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# PHENOTYPE VARIATIONS AMONG Moringa oleifera ACCESSIONS IN MALAYSIA

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**ABSTRACT** Moringa oleifera is a multipurpose tree as it is cultivated for vegetable, spice, cosmestic oil and medicinal plant. M. oleifera oil contains all the main fatty acid including oleic acid which is very stable when used for frying. Moreover, it has the potential to become a new source of high oleic acid oil. Previous study only focused on medicinal and nutritional aspects of the tree parts. However, there is lack of information on the morphological variations of M. oleifera grown in Malaysia. Thus, the objectives of the study were to determine the phenotypic variations among M. oleifera accessions, the performance of stem cuttings of two selected accessions as propagation material, and the growth and maturation of pod of a selected accession. Morphological characteristics were used to assess levels of polymorphism across 20 accessions of M. oleifera in situ and ex situ. There were variations on morphological characteristics between accessions of M. oleifera based on stem girth, leaf length, leaf width, pod length, pod diameter, pod weight, seed number/pod, seed diameter and seed weight indicating the presence of genetic variability among accessions. Among the 20 accessions S05 had the highest seed weight followed by S04, with a difference of 3.7% seed weight. As for seed number/pod, accession P05 had the highest value followed by P03, with a difference of 4.7%. These two characteristics are beneficial in the selection of M. oleifera as they contribute to the total yield.

**KEYWORDS :**

**1. Introduction**

M. oleifera (Synonym: Guilandina moringa L. or Moringa pterygosperma Gaertn.) belongs to the Moringaceae family, and it is one of the best known and most widely distributed and neutralized species . The tree is native to Northern India, Pakistan and Nepal. It has been cultivated and has become neutralized well beyond its native range, including throughout South Asia, South East Asia, the Arabian Peninsular, tropical Africa, Central America, the Caribbean and tropical South America. M. oleifera usually grows up to 10 or 12 m in height, with a spreading, open- crown of drooping, brittle branches, feathery foliage of tripinnate leaves, and thick, corky, deeply fissured off-white bark.

Although most of the plants are found to be domesticated in Malaysia, some plants have escaped from cultivation and adapt to the soil and climate conditions. Plant adaptations through mutation often resulted in morphological characteristic changes, and over a period of time and geographical barriers developed into new forms, varieties or subspecies. Although this brings new light in taxonomy, but agriculture often demands uniform planting materials in term of its genetic constituents and morphological characteristics, particularly in its yield. Morphological characterization plays an important part in plant identification and taxonomy through comparison of the quantitative and qualitative traits. For the purpose of registering new hybrids or cultivars, morphological data are considered as most reliable and accepted because appearance, may it be genetically controlled, or environmentally influenced, are the main interest in agriculture.

Prior to any cultivation, establishment of germplasm is a necessity and in this case, collection of M. oleifera genetic resources requires characterization of accessions for the purpose of database and also analysis of their diversity. In India, where the M. oleifera population is naturally distributed, the plants vary in quantitatively inherited traits. . Variations included differences in pod sizes that ranged from 15 to 120 cm. The bisexual, oblique, stalked, axillary and heteromorphic flowers are highly cross-pollinated due to heteromorphism. Occurrence of variations amongst non-cultivated M. oleifera from previous reports suggested that when establishing a new germplasm prior to cultivation, it is always prudent to determine the phenotypic variations in order to obtain the genotype profiles. This study was carried out to describe the phenotypic variations in morphological characteristics of M. oleifera accessions collected from Perak and Selangor, Malaysia.

