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Phenotypic characterization of selected Kenyan Khat (*Catha edulis)* cultivars based on morphological traits

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Khat (*Catha edulis* Vahl) is an evergreen shrub habitually ingested for its euphoric and stimulatory effects. The crop is grown in the Middle East, Somalia, East Africa and Ethiopia. It is referred to as *Miraa* in Kenya, while in Yemen it has several names like *Qat, Kat, Kath, Gat, Chat* and *Tschat*. It belongs to the sub-order Rosidae, family Celastraceae and characterized by astringent taste. Despite the daily use and consumption of khat by millions of people in Kenya, little is known about its phenotypic. Phenotypic characterization is an essential approach for assessment of khat diversity; however, it is limited by morphological plasticity and multiple lineage evolution. The study aimed at evaluating the phenotypic diversity of selected khat cultivars grown in Embu and Meru Counties. Ninety samples from 18 cultivars were collected for phenotypic characterization. MINITAB 17 Software was used for description of principal component and construction of dendrogram using the Euclidean distance tool where 58.7% variability was observed among 13 traits studied in 90 samples of khat. Phenotypes grouped into 2 clusters phenotypic diversity showed considerable variability based on 13 khat traits. This will be useful in breeding and characterization programmes of khat cultivars.

Key words: Phenotypic, khat, diversity, cultivar.

INTRODUCTION

Khat (*Catha edulis* Vahl) is ever green and an edible plant (Ngari et al., 2018). It is classified in the kingdom Plantae, class Magnoliopsida, order Celastrales, family Celastraceae, genus *Catha,* and species *edulis* (Sikiru, 2012). The plant was first described a by Swedish botanist called Peter Forskal. He encountered the plant as he travelled to Yemen through Egypt in an expedition that was paid for by King of Denmark Friederick, who wanted all the natural collections (Al Motarreb et al., 2002). Countries and communities have different names for the plant such as *Qat* and *Chat* in Yemen and Ethiopia, *Jaad* and *Qaad* in Somalia, *Muguka* and *Miraa* in Kenya, and *Jimma* in the Oromo language. In most western countries, it is recognized as khat (Ngari et al., 2018).

Khat is said to have originated from Ethiopia and then spread to East Africa and Yemen (El-Menyar et al., 2015). In Kenya, khat is grown in Meru and Embu Counties for commercial purposes. In Meru County, it is grown around Nyambene hills in Meru North, 320 km North East of Nairobi and the main outlet is Chyulu Maua Town (Nyongesa and Onyango, 2010). In Embu county, khat is mainly found in lowland area of Mbeere region

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which are usually dry (Kiunga et al., 2016).

Khat also flourishes in arid and semi-arid environments where temperatures range from 5 to 35°C with free draining soil (New Agriculturalist, 2007; Nyongesa and Onyango, 2010). Khat leaves are harvested in the morning then covered with fresh banana leaves and polythene bags to maintain freshness (Nyongesa and Onyango, 2010). Khat is more potent during the dry and sunny season of the year (Ng'ethe, 2012). Harvesting is done throughout the year and planting is spread over a period of time to obtain a continuous supply. Khat is usually consumed by chewing the leaves while fresh, although occasionally, leaves are dried, then consumed as a stimulating juice (Wabe, 2011).

Mainly, khat chewing is a male habit though it has gained popularity among women (Mwenda et al., 2003; Kiunga et al., 2016). More than 10 million people worldwide use khat for psychostimulating effect (Gitonga et al., 2017). In many countries, khat is chewed for social and psychological reasons (Al-Kholani, 2010). Current trends indicate that khat is used by all societal groups regardless of age, gender, affluence, class, education and occupation (Gesese, 2013).

Various khat cultivars are identified based on the communities that consume them. In Yemen, local cultivars of khat are described according to geographical location, growth habit and physical appearance (colour of the leaf, stem sizes and potency) (Al-Thobhani et al., 2008). It is documented that different kinds of khat vary in the extent of their pharmacological activity. Farmers in Yemen have four cultivars, namely, *Abyadh, Azraq, Aswad* and *Ahma*r categorized according to the colour of shoots and growing twig (Ngari et al., 2018). Also forty kinds of khat cultivar were recognized according to geographical origin. In Ethiopia, two major cultivars have been described as *Dimma* (red) or *Ahde* (white). Getahun and Krikorian, classified khat into three types, namely 'madness-causing, intoxicating-like spirit and insomnia-causing based on their effects (Al-Thobhani et al., 2008; Ayana and Mekonen, 2004). Little is known of how *C. edulis* differs morphologically, therefore, in this study the plant was characterized morphologically.

Morphological traits have been used for classification of variety duplicates, determination of genetic diversity and correlation with characteristics of agronomic significance (Zeng, 2015). Morphological characterization of khat cultivars in Embu and Meru counties was done by assessing variations in 13 khat traits. This has been traditionally used for classification of khat cultivars. It is clear that most users' knowledge is limited to only the narcotic effects of the plant (Al-Thobhani et al., 2008).

