Original Research Papers

Morphological traits as indicators of bitterness in traditional vegetables; the case of spider plant (*Cleome gynandra*) in Kenya

ABSTRACT

Aims: The study set to find out existence of variation in leaf bitterness of spider plant in six (6) agro ecological zones in Kenya and whether leaf bitterness levels correlated with morphological characters; colour of main stem including colour of leaf blade, stem pubescence and leaf waxiness.

Study design: Morphological characterization was done using purposive sampling method while organoleptic characterization was done using 'within participant's' method.

Place and duration of study: Agro-ecological zones; upper highlands, lower highlands, upper midlands, lower midlands, inland lowlands and coastal lowlands sampled between 2016-2017.

Methodology: Morphological characterization for *Cleome gynandra* was done using IPGRI descriptors in 18 sites representing agro-ecological zones. Mature healthy seeds of spider plant were collected from the accessions characterized and grown at Nairobi Botanic Garden using a randomized block design. Organoleptic testing was done on 40 spider plant accessions which grew to maturity. Qualitative analysis performed on four (4) qualitative traits; colour of main stem, stem pubescence, leaf waxiness and colour of leaf blade was correlated to levels of bitterness using Pearson correlational analysis. Leaf bitterness and colour of leaf blade showed significant variation in six agro-ecological zones. Leaf bitterness levels were high in coastal lowlands and lower midlands and low in highlands. Colour of leaf blade strongly associated with leaf bitterness while others correlated weakly. *Cleome gynandra* specimens were grouped into two (2) clusters which were further divided into eight (8) clusters in the dendogram based on level of leaf bitterness.

Changes in agro ecology had a significant effect on level of bitterness and colour of leaf blade was a strong indicator of level of leaf bitterness of spider plant. Colour of leaf blade was recommended for distinguishing non-bitter types from bitter types of spider plant.

Key words; spider plant, variation, morphology, leaf bitterness

1. INTRODUCTION

Spider plant (*Cleome gynandra*) belongs to the plant family Cleomaceae [7], is an erect herb 1 m high with hairy stems and sometimes waxy. Its leaves are petiolate usually divided into 3,5 and 7 leaflets. Petals are white or purple and the fruit is a capsule with greyish black seeds. It is widely distributed all over Kenya as a weed of cultivation and disturbed areas from 0-2400 m. It grows on soils rich in organic matter and is especially found in animal enclosures and abandoned homesteads and thrives during warm season.

The vegetable is among the list of important traditional African leafy vegetables used as a relish in Kenyan households [15,16] and other African countries whose market demand is on the rise [17] due to it rich nutritional and medicinal value. It is commonly referred to as spider plant, spider flower in English, mwangani in Swahili or by its trade name 'saga' or 'managu' in Kenya [5]. Spider plant follows the C4 photosynthesis pathway which enhances its ability to thrive in semi-arid areas and hencea key vegetable available during relish-gap period and therefore plays an important role in household food security during lean times [22]. It is relished for its taste and perceived medicinal value by various communities in Kenya such as the Luo, Luhya, Kisii, Kipsigis,Mijikenda among others. [15]. In Kenya, managu or spider plant vegetable is regarded as a powerful 'medicinal food' or nutraceutical consumed by recuperating individuals such as pregnant and lactating mothers, circumcised boys and invalids [21]. Spider plant stimulates the restoration of blood after delivery by increasing the number of red blood cells and the corpuscular hemoglobin concentration and also by stimulating the synthesis of iron biomarkers such as transferrin and ferritin in the body [3].

Nutritional studies have shown that it is rich in micronutrients such as calcium, magnesium, iron, zinc, vitamin A, C and E, hence suitable for combating hidden hunger and management of lifestyle diseases

[24]. It is an ingredient in a baby's weaning food where leaves are crushed and added to a meal to increase its nutritional value in Nigeria [9]. It is also reported that spider plant is a good source of vitamin A and Iron providing 50–75% RDA for children [26].

An infusion of the leaves is used to treat anaemia and as an eye wash [5]. In Kenya, fresh leaves are sold domestically in formal and informal markets whose price ranges from 0.40-0.50 USD/Kg when in plenty during wet season and sells twice as much in dry season and hence contributes 15-40% of the total income of small scale farmers in Kenya [23].