**2. Materials and methods**

**2.1 Collection sites and plant materials**

The experiment was carried from July 2015 to September 2016. The collection area covered latitudes of 1°46.018 N to 6°39.285 N (North), longitudes of 100° 09.81 E to 103°51.988 E (East) and altitudes of 10 to 634 m in the states of Perak and Selangor, where the M. oleifera plant is commonly found (Table 1). It is believed that the plant was brought into Malaya and planted by the early Indian settlers who came to work in the rubber plantations. As most of the rubber plantations were situated in Perak and Selangor in Malaysia, thus most of the Indians had settled in these two states. Normally, the Indian people grow the M. oleifera plant in their house compound or along roadsides where they can get easy access to the edible fruits, flowers and leaves. The collection sites included the house compound and along roadsides. Ten plants (accessions) were selected randomly from each Perak and Selangor. Selected trees were tagged at the stem base using plastic tags. The flowers, stems, pods, seeds and leaves were collected and packed into a plastic cooler box. The samples were transported in an air-conditioned vehicle to the Postharvest Laboratory, Crop Science Department, Faculty of Agriculture, Universiti Putra Malaysia (UPM). The morphological characteristics of M. oleifera were determined.

### 2.2 Plant parameters recorded at collection sites

Data regarding the collection site of the original sample was recorded in the passport data of each accession ––. A passport data was constructed by modifying and selecting appropriate passport information published by the FAO/IPGRI multi-crop passport descriptors and Plant Research International. The Global Positioning System (GPS-12, Garmin Ltd., U.S.A.) was used to determine latitude, longitude and altitude of collection sites. Morphological characteristics such as stem textures, stem girth, stem colour, leaf length, leaf width, flower weight, flower length, flower colour, pod length, pod diameter, pod weight, number seed per pod, seed size, and seed weight were obtained. For parameters on flower, leaf, pod and seed, five samples were taken from each accession and analyzed.

#### 2.2.1 Stem

Stem texture was visually differentiated based on the roughness or smoothness of the stem surface. Stem girth was measured by circling a string around the stem and the length of the string was measured using a ruler (cm). Point of measurement was made at 1.0 m from the ground level. Stem colour was determined based on the visual colour of the stem, mature stem being either light brown or brown colour.

**Table 1 . Geographical distribution of Moringa oleifera accessions collected from Perak and Selangor.**

| Population | Collection site | State | Latitude | Longitude | Altitude |
|---|---|---|---|---|---|
| S01 | Universiti Putra Malaysia | Selangor | 2°59.210 | 101°42.454 | 48 |
| S02 | Taman Serdang Jaya,Serdang | Selangor | 2°59.671 | 101°43.331 | 43 |
| S03 | Taman Universiti, Sri Kembangan | Selangor | 2°57.132 | 101°51.989 | 331 |
| S04 | Taman Equine | Selangor | 2°56.331 | 101°45.112 | 41 |
| S05 | Kampung Sungai Buah (Dengkil) | Selangor | 2°53.559 | 101°45.030 | 29 |
| S06 | Sungai Buloh | Selangor | 2°24.117 | 101°41.771 | 31 |
| S07 | Batang Kali | Selangor | 3°25.428 | 101°37.430 | 30 |
| S08 | Kerling (Hulu Selangor) | Selangor | 3°34.615 | 101°36.750 | 54 |
| S09 | Bukit Beruntung | Selangor | 3°21.657 | 101°41.222 | 41 |
| S10 | Rawang | Selangor | 3°21.122 | 101°51.909 | 11 |
| P11 | Kampung Jalan Paku (Bidor) | Perak | 4°04.511 | 101°18.090 | 52 |
| P12 | Kampar | Perak | 4°15.734 | 101°10.783 | 65 |
| P13 | Lenggong (Hulu Perak) | Perak | 4°10.621 | 100°59.101 | 91 |
| P14 | Chandan, Kuala Kangsar | Perak | 4°17.640 | 100°15.220 | 11 |
| P15 | Lawin, Kuala Kangsar | Perak | 4°28.729 | 100°15.373 | 28 |
| P16 | Gopeng | Perak | 4°25.229 | 100°16.002 | 689 |
| P17 | Taman Maju, Ipoh | Perak | 4°38.011 | 100°27.890 | 11 |
| P18 | Hospital Pantai, Ipoh | Perak | 4°12.950 | 100°33.973 | 29 |
| P19 | Taiping | Perak | 4°57.641 | 100°32.749 | 23 |
| P20 | Sungai Siput | Perak | 4°34.544 | 100°31.337 | 43 |

Note: Five samples of Moringa oleifera leaves, flowers, pods and seeds were collected in each collection site.

