

There is an inter and intra-specific variation in the stomatal size. Stomata ranged in size from 10-33 × 4-20 µm (length × width). However, the largest stomata are those of V. vexillata 30-33 µm long, and 17-20 µm wide. The smallest are those of V. luteola 10-14 µm long, 5-7 µm wide (Table 3).

The rim

Rims are usually broad and raised in all taxa. Most of the studied taxa possess stomata with one stomatal rim and peristomatal rim.

Cuticular and wax ornamentation

The reticulate cuticular ornamentation was recorded for V. luteola, section Vigna and V. unguiculata, V. pubescence collected from Saudia Arabia section Catiang while it is rugose in V. pubescens collected from Ethiopia. It is favularite in V. radiata and V. trilobata section Ceratotrops, while rugose in V. caracalla section Segmidotrops and ruminate in V. vexillata section Plectrotrops (Plate 1 (1b, 2b, 3b, 4b, 5b) and Plate 2 (6b, 7b, 8b, 9b, 10b and 11b; and Table 3).

Hilum

Position

The hilum in the examined seeds was located in central position as shown in V. radiata, V. trilobata and V. caracalla Plates 3 (Figures 1, 2, 3, 4 and 5); whereas, the hilum was subcentral in the remainder of the studied taxa (Table 4; Figures 6, 7, 8, 9, 10 and 11 (Plates 4)). The hilum shape was oblong in V. radiata, elliptical in V. vexillata, V. caracalla and V. luteola, ovate in the other taxa; and the level is sunken in V. caracalla while raised in the remaining taxa.
The size

The size of hilum varied in all studied species and their accessions, where the size ranged from 2.0-3.0 mm × 1.0-1.4 mm to 0.7-1.0 × 0.5-0.7 mm (Length × Width). The longest size (2.0-3.0 mm) was found in *V. unguiculata* from Egypt (No. 8), (Figure 8), followed by 1.8-2.0 mm in *V. unguiculata* from Ethiopia (No. 10), (Figure 10); the lowest one was found in *V. trilobata*, 0.7-1.0 mm long, 0.5-0.7 mm wide (Figure 3).

The data in Table 4 show that there is a high difference in the size of the hilum between the taxa of the same species of the studied genus as in *V. unguiculata* (2.0-3.0 mm length, 1.6-1.7 mm length and 1.8-2 mm length), *V. radiata* (1.4-1.6 mm length and 1.2-1.4 mm length) and *V. pubescens* (1.1-1.3 mm length and 1.3-1.5 mm length); *V. luteola* (1.3-1.5 mm length) *V. vexillata* (1.2-1.6 mm length), and *V. caracalla* (1.4-1.6 mm length).
The rim aril and colours

The term aril is used for a fleshy to hard structure that develops from the funiculus or ovule after fertilization and invests part or all of a seed. Rim aril was absent in *V. radiata* and *V. trilobata*; whereas, it is well developed with superficial expansion and a cleft margin in *V. vexillata* and *V. caracalla*. However, in the remainder of the taxa, the aril is well developed with superficial expansion without cleft margin. Diverse rim colours were observed varying from black, brown to yellowish brown (Figures 1a, 2a, 3a, 4a, 5a, 6a, 7a, 8a, 9a, 10a, 11a and 11a and Table 4).

Ornamentation

The examinations of hilum surface by S.E.M showed different types of ornamentations were present in cultivated *V. radiata*. Among the ornamentation, there was rugose structure type, wax flakes depositions; and heavy globular waxy depositions of various size were also clearly seen.

The hilum in wild *V. radiata* and *V. trilobata* was reticulate with some waxy flakes. In *V. vexillata* it is rugulose striate, while in *V. caracalla*, the cells of hilum are disposed in a uniserate compact filament and are fusiformed with the rim. However, in *V. pubescens* the hilum is reticulate colliculate. In *V. unguiculata* there is regulate pattern, and it contains a furrow-like array of longitudinal cells with light waxy deposition. The hilum of *V. luteola* is reticulate foveate (Figures 1b, 2b, 3b, 4b, 5b, 6b, 7b, 8b, 9b, 10b and 11b; Table 4).