Khat plant cultivars are notable by the level of cathionone (or the narcotic effect of the plant) present in the plant material and also their morphological differences. The varying morphological features and cathionone levels within khat may also be as a result of genetic variations. Morphologic studies have not been conducted for this crop in Mt. Kenya region where it has become a major income earner. As a traditional method, morphological traits are used to assess genetic divergence and classify existing germplasm materials. In addition, this technique is easier, cost effective, and easy to score and requires less time and finally it does not need any technical knowledge (Malek et al., 2014).

MATERIALS AND METHODS

Collection sites

The germplasm was collected from 12 major khat producing wards (Figures 1 and 2). The wards included: Maua, Kianjai, Gaiti, Kangeta, Muthaara in Meru County and Kaaga South, Kithimu, Kaaga North, Mbeti south, Mbeti North, Mavuria, and Muminji in Embu County. Meru County is found in eastern region of Kenya approximately 225 km northeast of Nairobi, it covers an area of 6,936 km^2. The area receives about 1366 mm per annum. The climate of Meru is described as cool and warm with temperature ranging between 16°C during cold season and 23°C during hot warm season. Embu County is located approximately at 120 km northeast of Nairobi towards Mt. Kenya. It covers an area of 2,818 km^2 and lies between latitude 0° 8" and 0° 35" South and longitude 37° 40" East.

Collection of plant materials

Khat fruits, flowers and fully grown leaves were harvested from selected khat plants for morphological characterization. A total of 90 samples were collected from locally available cultivars and three replicates picked randomly from each of the sample giving a total number of 90 khat samples where different local khat names were given.

For morphological characterization, a small branch of the khat plant was cut aseptically and each packed in between a newspaper and later transported to National Museums Herbarium Department, where identification and morphological measurements were carried out. All the information on these plants was recorded based on local names given by Meru and Embu communities as well as geographic distribution (Appendix 1). The local names given by farmers included: *Kira Kieru*-1*, Kira Kieru*-2*, Kira Kieru*-3*, Kira Kiiru*-1*, Kira Kiiru*-2*, Kira Gitune*-1*, Kira Gitune*-2*, Muchuri, Kithara, Mutimutiri, Mugiza*-1*, Mugiza*-2*, Mugumo*-1*, Mugumo*-2*, Mugumo*-3*, Mugumo*-4*, Mugumo*-5*, Muguka*-1*, Muguka*-2*, Muguka*-3*, Muguka*-4*, Muguka*-5, *Mugukawakarimi, Gitu,Mutamucii*, *Mukurukuru*, *Muruti*, *Muceke*, *Mitune*, and *Mumbu.*

A total of 13 morphologic traits of 90 khat samples were evaluated according to guidelines provided by Robson (1994). The traits studied were leaf length, leaf width, leaf margin, petiole length, inflorescence length, peduncle length, sepal length, petal length, and diameter of the stamen, diameter of the filament, ovary length, capsule length and the length of the wing (Appendix 2). For each of the khat sample, 3 leaf samples were randomly selected and their measurement taken using a digital vernier caliper. Flowers and fruits measurements were taken using wild MSA Switzerland dissecting microscope at magnification of ×10.

Data management and statistical analysis

Raw data was entered into Microsoft excel spreadsheet which was then imported to Minitab software version 17.0 (State College Pennsylvania-USA) software. Data was analyzed statistically for the

Figure 1. A map showing Meru County.
Source: Survey of Kenya (2011).

Figure 2. A map showing Embu County.
Source: Survey of Kenya (2011).

differences in means for 13 khat traits, through one-way ANOVA followed by Tukey's post hoc test. Statistical significant differences were set at p≤0.05. Cluster analysis yielded a dendrogram that was used to examine the morphological relatedness among the 90 khat samples while Principal Component Analysis (PCA) was used to assess the underlying source of variation in morphology.

RESULTS

The measurements of the 13 phenotypic traits studied were found to vary across the 90 khat samples, *Muguka*-4 had the highest mean leaf length of 99 mm, while *Mutimutitiri* had the lowest mean leaf length of 53.67 mm. The mean leaf length of the 90 khat samples had significant differences. Regarding leaf width, *Muguka*-4 had the highest mean value of 45.67 mm, while *Kira Kieru*-1 had the lowest mean leaf length of 19.33 mm (Appendix 2).

The mean sepal length of the studied samples did not show significant differences among themselves with the highest mean length of 1.47 mm being recorded in *Mugumo*-1, *Mugiza*-1, and *Kithara*. The lowest mean sepal length of 1.03 mm was observed in *Mugumo*-3 and *Mugumo*-4, *Mugukawakarimi*, *Mukurukuru*, *Mumbu*, *Muruti, Mutamucii* and *Mutimutiri*. The highest mean petal length of 1.67 mm was observed in *Kira Gitune*-1, *Kira Kieru*-1 and *Kira Kiiru*-1 cultivar from Meru, while the lowest mean petal length of 1.00 mm was observed in *Muguka*-2. The mean petal length of the 90 khat samples did not show significant differences among themselves. The mean stamen diameter of the studied khat samples did not show significant differences with the highest diameter of 2.37 mm being shown in *Kira Gitune*-1 and the lowest mean diameter being 1.07 mm in *Kira Kieru*-2 (Appendix 2).