Global Positioning System (Garmin GPS-12) was used to determine position of collection sites.

#### 2.2.1 Leaf

Leaves on rachis number 10, 15 and 20 from the top of the plants were measured. The leaf was measured from the petiole to the tip (cm). Leaves on rachis number 10, 15 and 20 from the top of the plants were measured. The leaf was measured at the widest point of the leaf (cm).

#### 2.2.2 Flower

Flower fresh weight (mg) was determined by weighing 10 harvested flowers from each accession with a weighing scale (Mettler scale B303-S, Switzerland). The flower width (cm) was measured at the longest point of a petal by using a ruler. The flower colour was recorded based on visual observation. The colour of the flower was either white or off-white.

#### 2.2.4 Pod

Pods were harvested when the pod had dried and turned brown (suitable maturity stage for seed-oil extraction). Pod length (cm) was measured on the outer pod in a straight line, from the distal to proximal end, with a ruler. Pod diameter (cm) was measured at the equatorial position of each pod using a varnier caliper (500-171, Mitutoyo Corporation, Japan. Pod weight (g) was obtained with a weighing scale (B303-S College, Mettler Toledo, U.S.A).

#### 2.2.5 Seed

Harvested pods (from the above measurement) were carefully cut along the middle of the pod length. The number of seeds in the individual pods was counted and recorded. Diameter (cm) of seeds was measured using a varnier caliper. The mean diameter of 20 seed samples was calculated. The measurement for seed weight was made by weighing all the seeds in each pod (from the above measurement) using a weighing scale (Mettler scale B303-, Switzerland). Weight per seed (g) was calculated based on average of all seeds per pod.

### 2.3 Experimental design and data analysis

The data were transformed to suit cluster analysis in Table 2. The quantitative data were presented as 1 or 2 for stem texture, representing rough or smooth categories, as 1 or 2 for stem colour, representing light brown and brown colour and as 1 or 2 for flower, representing white or off-white cream categories, respectively. The experiment was arranged in a completely randomized design (CRD) with five samples per accession of M. oleifera. Data were analyzed using analysis of variance and the significant means were separated by least significant difference (LSD) at $P \leq 0.05$. Correlation analysis was performed to indicate the strength of relationship between the parameters measured for pod and seed.

**Table 2 Morphological characteristics of stem and flower from 20 accessions of Moringa oleifera from Perak and Selangor**

| Morphological characteristics | Description |
|---|---|
| Stem colour | 1= light brown and 2= brown |
| Stem texture | 1= rough and 2 = smooth |
| Flower colour | 1= white and 2 = off-white |

The morphological data were further analyzed using Euclidean distance coefficient, NTSYS-pc Versions 2.1 program. These similarity coefficient were used to produce a dendrogram for which the UPGMA algorithm and SAHN clustering (unweighted pair group method using arithmetic average) was employed for depicting the genetic relationships. Data were subjected to principal component analysis (PCA) using the same program. The eigen values were determined in PCA. Eigenvalues indicate the amount of variance explained by each principal component. In order to reduce the influence of scale differences during cluster and principal component analyses, data of quantitative characters were standardized using the STAND program in NTSYS-pc. During principal component and cluster analyses, 20 accessions were represented by rows and 14 taken as variables by columns.

## 3. Results and discussion

The results of morphological characteristics among 20 accessions of M. oleifera showed that values were significantly different for all parameters evaluated (Table 3 and Table 4) except for stem texture, stem colour, flower weight, flower diameter and flower colour. These suggest that selection for relevant characters could be possible. All parameters that statistically showed significant differences with respect to the different accessions indicated a high level of morphological variations. Some characters of this plant such as stem girth, leaf length, leaf width, pod length, pod diameter, pod weight, number seed/pod, seed diameter and diameter weight could be used in the identification and classification of M. oleifera.