**DISCUSSION**

In this study, our observations of the macro and micromorphological features revealed the presence of considerable morphological variations between the different subgenus species. In subgenus *Ceratotropis*, the pods are pendent, non-shattered and brown in colour in cultivated *V. radiata*. Whereas, it is erect, shattered and black in colour in wild *V. radiata* and *V. trilobata* at maturity. In subgenus *Plectrotropis*, the pod is erect, shattered and brownish black in colour. In subgenus *Sigmoidotropis*, the pod is pendent and non-shattered; and grey in colour. In the subgenus *Vigna*, the pod is pendent and shattered or non-shattered/erect and shattered. In this respect these variations were previously observed by many authors such as Garba and pasqet (1998), Sangwan and Lodhi (1998), Peksen and Peksen (2013), and Popoola et al. (2015, 2017). Many accessions within *V. unguiculata* show long and relatively large seeds with seed coat colour or patterns similar to the cultivated cowpea; although, some of their character's pod structure, pod position on the raceme and pod shattering are characteristics of the wild species. Also, diversity of *V. vexillata* is also well represented on the variability of the surface pubescence (that is, plant almost glabrescent, densely pubescent to bristly) as reported by Padulosi and Ng (1993).

The number of seeds per pod of the cultivated *V. unguiculata* (7-12 seeds) was relatively small compared with the cultivated (No. 8), (20-25 seeds) followed by the wild form of this species.

### Table 4. Hilum characters and cuticular ornamentation of the studied taxa.

<table>
<thead>
<tr>
<th>Species</th>
<th>shape</th>
<th>level</th>
<th>Position</th>
<th>Size</th>
<th>Rim aril</th>
<th>Rim colour</th>
<th>ornamentation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Length (mm)</td>
<td>Width (mm)</td>
<td></td>
</tr>
<tr>
<td><em>V. radiata</em></td>
<td>Oblong</td>
<td>Raised</td>
<td>Central</td>
<td>1.4-1.6</td>
<td>0.5-0.6</td>
<td>Absent</td>
<td>Yellowish brown</td>
</tr>
<tr>
<td><em>V. radiata</em></td>
<td>Oblong</td>
<td>Raised</td>
<td>Central</td>
<td>1.2-1.4</td>
<td>0.4-0.5</td>
<td>Absent</td>
<td>Black</td>
</tr>
<tr>
<td><em>V. trilobata</em></td>
<td>Oval</td>
<td>Raised</td>
<td>Central</td>
<td>0.7-1.0</td>
<td>0.5-0.7</td>
<td>Absent</td>
<td>Brown</td>
</tr>
<tr>
<td><em>V. vexillata</em></td>
<td>Elliptical</td>
<td>Raised</td>
<td>Sub central</td>
<td>1.2-1.6</td>
<td>0.5-0.7</td>
<td>Present with superficial expansion and cleft margin</td>
<td>Black</td>
</tr>
<tr>
<td><em>V. caracalla</em></td>
<td>Elliptical</td>
<td>Sunken</td>
<td>Central</td>
<td>1.4-1.6</td>
<td>0.6-0.7</td>
<td>Present with superficial expansion and cleft margin</td>
<td>Black</td>
</tr>
<tr>
<td><em>V. pubescens</em></td>
<td>Oval</td>
<td>Raised</td>
<td>Sub Central</td>
<td>1.1-1.3</td>
<td>0.4-0.5</td>
<td>Present with superficial expansion</td>
<td>Black</td>
</tr>
<tr>
<td><em>V. unguiculata</em></td>
<td>Oval</td>
<td>Raised</td>
<td>Sub Central</td>
<td>1.3-1.5</td>
<td>0.6-0.7</td>
<td>Present with superficial expansion</td>
<td>Brown</td>
</tr>
<tr>
<td><em>V. unguiculata</em></td>
<td>Oval</td>
<td>Raised</td>
<td>Sub Central</td>
<td>2.0-3.0</td>
<td>1.0-1.4</td>
<td>Present with superficial expansion</td>
<td>Brown</td>
</tr>
<tr>
<td><em>V. unguiculata</em></td>
<td>Oval</td>
<td>Raised</td>
<td>Sub Central</td>
<td>1.8-2.0</td>
<td>0.7-0.8</td>
<td>Present with superficial expansion</td>
<td>Brown</td>
</tr>
<tr>
<td><em>V. luteola</em></td>
<td>Elliptical</td>
<td>Raised</td>
<td>Sub Central</td>
<td>1.3-1.5</td>
<td>0.5-0.6</td>
<td>Present with superficial expansion</td>
<td>Brown</td>
</tr>
</tbody>
</table>
Plate 2. Photograph and S.E.M micrographs of pod. (6a, b) *V. pubescens* Ethiopia; (7a, b) *V. pubescens*; (8a, b) *V. unguiculata*; (9a, b) *V. unguiculata*; (10a, b) *V. unguiculata*, wild, (11a, b) *V. luteola*.