The mean filament diameter of all samples ranged from 1.53 to 1.07 mm. The highest mean filament diameter was observed in *Muchuri* and *Kira Kiiru*-1, while the lowest filament diameter was shown in *Gitu*, *Kira Kieru*-2, *Muguka*-4, *Muguka*-5, *Muguka*-1, *Muguka*-2*, Mugukawakarimi*, *Mugumo*-3, *Mukurukuru*, *Muruti* and *Mutimutiri* cultivars. There were no significant differences in ovary length mean among all cultivar with *Mugiza*-1 showing the highest length of 1.57 mm, while *Kira Kieru*-3 and *Kira Kiiru*-2 showed the lowest mean ovary length of 1.10 mm. There were significant differences in mean capsule length among the 90 khat samples with the highest capsule length of 8.43 mm being observed in *Gitu* and lowest mean capsule length of 5.1 mm in *Kira Gitune*-1, and *Kira Kieru*-3. The mean wing length varied significantly among various cultivar with the highest mean wing length of 3.5 mm in *Muguka*-1 and lowest wing length of 2.07 mm in *Kira Gitune*-1.

The leaf margin had wide variations ranging from a mean of 107.33 mm (*Muguka*-4) to a mean of 55.00 mm (*Mutimutiri*). The mean leaf margin of *Mutimutiri* cultivar was significantly lower than the other cultivars (p≤0.05). Besides, the mean petiole length did not show significant differences among the cultivar with the highest mean petiole length being 5.67 mm (*Muguka*-1 and *Muguka*-2) and the minimum mean petiole length being 2.33 mm observed in *Mugiza*-1 and *Muguka*-3 cultivars (p>0.05). The mean inflorescence length of 90 khat samples did not show significant difference among the 90 samples. *Kira Kieru*-1 had the highest mean inflorescence length of 21.00 mm, while *Mutimutiri* had the lowest inflorescence length of 14.33 mm. The mean peduncle length of 30 samples was not significantly different with *Kira Gitune*-1 having the highest mean of 7.33 mm, and *Mugumo*-1, *Mugumo*-2 and *Mugiza*-1 having lowest mean peduncle length of 4.00 mm (p>0.05).

A dendrogram constructed from data set of mean values of the 13 khat traits showed 2 super clusters namely I and II. Super cluster I had two sub clusters Ia and Ib. The sub cluster Ia comprised *Mugukawakarimi* and *Kira kieru*-3 cultivars from Embu and Meru counties, respectively, while sub cluster Ib had *Muguka*-4 cultivars from Embu county, clustering independently. The sub cluster II was more diverse and clustered into two sub clusters IIa and IIb. The sub cluster IIa was divided into IIai and IIaii sub clusters. The sub cluster IIai had three groups where the first group comprised *Gitu* and M*uguka*-3 cultivars from Embu County while the second group comprised *Kira kiiru*-2, *Kira kieru*-2 and *Kiithara* cultivars from Meru County. The third group comprised *Kira gitune*-2 and *Kira kieru*-1 cultivars from Meru county and *Mugumo*-4 cultivar from Embu county*.* The sub cluster llaii had one group which comprised *Muguka* -5 and *Mugumo*-1 genotypes both from Embu County.

The sub cluster IIbi had four groups. The first group comprised of *Gitune*, *Muruti, Mukurukuru Mugumo*-5, *Mugiza*-2 and *Muguka*-1 cultivar from Embu County and *Kira kiiru*-1 cultivar from Meru County. The second group comprised *Muceke* and *Mugiza*-1 from Embu County. The third group comprised *Mugumo*-2, *Muguka*-2, *Mugumo*-3 and *Mumbu* from Embu County, while the fourth group comprised *Kira gitune*-1 and *Muchuri* cultivar from Meru county, and *Mutamucii* from Embu county. The sub cluster IIbii had *Mutimutiri* cultivar which originated from Embu County (Figure 3).

Principal component analysis (PCA)

The PCA was performed for all the 13 traits in the 90 khat samples as indicated in Table 1. Out of the 13 traits, three principal components (pc1, pc2 and pc3) exhibited more than one Eigen value and showed about 58.7% variability among the 13 studied traits. The three principal components: pc1, pc2 and pc3 had 25.0, 18.4 and 15.3% variability, respectively among the cultivars for the traits under study. The pc1, pc2 and pc3 had Eigen values of 3.247, 2.386, and 1.995, respectively.