### 3.1 Stem

The stem texture is an underappreciated ornamental aspect of woody plants. Stem texture deals with the visible exterior and tactile feel of stems. This plant can be distinguished into two groups, smooth and rough stem, by looking at the bark texture (Table 3). This indicates that the two types of M. oleifera can easily be differentiated based on their stem textures. M. oleifera with smooth stem textures were predominately smooth to the touch and lack any raised protuberances or surface irregularities. M. oleifera with rough stem texture were rough to the touch and appear uneven in texture. Table 3 shows that all M. oleifera from the Perak area had a smooth bark texture. Stem girth of M. oleifera showed that the stem shape of M. oleifera widens down and both M. oleifera from Perak and Selangor had this characteristic.

**Table 3. Descriptive statistics for eight morphological characteristics (stem, leaf and flower) 10 accessions of Moringa oleifera collected from each state, Selangor (S01-S10) and Perak (P11-P20).**

| State | Code | Stem | | | Leaf | | Flower | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Texture | Girth (cm) | Colour | Length (cm) | Width (cm) | Weight (g) | Diameter (cm) | Colour |
| **Selangor** | S01 | 1.0 a | 14 j | 1 a | 2.6 a | 1.5 cde | 24.5 a | 0.72 a | 1.98 a |
| **Selangor** | S02 | 1.0 a | 22 h | 1 a | 2.1 b | 1.4 bcd | 23.1 a | 0.77 a | 1.85 a |
| **Selangor** | S03 | 1.0 a | 13 j | 1 a | 1.7 cd | 1.3 cde | 22.5 a | 0.83 a | 1.83 a |
| **Selangor** | S04 | 1.0 a | 49 c | 1 a | 2.0 b | 1.5 a | 23.2 a | 0.88 a | 1.97 a |
| **Selangor** | S05 | 2.0 a | 43 d | 2 a | 1.9 bc | 1.2 ef | 23.5 a | 0.73 a | 1.86 a |
| **Selangor** | S06 | 2.0 a | 15 ji | 2 a | 2.0 bc | 1.3 bc | 21.2 a | 0.91 a | 1.87 a |
| **Selangor** | S07 | 2.0 a | 14 j | 2 a | 2.2 b | 1.5 ab | 21.4 a | 0.92 a | 1.88 a |
| **Selangor** | S08 | 1.0 a | 18 i | 1 a | 2.1 b | 1.4 bc | 23.2 a | 0.82 a | 1.93 a |
| **Selangor** | S09 | 1.0 a | 13 j | 1 a | 1.8 bcd | 1.2 def | 21.3 a | 0.85 a | 1.92 a |
| **Selangor** | S10 | 2.0 a | 9.8 k | 2 a | 1.9 bcd | 1.3 cde | 22.1 a | 0.84 a | 1.88 a |
| **Perak** | P11 | 1.0 a | 28 g | 1 a | 1.5 ef | 1.1 f | 23.5 a | 0.84 a | 1.87 a |
| **Perak** | P12 | 1.0 a | 40 de | 1 a | 1.6 de | 1.1 ef | 21.2 a | 0.84 a | 1.88 a |
| **Perak** | P13 | 1.0 a | 43 d | 1 a | 1.7 cde | 1.4 bc | 21.4 a | 0.84 a | 1.93 a |
| **Perak** | P14 | 1.0 a | 58 b | 1 a | 1.5 ef | 1.2 def | 22.0 a | 0.87 a | 1.92 a |
| **Perak** | P15 | 1.0 a | 31 gf | 1 a | 1.3 fg | 1.3 cde | 23.3 a | 0.85 a | 1.88 a |
| **Perak** | P16 | 1.0 a | 32 f | 1 a | 1.3 fg | 0.9 g | 22.1 a | 0.83 a | 1.98a |
| **Perak** | P17 | 1.0 a | 40 e | 1 a | 1.6 e | 1.2 ef | 22.0 a | 0.75 a | 1.85 a |
| **Perak** | P18 | 1.0 a | 69 a | 1 a | 1.3 fg | 0.8 g | 22.5 a | 0.71 a | 1.83 a |
| **Perak** | P19 | 1.0 a | 33 f | 1 a | 1.3 fg | 0.91g | 21.9 a | 0.87 a | 1.87 a |
| **Perak** | P20 | 1.0 a | 32 gf | 1 a | 1.2 g | 0.8 g | 23.2 a | 0.77 a | 1.88 a |

PerakP201.0 a32 gf1 a1.2 g0.8 g23.2 a0.77 a1.88 az Means within a column followed by the same letter are not significantly different by LSD at P≤0.05.