Cleome gynandra is a herb [2], quite variable in its range [5] because it is adapted to a wide agro ecological range [14]. Variation has been reported in stem colour, stem pubescence, large or small leaves, flower colour, shape of pods, range in bitterness among others [11, 21]. The bitter taste in spider plant is said to vary from slightly bitter to extreme bitterness [21]. Various cooking techniques are applied to reduce the bitter taste [11, 15]. Spider plant is referred to as *bilolo* which means bitter in Lingala language due to its bitter taste [14]. Some communities in Kenya (western and coast) and West Africa appreciate the bitter taste as it is said to be appetizing and also 'good for the stomach' [21]. The bitter taste of spider plant deters some people from consumption such as the youth, children and certain community groups in Kenya because it is considered unpalatable. Spider plant contains condensed tannins that cause the bitter taste [11]. More so the concentration of condensed tannins varies within *Cleome gynandra* genotypes and that bitterness is amenable by breeding by reducing concentration of condensed tannins [11]. Condensed tannins are involved in biochemical plant defense mechanisms with the variation arising from evolutionary history in terms of pathogen/pest pressure in areas of origin. These adaptations are genetically controlled and highly heritable [8].

Indigenous leafy vegetables such as spider plant are cooked traditionally by first boiling to eliminate unwanted non-nutrient bio-active compounds [1]. Extreme bitter types of spider plant are cooked for long periods (1-2 hours) with several water changes to reduce leaf bitterness which leads to extreme losses of thermo-labile vitamins beta carotene, vitamin C and other useful medicinal compounds [1,12]. Other spider plant preparation methods include boiling and fermenting for two (2) days with addition of fresh

milk or coconut milk to neutralize the bitter taste [15]. It is also cooked mixed with other vegetables such as amaranth or Ethiopian kale to neutralize the bitter taste. Local communities in Kenya and Tanzania have developed knowledge on how to select spider plant types which are not bitter by use of morphological traits such as stem colour or colour of petiole [21]. Green colored types are preferred to purple ones as they are reported to be less bitter and more tender [21]. The Duruma of coastal Kenya distinguish two (2) types of spider plant; light green (*changani cheruhe*) and dark green coloured leaves (*changani chiru*) where the former light green leaved types are considered to be more bitter than the latter [13]. This knowledge on selection of bitter and non-bitter types has not been verified scientifically.

A number of past research studies have characterized *Cleome gynandra* based on different criteria. It has been demonstrated that there is distinct genetic variation where the five (5) identified *Cleome gynandra* genotypes being tested for levels of bitterness and genetic variability on levels of condensed tannins [11]. On the basis of the above tannin test, two (2) genotypes were recommended for direct use because they had low levels of condensed tannins, hence were less bitter, required less amount of cooking time and therefore the thermo-labile vitamins were preserved. They [11]. This variation was explained as a result of evolutionary history in terms of pathogen/pest pressure in areas of origin of these genotypes. Such evolutionary adaptations are often genetically controlled, highly heritable and amenable to breeding [11].

The objective of this study was to evaluate existence of variation in leaf bitterness in spider plant and determine whether bitterness has a relationship with morphological characters; colour of leaf blade, colour of main stem, stem pubescence and leaf waxiness in six (6) agro ecological zones.

2. MATERIALS AND METHODS

2.1 Morphological characterization

Purposive method of sampling was used to sample only those sites such as the wild and farms which had spider plant growing naturally. The sites sampled are shown in figure 1 which occur within six agro ecological zones which are upper highlands, lower highlands, upper midlands, lower midlands, inland lowlands and coastal lowlands. The sites samples were Baringo, Bungoma, Elgeyo Marakwet, Garissa,

Homabay, Kakamega, Kericho, Kilifi, Kitui, Kisii, Kwale, Makueni, Nakuru, Taita, Trans Nzoia, Uasin Gishu, West Pokot and Nyeri (Table 1).