The pc1 was positively correlated to leaf margin, petiole

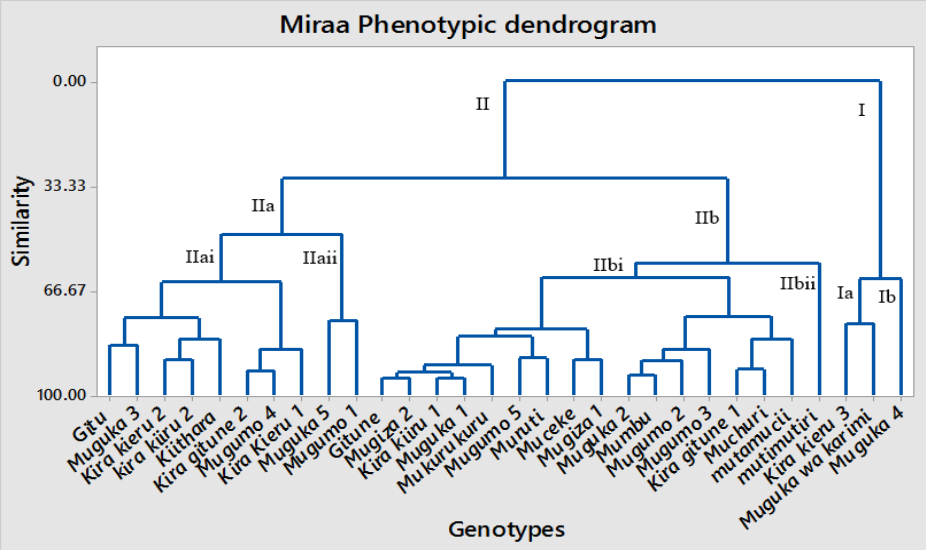

Figure 3. Euclidean distance based dendrogram developed from mean values of the 13 traits of 90 khat samples.

Table 1. Principal Component Analysis among the 30 khat cultivars.

Parameter	PC1	PC2	PC3
Eigen value	3.247	2.386	1.995
% Total variance	25.0	18.4	15.3
% Cumulative	25.0	43.3	58.7
Trait	**Eigen vectors**		
Leaf length (mm)	-0.062	0.310	-0.546
Leaf width (mm)	-0.323	0.030	-0.305
Leaf margin (mm)	0.107	0.488	-0.359
Petiole length (mm)	0.191	0.163	0.247
Inflorescence length (mm)	-0.120	0.037	-0.167
Peduncle length (mm)	0.466	0.159	0.090
Sepal length (mm)	0.124	-0.279	-0.541
Petal length (mm)	0.402	-0.148	-0.004
Diameter of stamen	0.386	-0.143	-0.111
Diameter of filament	0.224	-0.391	-0.147
Ovary length	0.074	-0.485	-0.214
Capsule length	-0.416	-0.314	0.053
Wing length	-0.241	-0.077	0.087

length, peduncle length, sepal length, petal length, stamen diameter, filament diameter and ovary length. However, it was negatively correlated to leaf length, leaf width, inflorescence length, capsule length and wing length. The pc2 showed a highly positive correlation to leaf length, leaf margin, petiole length, inflorescence length and peduncle length. However, it was negatively correlated to sepal length, petal length, stamen diameter, filament diameter, ovary length and wing length. The pc3 showed a positive correlation to petiole length, capsule length, peduncle length and wing length but negatively correlated to leaf length, leaf width, leaf margin, inflorescence length, sepal length, petal length, stamen diameter, filament diameter and ovary length (Table 1).

The scatter plot of the 30 khat samples based on the 13 studied traits was also drawn in this study. It was observed that the first quadrant comprised of *Muguka*-1, *Muguka*-2, *Muguka*-3 and *Mugiza*-2, *Muguka wa karimi, muguka*-4, *Muguka*-5 and *Gitu* cultivars, which were from Embu county. The second quadrant had six cultivars,