Stem texture: 1= smooth, 2 = rough, Stem colour: 1 = light brown, 2 = brown, Flower colour:1 = white 2 = off-white.

**Table 3.4. Descriptive statistics for six quantitative morphological characteristics analysed (pod and seed) on the 10 accessions of Moringa oleifera collected from each state, Selangor (S01-S10) and Perak (P11-P20).**

| State | Code | Pod | | | Seed | | |
|---|---|---|---|---|---|---|---|
| | | Length (cm) | Diameter (cm) | Weight (g) | Number per pod | Diameter (cm) | Weight (g) |
| **Selangor** | S01 | 42 e | 2.4 fghij | 13 fghi | 21.5 abc | 1.12 h | 22.6 f |
| **Selangor** | S02 | 44 de | 2.0 h | 15 de | 20. 5bcd | 1.41 ef | 20.9 fg |
| **Selangor** | S03 | 44 de | 2.2 ghij | 15 de | 20. 5bcd | 1.36 f | 15.6 h |
| **Selangor** | S04 | 59 a | 2.5 fgh | 19 a | 20. 5bcd | 1.48 de | 32.1 ab |
| **Selangor** | S05 | 45d | 2.5 efg | 12 hi | 20.5bcd | 1.55 d | 33.3 a |
| **Selangor** | S06 | 49 c | 2.4 fghi | 13 gh | 20.0 bcd | 1.40 ef | 31.1 bc |
| **Selangor** | S07 | 55 b | 3.1 a | 15 cd | 20.0 bcd | 2.20 a | 29.9 cd |
| **Selangor** | S08 | 50 c | 2.8 bcd | 14 def | 18.5 d | 2.04 b | 24.1 e |
| **Selangor** | S09 | 31 fg | 3.1 a | 16 bc | 22.0 ab | 0.81 j | 23.9 e |
| **Selangor** | S10 | 46 d | 3.0 ab | 11 i | 21.5 abc | 1.26 g | 21.1 fg |
| **Perak** | P11 | 30.g | 2.0 h | 9.5 j | 19.5 cd | 1.11 h | 12.5 i |
| **Perak** | P12 | 32 fg | 2.4 fg | 11 i | 18.5 d | 1.05 h | 22.2 f |
| **Perak** | P13 | 31 fg | 2.8 bc | 17 b | 22.0 ab | 0.83 j | 25.1 e |
| **Perak** | P14 | 24 h | 2.7 cde | 12 ghi | 13.5 f | 0.95 i | 20.3 g |
| **Perak** | P15 | 23 h | 2.3 ghij | 18j | 23.0 a | 0.97 i | 17.6 h |
| **Perak** | P16 | 34 f | 2.0 gh | 15 cd | 21.5 abc | 1.06 h | 17.0 i |
| **Perak** | P17 | 18 h | 2.0 gh | 9.5 j | 13.0 f | 0.96 i | 18.3 h |
| **Perak** | P18 | 25 h | 2.4 fg | 13 efgh | 14.5 ef | 1.08 h | 19.0 h |
| **Perak** | P19 | 41 e | 2.4 fghi | 14 def | 20.5 bcd | 2.15 a | 29.0 d |
| **Perak** | P20 | 32 fg | 2.6 def | 13 fghi | 15.5 e | 1.81 c | 22.1 f |

z Means within a column followed by the same letter are not significantly different by LSD at P≤0.05.