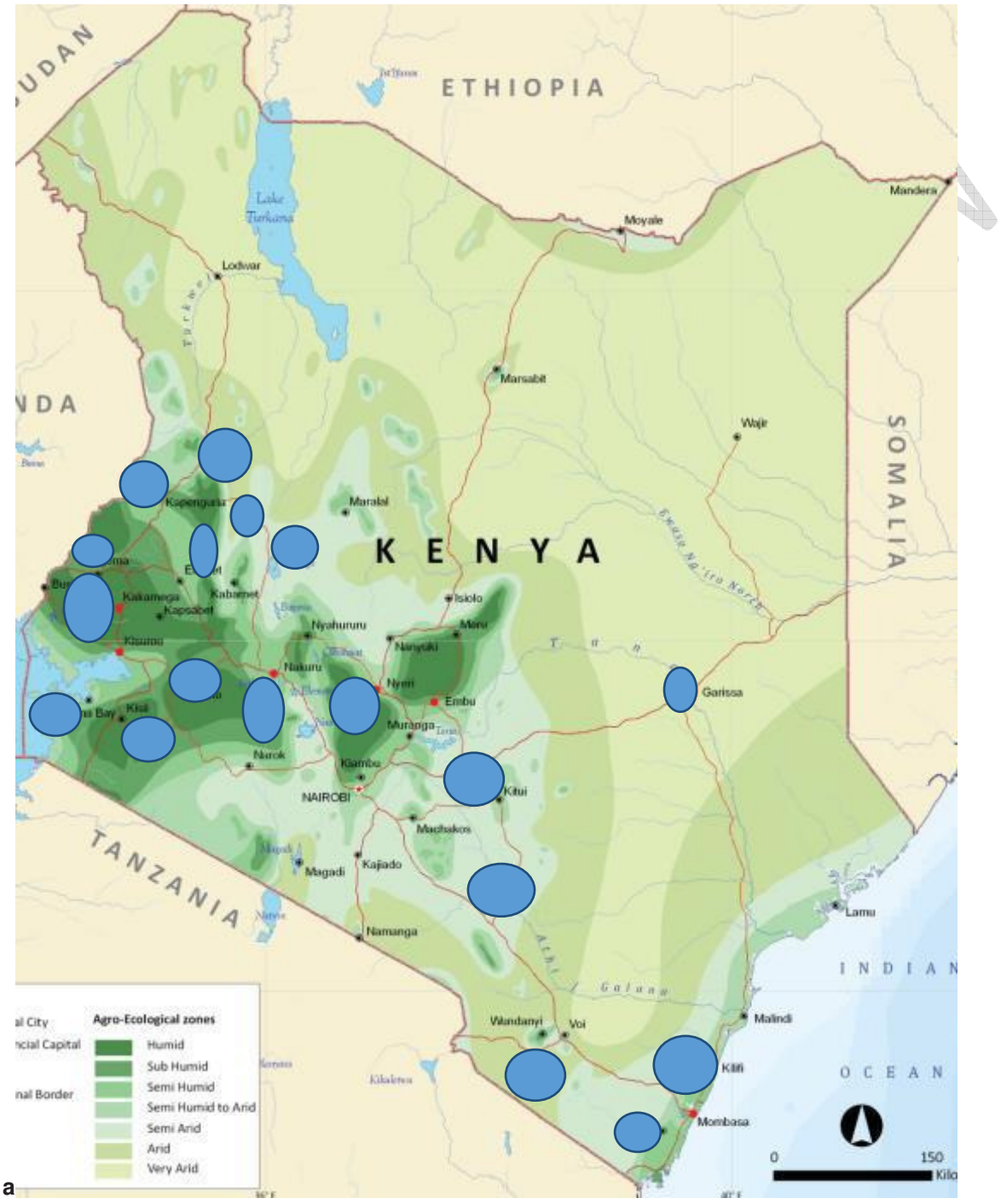

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Figure 1: Map of collection sites for *Cleome gynandra*