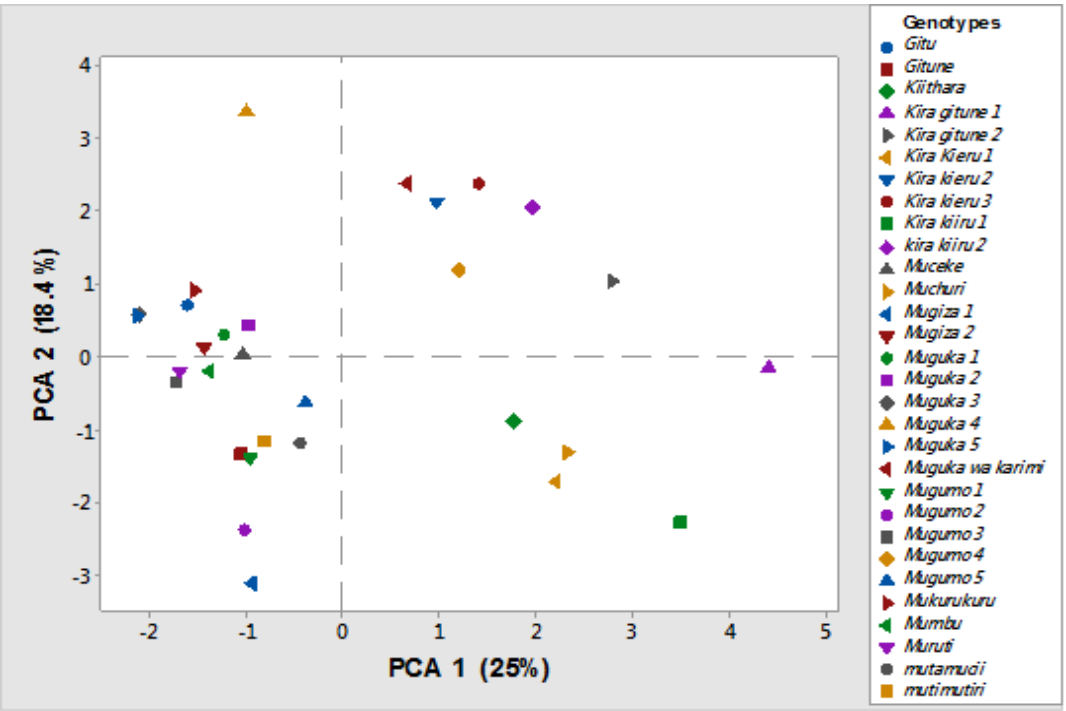

Figure 4. The scatter plots of 30 khat cultivars based on 13 traits

comprising *Kira kieru*-2, *Kira kieru*-3, *Kira kiiru*-2, *Kira gitune*-2, from Meru County, as well as *Mugumo*-4 and *Mukurukuru* cultivar from Embu County. The third quadrant comprised *Mugumo*-3, *Mugiza*-1, *Mugumo*-1, *Mugumo*-2, *Gitune*, *Mutimutiri, Muruti*, *Mumbu, Muceke* and *Mugumo*-5 cultivar from Embu County. The fourth quadrant comprised five cultivar, which originated from Meru county and they included *Kira gitune*-1, *Kiithara, Muchuri*, *Kira kieru*-1 and *Kira kiiru*-1 (Figure 4).

DISCUSSION

The mean leaf length of different cultivars indicated significant differences, whereby *Muguka*-4 cultivar recorded the highest mean leaf length while *Mutimutiri* cultivar showed the lowest mean leaf length. Both cultivars were collected from the same geographical location and environmental conditions. The lowest leaf length of *Mutimutiri* cultivar could be due to the ability of the cultivar to adapt to relatively drier and saline environment. Plants growing in such conditions usually have reduced leaf area in order to minimise loss of water during evaporation (Deblonde and Ledent, 2001).

The mean leaf width of different cultivar was significantly different, *Muguka*-4 cultivar recorded the highest mean leaf width while *Kira Kieru*-1 recorded the lowest leaf width. The lowest leaf width of *Kira Kieru*-1 could be as a result of adaptations to conserve water. *Kira Kieru*-1 had narrow leaves and less total surface area than *Muguka*-4 and, therefore, loses more water compared to *Kira Kieru*-1. Studies have shown that broad leaves heat up more than narrow leaves of the same length, hence narrow leaf plants are well adapted to dry and hot environments. The cultivar with the highest mean leaf width (*Muguka*-4) could have been in moist and shady environments, which enhanced their ability to absorb sunlight. This was confirmed in tropical vines (Tardieu, 2013).

Cultivars with long inflorescence produce more flowers, fruits and seeds compared to cultivars with short inflorescence (Rahman et al., 2009). This suggests that both the number of inflorescences and the number of female flowers are the main factors in determining yield. On the other hand, determining the number of inflorescences per plant and the number of female flowers both depend on environmental factors (Sangoi, 2001; Domiciano et al., 2014). Flowering and determination of the flower type are influenced by the occurrence of low temperatures and high rainfall (Inouye, 2008; Prasad et al., 2001). Therefore, the highest inflorescence length in *Kira Kieru*-1 could have resulted from high rainfall and low temperature, while the lowest inflorescence length in *Mutimutiri* could be due to low rainfall and high temperature.

The mean peduncle length showed significant differences among different cultivars. Drought and stress cause reduction in peduncle length. From this study *Kira Gitune*-1, which had the highest peduncle length was not drought stressed while *Mugumo*-1, *Mugumo*-2 and

Mugiza-1 which had the lowest peduncle length were drought stressed. This was confirmed by Amiri et al. (2013) on 80 bread wheat genotypes.

The mean sepal length of different genotypes showed significant differences among different cultivars. The difference in sepal and petal length could be related to availability of water (Kwak et al., 2007). Therefore, the longer the petal and sepal the higher the water availability. Hence cultivars *Mugumo*-1, *Mugiza*-1, *Kiithara*, *Kira Gitune*-1, *Kira Kieru*-1 and *Kira Kiiru*-1 were in place with enough water. The mean stamen length of many cultivars showed significant differences with the highest diameter of 2.37 mm being recorded in *Kira gitune*-1 cultivar from Meru and the lowest mean diameter of 1.07 mm was recorded in *Kira kieru*-2 cultivar from Meru. Availability of water results to increased flowers size and stamen length. Consequently, the traits usually related to floral attractiveness are increased (Natalia et al., 2015). However, fecundity potential is not increased. These results suggest that population differentiation in floral characters could be a caused by random genetic drift that occur in relatively small or isolated populations. It may also be attributed to restricted gene flow (Medrano et al., 2005).