Stem texture: 1 = smooth, 2 = rough, Stem colour: 1 = light brown, 2 = brown, Flower colour:1 = white 2 = off-white.The high variability in stem girth of M. oleifera could be due to age differences among the accessions (Table 3). The bark of the young plants could differ markedly according to the stem girth naturally. A young M. oleifera plant has a smaller girth than an old M. oleifera plant. Accession P18 had the biggest stem girth (69 cm) compared to other accessions. Based on the visual observation, there is very distinguishable appearance in terms of stem colour between M. oleifera. The M. oleifera with smooth stem texture is light brown or buff in colour while the M. oleifera with rough stem texture is brown in colour. The light brown colour is a pale yellow toned with gray brown for example a dingry yellowish brown or very pale tan just like the smooth M. oleifera stem colour. The brown colour is a colour that is a dark yellow, orange or red, of low luminance relative to lighter or white coloured just like the rough M. oleifera stem colour. These distinguished colours indicate that the two types of M. oleifera with smooth and rough can easily be differentiated.

### 3.2 Leaf

In all of the accessions, the general appearance of the leaves of M. oleifera were alternate, composite, bippinate, with 2 to 6 pairs of opposite pinnae bearing opposite leaflet in 3 to 5 pairs suggest it to be inherent characters. Similarly, possession of terminal leaflet that is larger more or less glaborous, ovate to elliptic, rounded at apex and base, with the petiole being pubescent and generally with mean leaf sizes considerably variable even among samples from same location. Both of the M. oleifera from Perak and Selangor had the same leaf type, which was compound tripinnate. Compound leaves are made of leaves divided into smaller group leaflets. Compound leaf has the blade part separated into smaller pieces and the botanical term is pinnate.

Thus, leaf of M. oleifera cannot be differentiated between Perak and Selangor. If a plant has its leaves on a stalk, there are several ways in which they may be arranged. Most commonly, they are either in pairs, opposite one another on the stem, or they are produced singly in an alternate arrangement. Leaves can also be three at a node, or several of them in which case they are said to be whorled. M. oleifera from Perak and Selangor had the same leaflet arrangement which was opposite (Table 3). Most leaves appear simple at first sight and they consist of only a few cell types. Despite that, many developmental processes are involved in leaf ontogeny, including positioning and initiation of leaf primordia, specification of leaf identity, establishment of dorsiventrality, the control of cell division and expansion and pattern formation. Thus, it shows that genetic content in the leaves influenced leaf growth and development (Duke, 1985).

One way of determining the size of M. oleifera leaflets in order to differentiate between the two types of M. oleifera is by measuring the leaflet length. Table 3 shows that there were significant differences (at P≤0.05) in leaflet length for M. oleifera accessions. This could be due to genetic constituents in Selangor M. oleifera leaves that produce longer leaflets compared to the genetic content of the Perak M. oleifera that produce relatively shorter leaflets. Despite that, the leaflet shape of the Perak and Selangor M. oleifera were similar with each other, the Selangor M. oleifera had a bigger size of elliptic leaflets in terms of leaflet length (Table 3.3). Leaf morphological characteristics after clustering as in this work have shown region non-specificity groupings in all clusters, indicating a high possibility of exchange of genetic information between samples since they are not completely isolated.

### 3.3 Flower

Table 3 shows that there were no significant differences (P≤0.05) in flower size for Perak and Selangor M. oleifera. Both have a relatively similar weight of 10 fresh flowers. Thus, flower size cannot be differentiated between the M. oleifera morphological characteristics.

A flower, also known as a bloom or blossom, is the reproductive structure found in flowering plants (plants of the division Magnoliophyta, are also called angiosperms). The biological function of a flower is to mediate the union of the male sperm with female ovum in order to produce seeds. The process begins with pollination, is followed by fertilization, leading to the formation and dispersal of the seeds. The grouping of flowers on a plant is called the inflorescence. M. oleifera flowers were obliquely monosymmetric and papilinoid (the median petal is adaxial) with five stamens, were in axillary pendulous panicles 1.5 to 2 cm long from leaf corners . The flowers were borne on slender, hairy stalks in spreading or drooping lateral flower clusters 10 to 25 cm long. Individual flowers were approximately 0.7 to 1 cm long and 2 cm broad with five thinly veined petals . Based on the visual observation, both the Perak and Selangor M. oleifera have similar flower colour which is off-white.