(15-18 seeds) from subgenus *Vigna*; and the other studied species which had a range of 6-10 seeds. However, the seeds per pod of the two accessions of *V. radiata* and *trilobata* (5-6 seeds) from subgenus *Ceratotropis* were relatively small compared with the other studied species of *Vigna*. A similar variation in the pod size for different taxa of *Vigna* was reported by many investigators, and it could be attributed to the geographical distribution of these taxa (Barrett, 1990; Hymowitz, 1990; Pasquet, 1998; Pasquet and Vanderborght, 2000; Fery, 2002; Bisht et al., 2005). The usage of the scanning electron microscope (SEM) studies in examination of legume surface of 7 species of *Vigna* (11 accessions) revealed the importance of this technique as a taxonomic tool. The results in this investigation showed fairly heterogeneous stomatal surface patterns of the legume in the different species of *Vigna*; and also this data offer significant information to be used in classification similar to that studied by El-Hadidy (2004) for the genus *Lotus*, family Fabaceae.

The seed of *V. radiata* and *V. trilobata* section *Ceratotropis* has no rim-aril, while the other species...
studied have a superficial expansion of the rim aril. In comparison, *V. vexillata* section Plectrotropis and *V. caracalla* section Sigmidotropis have a superficial expansion with cleft margin, which is comparable to the dry tongue aril illustrated by Gunn (1981) for some Papilionoideae taxa. These observations are also in

Plate 3. (Figures 1-5). Photograph of seed and S.E.M micrographs of Hilum. (1–a, b) *V. radiata* wild; (2–a, b) *V. radiata* cultivated; (3–a, b) *V. trilobata*; (4–a, b) *V. vexillata*; (5–a, b) *V. caracalla*. 
agreement with the studies of Fabiana et al. (2013) and Khedia et al. (2017), on some species of *Vigna*.
have demonstrated the existence of diversity in various taxa; and revealed that the mature seed surface pattern is not identical; rather it showed the presence of a remarkable difference between the two collected accessions of each species such as in wild and cultivated forms collected from different countries. In contrast, the spermoderms of both collected taxa of *V. pubescens* are reticulate colliculate, regulate in *V. vexillata* and *V. unguiculata*, compact in *V. caracalla*, rugae in *V. radiata* wild while reticulate in *V. radiata* cultivated, and reticulate foveate in *V. luteola*; as also was illustrated by Kumar and Rangaswamy (1984), Fabiana et al. (2013) and Khedia et al. (2017).

**Conclusion**

The macro and micro-morphological characteristics of legume and hilum were reported. This includes legume features such as shape, colour, size, surface texture, number of locules, number of legumes per peduncle, seed set percentage, and orientation. Moreover, by using scanning electron microscopy, additional detailed information was obtained such as stomatal shape, size, type, cuticular and wax ornamentation of the mature pod surface. In addition, hilum shape, size, position, and ornamentation were elucidated by using scanning electron microscopy. The results provide clear and important attributes of these species for the definition and identification of the taxa collected from different countries.

**CONFLICT OF INTERESTS**

The authors have not declared any conflict of interests.

**ACKNOWLEDGEMENT**

The authors express their gratitude to Prof. Dr. Abbas, A. El-Ghamery, Botany Department, Faculty of Science, Al-Azhar University for revision and valuable discussions in this manuscript.

**REFERENCES**