The mean filament diameter showed significant difference among the cultivar with mean filament diameter ranging between 1.53 and 1.07 mm. *Kira Kiiru*-4 had the highest, while *Gitu*, *Muguka*-4 and *Muguka*-5 had the least 1.07. The floral parts of a plant are highly affected by the environment and this could be reasons as to why significant difference was recorded in filament diameter. The mean ovary length of all cultivars indicated no significant difference. This could be due to similar ancestry of all the cultivars studied. The mean capsule length showed significant difference among 30 khat samples, such could be used to discriminate cultivars. *Gitu* had the highest capsule length while *Kira gitune*-1 and *Kira kieru*-3 cultivar had the lowest. The difference in position of the inflorescence or the fruit could account for the variation in the capsule length (Buide, 2008). From the results it indicates that *Gitu* fruit was far from the stalk, while *Kira Gitune*-1 and *Kira Kieru*-1 fruit was near the stalk.

The mean length of the wing was significantly different among the various cultivars. *Muguka*-1 cultivar from Embu had the highest and *Kira Gitune*-1 had the lowest. This could be explained by the possibility that the age of the plant could have had an impact on the length of the seed wing (Georg-kraemer et al., 2004). The one with high mean wing length could be older compared to the one with short wing length. From the results this could mean *Muguka*-1 cultivar was older than *Kira Gitune*-1 cultivar.

Principal Component Analysis (PCA) is a method that summarizes the data without much loss of information based on the similarities and the differences of the data. The PCA results of 13 traits among 30 khat samples indicated that the first principal component had a higher contribution to the total variation compared to the second and third principal components. The traits which contributed the highest variability value in PC1 included leaf margin, petiole length, peduncle length, sepal length, petal length, diameter of stamen, diameter of filament and ovary length (Table 1). This showed that these traits were responsible for the most of the diversity exhibited in the first principal component. The cumulative value of the first three principal components (58.7%) was slightly above the first principal component obtained by Mawia et al. (2015) (53.8%) in 13 rice genotypes. The principal component two showed variability of six traits while principal component three showed variability of four traits (Table 1). Leaf margin, petiole length and peduncle length contributed to variation across all the three principal components, indicating that they were most important agronomic traits in *C. edulis*. The first Eigen value (3.247) was close to that found by Asudi et al. (2010) who reported a value of 3.147 on phenotypic features of Kenyan papaya. The second Eigen value of 2.386 was lower than that obtained by Mawia et al. (2015) in 13 rice genotypes. Conversely, the third Eigen value of 1.995 was higher than that obtained by Mawia et al. (2015) (0.38) in 13 rice genotypes.

The distribution of the cultivars on the scatter plot indicated the existence of wide variations among the studied cultivars. Cultivars that were close to each other on the same scatter plot showed phenotypic relatedness, while cultivars that were far away from each other were regarded as phenotypically distant. For example, the graphical closeness of *Mugumo*-5 and *Muguka*-3 indicated morphological relatedness. Most of the cultivars, from the same quadrant were collected from the same geographical region. For example *Mugumo*-5, *Mugumo*-2, *Mugiza*-2 and *Muguka*-3 were found in the same quadrant and were collected from the same geographical region. *Gitune*, *Muceke* and *Mumbu* were from the same county. However, cultivars from different geographical regions were found clustering together for instance *Kirakiiru* from Meru clustered together with *Muguka*-1*, Mutimutiri and Muguka*-4 which were cultivars from Embu county, while *Mugumo*-4,*Gitu,Mugumo*-1,*Muruti, MugukawaKarimi* clustered together with *Kira Gitune*-2, *Kira Kieru*-1 which were cultivars from Meru county.

The dendrogram generated from 13 phenotypic traits showed two main clusters (clusters I and II), which indicated an existing variation in the cultivars. Cluster II showed high phenotypic variation, indicating the enormous phylogenetic divergence among cultivars in this cluster. Cluster II had two sub-clusters, IIa and IIb each of these sub-cluster, clustered into two subcluster again, IIai, IIaii and IIbi and IIbii. Cultivars in these cluster I included *Kira Kieru*-3, *Mugukawakarimi* and *Muguka*-4 cultivars where 2 cultivars were from Embu and 1 from Meru County. Most of the cultivars clustered in cluster II, some of the cultivars included *Kira Kiiru*-2 and *Kira Kieru*-2 which

were collected from Meru in cluster IIai, *Muguka*-2*, Mumbu, Mugumo*-2*,* and *Mugumo*-3 cultivars from Embu was found in IIbi. In these findings, those cultivars which clustered together were collected from the different geographical location. For the scatter plot, for example *Kira Kieru*-2 and *Kira Kiiru*-2 cultivars which were on the same scatter plot showed little variation in dendrogram clustering. Further studies are recommended due to the limitations associated with morphological traits as a means of determining variations, the diversity of these khat cultivars should be validated using superior traits such molecular markers.