### 3.4 Pod

The pod diameter ranged from 2.0 cm to 3.1 cm. Accession S07 had the largest pod diameter while P11 had the smallest pod diameter (Table 4). In pod length, accession S04 had the longest pod followed by accession S07 while accessions P15, P17 and P18 had the shortest pod, with pod length between 18.5 to 25.7 cm. Accessions S07 and P19 had the highest seed diameter. The differences could be caused by environmental factors or genetic diversity among these accessions of M. oleifera. These results agree with that the pod size is markedly influenced by the environment during the pod enlargement period of growth. Pod size reduction caused by genetic variation or other stress during the pod enlargement period could reduce yield substantially. Edible-pod accessions with largest and longest pods could be introduced to the farmers.

### 3.5 Seed

The number of seeds/pod was between 13-23. Accession P15 had a significantly higher number of seeds, which were 23 seeds per pod compared to other accessions. Accessions S05 had the highest seed weight (Table 4). A total of three quantitative variables (number seed per pod, seed diameter and seed weight) were showed highly significant differences between M. oleifera accessions. Thus, these variables might be useful for morphological identification although these are also variables caused by age of the M. oleifera planted.

Accessions that originated from Selangor have dominant values in pod and seed production. Accessions number S05 and S07 had higher number of seeds which can be a potential characteristic as a planting material. This result showed that there was a variation in seed production among accessions. The significant differences revealed by the listed attributes of yield could be due to environmental influences on the parental genetic constitution. Similar findings for pea were reported by . Environmental factors and genetic variability among accessions contributed to the different seed yield of M. oleifera. reported that seed size variation reflects the adjustment to both previously fixed yield components and changes in environmental conditions.

### 3.6 Correlation of pod and seed

Strong correlations are observed between pod size (pod diameter and pod length) and seed number, seed length, seed diameter and seed weight (Table 5). There were significant and positive correlations between pod diameter and weight per seed (r=0.86) and pod length (r=0.91). Weight per seed was also positively correlated with seed number (r = 0.80), seed length (r= 0.90) and seed diameter (r=0.87).

Correlations between morphological characteristics were highly significant. This shows that there is a strong relationship between the pod size and the production of seed. The seed growth rate is determined by the sink size of pod . The tightly packed arrangement of M. oleifera seeds in each pod indicated that physical restriction of each seed could have occurred. The high positive correlation of pod length with seed number, seed

**Table 5. Correlation coefficients between pod diameter, pod length, seed number, seed length, seed diameter and weight of seeds for the 20 accessions of Moringa oleifera collected from Selangor and Perak.**

| Parameter | Pod diameter (cm) | Pod length (cm) | Seed number | Seed length (cm) | Seed diameter | Weight/ seed |
|---|---|---|---|---|---|---|
| Pod diameter cm) | -- | 0.89** | 0.58* | 0.97** | 0.85** | 0.86** |
| Pod length (cm) | | -- | 0.79** | 0.91** | 0.93** | 0.91** |
| Seed Number | | | -- | 0.63* | 0.79** | 0.80** |
| Seed length (cm) | | | | -- | 0.90** | 0.90** |
| Seed diameter (cm) | | | | | -- | 0.87** |

For correlation coefficients, n=5
*,** Significant at 5% and 1% levels of probability, respectively.

length, seed diameter and seed weight indicated that when pod length increased, yield would also be increased. This is because the main yield production of M. oleifera is for seed .

Positive association of seed number with seed length, seed diameter and seed weight showed that when the seed number was high, the seed length and seed diameter determined were longer and larger. Seed diameter and weight were significantly and positively correlated with seed length. Also, the seed diameter was highly and significantly correlated with the seed weight. These findings would increase the utility of the M. oleifera's collection for future selection program in order to produce cultivars, which are potentially suitable for breeding purposes.