 -denotes collection sites

Table 1: Collection sites for *Cleome gynandra*

Accessions	Area	Lat/Long	Elevation (m)	Soils	Temp (℃)	Annual Rainfall (mm)	Humidity (%)	AEZ
BAR 1807B	Baringo	N00.46544 E036.01440	1012	Clay soil	21.9	684	56	IL
BAR 1807A	Baringo	N00.46777 E036.01886	1013	Loam soil	21.9	684	56	IL
BGM 2107A	Bungoma	N00.76367 E034.72483	1713	Loam soil	21.1	1628	62	UM
BGM 2107C	Bungoma	N00.61241 E034.61735	1506	Loam soil	21.1	1628	62	UM
ELG/1907B	Elgeyo Marakwet	N00.46728 E035.60924	1247	Rocky soil	14-24	400-1400	70	UM
ELG/1907C	Elgeyo Marakwet	N00.46728 E035.60924	1253	Rocky loam soil	14-24	400-1400	70	UM
ELG/1907A	Elgeyo Marakwet	N00.46728 E035.60924	1247	Rocky soil	14-24	400-1400	70	UM
GA-01	Garissa	S00 28.292 E039 38.193	1035	Clay	29.3	362	35	IL
HBY 2307A	Homabay	S00.54397 E034.28947	1155	Black cotton soil	22.5	1226	65	LM
HBY 2307B	Homabay	S00.54397 E034.28947	1155	Black cotton	22.5	1226	65	LM

				soil				
KK 2207	Kakamega	N00.23009 E034.83690	1568	Loam soil	20.4	1971	64	UM
KRC 2507B	Kericho	S00.31112 E035.28067	2063	Rocky soil	18.1	1735	69	LH
KRC 2507A	Kericho	S00.58607 E035.19402	1948	Loam	18.1	1735	69	LH
KF-11	Kilifi	S03 32.364 E039 31.812	282	Sandy loam	26	1063	56	CL
KF-05A	Kilifi	S03.2654 E40.01466	50	Sandy clay	26	1063	56	CL
KF-01	Kilifi	S03 03.239 E040 08.197	10	Clay	26	1063	56	CL
KF-05	Kilifi	S03.2653 E40.01468	50	Sandy clay	26	1063	56	CL
KF-09	Kilifi	S03 33.609 E039 30.362	236	Sand	26	1063	56	CL
KF-07	Kilifi	S03 35.569 E039 51.324	29	Clay	26	1063	56	CL
KIS/2407A	Kisii	S00.88754 E034.79023	1787	Loam	19.6	1922	67	UM
K1-01	Kitui	S 01 12.774 E037 50.763	1240	Alluvial sandy	21.4	1068	35	LM
K1-02	Kitui	S01 12.775 E037 50.772	1240	Clay with little sand	21.4	1068	35	LM
KW-02	Kwale	S04.29055 E039.56532	20	Clay rich in	23.5	1118	47	CL

				organic matter from goats				
MK-01	Makueni	S02 33.747 E03800.610	920	Clay	21.5	834	29	LM
MK-02	Makueni	S02 33.246 E038 00.390	925	Clay	21.5	834	29	LM
NKR 1807	Nakuru	N00.01964 E036.22417	2016	Loam	17.5	895	69	UH
KIS/2407B	Kisii	S01.00975 E034.88179	1756	Black cotton	19.6	1922	67	UH
TT-01	Taita	S03 28.678 E038 22.791	864	Sand	23	754	35	UM
TT-02	Taita	S03 28.678 E038 22.791	867	Sand	23	754	35	UM
TT-00	Taita	S03 47.263 E039 22.842	188	Sandy clay	25	650	36	LM
TNZ 2107B	Trans Nzoia	N00.89914 E034.97070	1791	Loam	18.3	1097	76	UM
UAG 1907C	Uasin Gishu	N00.62237 E035.43269	2252	Clay	16.8	995	73	LH
UAG 1907A	Uasin Gishu	N00.62237 E035.43269	2252	Clay	16.8	995	73	LH
UAG 1907B	Uasin Gishu	N00.62237 E035.43269	2252	Clay	16.8	995	73	LH
WPK 2007A	West Pokot	N00.01309 E035.20480	1722	Black	23.6	623	90	LM

				cotton soil				
WPK 2007D	West Pokot	N00.01309 E035.20480	1722	Black cotton soil	23.6	623	90	LM
WPK 2007E	Kapenguria	N01.23054 E035.11934	2016	Black cotton soil	16.9	1140	76	LH
NYR 2107C	Nyeri	S00.33867 E036.85818	2016	Loam soil	17.1	1004	73	UH
NYR2107B	Nyeri	S00.33867 E036.85818	2016	Loam soil	17.1	1004	73	UH
NYR2107A	Nyeri	S00.33867 E036.85818	2016	Loam soil	17.1	1004	73	UH

UH (Upper Highland), UM (Upper Midland), LH (Lower Highland), LM (Lower Midland), IL (Inland Lowland), CL (Coastal Lowland)

An IPGRI descriptor based on FAO standards was used to evaluate morphological qualitative characters across six (6) agro ecological zones as shown in table 2. A total of four (4) qualitative traits were evaluated for morphological variation were; colour of main stem, stem pubescence, colour of leaf blade and leaf waxiness. Colour of main stem, stem pubescence and colour of leaf blade were observational traits evaluated by using eyes. On the other hand, leaf waxiness and leaf bitterness were sensory traits evaluated using hands (feeling) and taste respectively.