Conclusions

From this study, it was concluded that phenotypic diversity showed considerable variability; clustering of the phenotypes was not based on the geographical origin of the plant.

CONFLICT OF INTERESTS

The authors have not declares any conflict of interests in phenotypic variations in *Catha edulis* along different growth intervals should be determined.

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Appendix 1. The name of the cultivar, place of collection and the type.

Code	Local name	County	Sub-County	Ward	Type habit
1	*Kira kieru* 1	Meru	Igembe South	Maua	Tree
2	*Kira kieru* 2	Meru	Igembe Central	Kangeta	Tree
3	*Kira kieru* 3	Meru	Igembe West	Kianjai	Tree
4	*Kira kiiru* 1	Meru	Igembe South	Maua	Tree
5	*Kira kiiru* 2	Meru	Igembe Central	Kangeta	Tree
6	*Kira gitune* 1	Meru	Igembe South	Maua	Tree
7	*Kira gitune* 2	Meru	Igembe South	Gaiti	Tree
8	*Muchuri*	Meru	Tigania West	Kianjai	Tree
9	*Kithara*	Meru	Tigania east	Muthaara	Tree
10	*Mutimutiri*	Embu	Mbeere South	Mavuria	Shrub
11	*Mugiza*1	Embu	Runynjes	Kagaari south	Shrub
12	*Mugiza* 2	Embu	Manyatta	Kithimu	Shrub
13	*Mugumo* 1	Embu	Mbeere South	Mavuria	Shrub
14	*Mugumo* 2	Embu	Mbeere North	Nthawa	Shrub
15	*Mugumo* 3	Embu	Mbeere South	Mbeti south	Shrub
16	*Mugumo* 4	Embu	Manyatta	Kithimu	Shrub
17	*Mugumo* 5	Embu	Runyenjes	Kaagari north	Shrub
18	*Muguka* 1	Embu	Mbeere South	Mavuria	Shrub
19	*Muguka* 2	Embu	Mbeere North	Muminji	Shrub
20	*Muguka* 3	Embu	Mbeere North	Nthawa	Shrub
21	*Muguka* 4	Embu	Manyatta	Mbeti north	Shrub
22	*Muguka* 5	Embu	Runyenjes	Kaagaari north	Shrub
23	*Muguka wa karimi*	Embu	Mbeere South	Mavuria	Shrub
24	*Gitu*	Embu	Mberee South	Mbeti south	Shrub
25	*Mutamucii*	Embu	Mbeere South	Mavuria	Shrub
26	*Mukurukuru*	Embu	Mbeere North	Nthawa	Shrub
27	*Muruti*	Embu	Mbeere North	Muminji	Shrub
28	*Muceke*	Embu	Mbeere South	Mavuria	Shrub
29	*Gitune*	Embu	Mbeere South	Mbeti south	Shrub
30	*Mumbu*	Embu	Mbeere North	Muminji	Shrub

Appendix 2. Phenotypic traits of 30 khat cultivars.

Genotype	Leaf length	Leaf width	Leaf margin	Petiole length	Inflorescence length	Peduncle length
Gitu	74.00±1.15^{ef}	34.00±0.58^{edfg}	92.00±1.35^{c}	2.67±0.33^{cd}	20.00±0.57^{ab}	5.33±0.33^{abcd}
Gitune	73.00±0.58^{efgh}	37.33±0.88^{cde}	71.33±1.20^{ghi}	4.33±0.33^{abc}	15.33±0.67^{ef}	4.67±0.33^{bcd}
Muguka 2	67.33±1.45^{fghi}	30.67±0.67^{gh}	63.67±0.88^{jk}	5.67±0.33^{a}	20.00±0.58^{ab}	5.33±0.33^{abcd}
Kira gitune 1	60.00±1.00^{jkl}	26.33±0.88^{ijk}	72.67±1.45^{gh}	4.33±0.33^{abc}	15.00±0.58^{f}	7.33±0.08^{a}
Kira gitune 2	75.33±1.45^{de}	21.33±0.88^{lm}	82.00±1.53^{de}	4.33±0.33^{abc}	19.67±0.33^{abc}	7.00±0.29^{ab}
Kira kieru 1	72.00±1.53^{efgh}	19.33±0.67^{m}	74.67±1.20^{fg}	4.67±0.33^{ab}	21.00±0.58^{a}	6.00±0.58^{abcd}
Kira kieru 3	94.33±1.76^{a}	30.33±0.88^{ghi}	99.00±0.58^{b}	4.67±0.33^{ab}	14.68±0.88^{f}	6.00±0.58^{abcd}
Mugumo 4	72.33±1.67^{efgh}	24.00±0.58^{kl}	80.33±0.88^{ef}	4.33±0.33^{abc}	20.33±0.33^{a}	6.00±0.58^{abcd}
Kira kieru 2	73.67±1.86^{ef}	30.00±0.57^{ghi}	87.67±1.45^{cd}	4.67±0.33^{ab}	17.00±0.58^{bcdef}	5.33±0.33^{abcd}
Kira kiiru 1	73.33±1.45^{efg}	35.33±0.67^{def}	71.67±0.88^{ghi}	4.67±0.33^{ab}	17.00±1.15^{bcdef}	5.33±0.33^{abcd}
Kira kiiru 2	71.33±0.88^{efgh}	24.00±1.00^{kl}	87.33±1.20^{cd}	4.33±0.33^{abc}	19.67±0.33^{abc}	6.83±0.44^{ab}
Kiithara	82.00±1.15^{cd}	23.00±1.15^{klm}	88.00±1.15^{cd}	4.67±0.33^{ab}	16.33±0.88^{def}	6.67±0.88^{abc}
*Muchur*i	56.00±1.15^{kl}	25.00±0.58^{jkl}	72.33±0.88^{ghi}	4.67±0.33^{ab}	16.67±0.88^{def}	6.33±0.33^{abcd}
Muguka 5	91.67±0.88^{b}	41.68±0.88^{ab}	87.67±1.20^{cd}	3.33±0.33^{bcd}	20.67±0.33^{a}	4.33±0.33^{cd}
Muguka 4	99.00±0.58^{a}	45.67±0.33^{a}	107.33±1.45^{a}	4.33±0.33^{abc}	19.33±0.33^{abc}	4.67±0.33^{bcd}