### 3.7 Cluster analysis

The standardized morphological data were used to calculate the Euclidean distances between the M. oleifera accessions and an UPGMA dendrogram was constructed from these values (Figure 1). In this dendrogram, the 20 M. oleifera accessions appeared to form six major clusters. Cluster I has 3 members, S01, S02 and S03. Cluster II consisted of 3 members, S05, P19 and P20. Cluster III has 5 members, P11, P12, P15, P16 and P18. Cluster IV has only one accession which is P13 and Cluster V consist of P14 and P17. Cluster VI has the most number of members namely S04, S06, S07, S08, S09 and S10. The accessions collected from different region still clustered in the same group (Figure 1), it may suggest the possibility of human transfer of seed source across state. The implication of this analysis suggests a basis of morphological differences exhibited by the accessions albeit the fact that they are collected from different geographical locations.

Some characters of trees, stems, flowers, leaves, pods and seeds may be useful for the identification of M. oleifera in Malaysia. A total of 14 qualitative and quantitative variables were used to classify the M. oleifera into six groups corresponding to their geographical origin (Table 6). From the quantitative analysis, there were highly significant difference in leaf, pod and seed parameter, they might be useful for morphological identification although there are also effects from environmental factors. However both the qualitative and quantitative measurement of flowers might not be useful for future identification, as no significant variations were found among accessions.

**Table 6. Moringa oleifera accession groupings revealed by cluster analysis.**

| Cluster | No. of accessions | Accessions names |
|---|---|---|
| I | 3 | S01, S02, S03 |
| II | 3 | S05, P20, P19 |
| III | 5 | P11, P15, P16, P12, P18 |
| IV | 1 | P13 |
| V | 2 | P14, P17 |
| VI | 6 | S04, S09, S06, S10, S07, S08 |

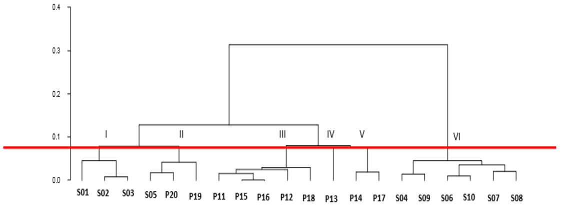


**Figure 1. The dendrogram of 14 morphological traits derived from 20 M. oleifera accessions constructed using the UPGMA method. The scale is based on Euclidean distance coefficient.**

More importantly, insignificant differences in flower morphology of these selected accessions showed that these accessions did not go through extensive mutation to develop into new form, varieties nor sub-species, indicating that this species was able to adapt easily into a new environment and retain their genetic makeup.

The selection of the qualitative and qualitative morphological characteristics was based on the stability of the characteristics which caused it to be influenced either by the environmental factors or the interaction between the genetic and its environment. However, the probability of variations, which might appear on the characteristics, was purely due to the influence of the genetic variations. The use of the qualitative morphological descriptors was found to decrease the refraction shown by the variations in the plant environmental conditions.

Morphological characterization has been widely used to describe varieties, although new molecular techniques have been deployed increasingly in recent years for the identification of different crops. Morphological characters were used to describe three potential citrus rootstocks . The advantages of morphological characters are saving in cost and time and the provision of horticultural characters. Because of the plasticity and instability of phenotypic characters, cultivar identification and diversity were often in contrast with actual genetic diversity. Previous author,  used both morphological data and molecular analysis. Furthermore, important horticultural characters are reported to be controlled by multiple genes . Thus, morphological characterization could be an essential component since most of the horticultural characters cannot be evaluated through molecular markers.

## 4 Conclusion

This study showed the existence of wide variation in morphological characters among M. oleifera accessions. The cluster constructed using qualitative and quantitative characters of plant was able to identify the accessions to specific locations. Furthermore, stem, leaf, pod and seed differed significantly among the locations. These variations are indicative of the underlying genetic diversity and the influence of environmental factors. This study also unravels basic information on important horticultural characters of different accessions. This research provide an understanding of the tree. Introduction of this plant into a farm which has a biodiversity environment can be beneficial for both the owner of the farm and the surrounding eco-system. The planting of M. oleifera trees by smallholders should be encouraged because it will improve both their health and income. From this result, it can be concluded that both environmental and genetic parameters are very important in choosing the best accessions to be promoted.

## Conflict of interest statement

We declare that we have no conflict of interest

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