Appendix 2. Contd.

Mugumo 5	72.33±1.20^{efgh}	30.67±0.67^{gh}	71.67±0.88^{ghi}	5.00±0.57^{ab}	19.67±0.33^{abc}	4.67±0.33^{bcd}
Muguka 1	73.00±1.73^{efg}	33.67±0.88^{efg}	73.00±1.00^{gh}	5.67±0.33^{a}	16.67±0.88^{cdef}	4.33±0.33^{cd}
Muguka 3	72.00±1.53^{efgh}	32.00±1.15^{fgh}	82.33±1.45^{de}	2.33±0.33^{d}	20.67±0.67^{a}	4.33±0.33^{cd}
Muceke	70.00±0.58^{efgh}	40.00±0.58^{bc}	69.33±0.88^{ghij}	5.33±0.33^{a}	15.33±0.88^{ef}	4.67±0.3^{bcd} 3
Mugiza 2	73.33±1.76^{efg}	38.00±0.58^{bcd}	74.33±0.88^{fg}	4.33±0.33^{abc}	16.67±0.88^{cdef}	4.67±0.33^{bcd}
Muguka wa karimi	81.00±1.00^{d}	31.33±0.88^{fgh}	102.67±1.20^{ab}	4.67±0.33^{ab}	19.33±0.33^{abcd}	4.67±0.33^{bcd}
Mugiza 1	70.67±1.20^{efgh}	41.67±0.88^{ab}	73.00±0.88^{gh}	2.33±0.33^{d}	19.67±0.33^{abc}	4.00±0.58^{d}
Mugumo 1	88.67±1.20^{bc}	41.67±0.88^{bc}	102.33±1.20^{ab}	3.33±0.33^{bcd}	18.33±0.33^{abcde}	4.00±0.58^{d}
Mugumo 2	61.33±0.88^{hij}	28.67±0.88^{ijk}	63.33±0.88^{jk}	3.33±0.33^{bcd}	20.67±0.33^{a}	4.00±0.58^{d}
Mugumo 3	62.33±0.33^{ijk}	33.67±0.33^{efg}	61.67±0.33^{jk}	4.33±0.33^{abc}	15.67±0.88^{ef}	5.00±0.58^{abcd}
Mukurukuru	70.00±1.15^{efgh}	38.00±0.58^{bcd}	75.33±0.33^{fg}	4.33±0.33^{abc}	15.33±0.33^{ef}	4.67±0.33^{bcd}
Mumbu	66.33±0.33^{hij}	28.33±0.88^{hij}	66.33±0.88^{ijk}	4.33±0.33^{abc}	20.33±0.33^{a}	4.67±0.33^{bcd}
Muruti	66.67±0.88^{ghij}	33.00±0.58^{fg}	65.00±1.00^{jk}	4.33±0.33^{bcd}	19.67±0.33^{abc}	4.67±0.33^{bcd}
Mutamucii	73.33±1.76^{efg}	33.00±0.58^{fg}	68.00±0.58^{hij}	4.33±0.33^{abc}	16.33±0.33^{def}	4.67±0.33^{bcd}
Mutimutiri	53.67±1.20^{l}	21.33±0.33^{lm}	55.00±0.58^{l}	2.67±0.33^{cd}	14.33±0.33^{f}	4.33±0.33^{cd}

Values are expressed as Mean±SEM. Values with the same superscript letter are not significantly different (one way ANOVA followed by Tukey's test ($p>0.05$).