Table 2: Descriptor codes for qualitative traits of spider plant accessions

Character	Descriptor and code
Colour of the main stem	Green (1), green tinged purple (2), purple tinged green (3), light purple (4), purple (5), dark purple (6), mixed (7)
Stem pubescence	Slightly hairy (1), hairy (2), very hairy (3), wooly (4)

Colour of leaf blade	Light green (1), green (2), dark green (3)
Raw leaf bitterness	Not bitter (1), mild (2), bitter (3), very bitter (4), extremely bitter (5)
Leaf waxiness	Not waxy (1), fairly waxy (2), waxy (3), very waxy (4)

2.2. Propagation of spider plant

Mature healthy seeds of spider plant were harvested from accessions in which morphological variation had been done and were grown in the field at the Nairobi Botanic Garden. Nairobi Botanic Garden is located in Nairobi, Kenya on latitude -1.274469 and long 36.813941 and falls under upper midland zone (33). Nairobi has a bimodal distribution of rainfall with long rains starting early March to late May and short rains from October to December with an average rainfall of 1000 mm [34]. Mean annual temperatures are 13℃ -23℃ [33]. A pre-germination test was first conducted to ensure that spider plant seeds were viable. 56 accessions were planted in the field garden in December 2017, arranged in a randomized complete block design with three (3) replications. The site (land) had no shading as spider plant does not tolerate shaded condition as this hampers growth. Land was prepared by ploughing using a jembe by hand and making plots or beds. Planting rows were made in each plot, and 10 seeding holes were made in each row. Inter row spacing was 30 cm and intra row spacing of 30 cm. Each plot had a total of 20 plants per plot. Each accession was planted in each row with three (3) replicates. A single seed was placed in each hole 5 cm deep and covered with soil but not compacted. The garden was kept weed free by hand weeding. No fertilizer or pesticides were used in growing the spider plants. The experiment was conducted under rain-fed conditions though supplemental overhead irrigation was applied two times, at two weeks after planting and two weeks after flower initiation. The spider plant leaves were ready for harvesting at 50% flowering to conduct the organoleptic test.

2.3. Organoleptic characterization

Organoleptic testing of leaf bitterness in spider plant accessions was done using a 'within participants' design' where all the testing participants were given the same raw spider plant leaves accessions to evaluate taste by chewing in the mouth and indicate level of bitterness. Spider plant leaves were ready for harvesting after two (2) months at 50% flowering to conduct the organoleptic test. Organoleptic test was done on 40 accessions because the other 16 accessions failed to germinate. Fresh leaves of spider

plant were harvested from each accession and used for organoleptic testing for evaluation of differences in levels of bitterness which ranged from 1-5. The raw leaves were tested for level of bitterness by chewing in the mouth.

Leaves from three (3) plants in each accession were randomly collected, put in a clean container and labeled. Only a handful of the leaves was collected. The three selected plants were tagged for evaluation.

A Panel of 10 people; five (5) men and five (5) women aged 20-55 years were selected based history of consuming the vegetable and those who did not consume it. Each person collected the leaves from each accession, then cleaned it in water and tasted. The level of bitterness was recorded, then the tester rinsed mouth and waited for five (5) minutes before tasting the next accession. The bitter taste of the leaves was ranked from 1 (not bitter) to 5 (very bitter) (Table 3).

Table 3: Range of leaf bitterness levels based on based on the perception of the ten selected tasters

	Taste description	Value
1.	Not bitter	1
2.	Mild	2
3.	Bitter	3
4.	Very bitter	4
5.	Extremely bitter	5

The Data was entered in an excel file; then mode and mean were conducted on the results of each accessions.

2.4Data analysis

Data collected in this study was analyzed using qualitative approach to discuss morphological characterization data collected in the field and garden and organoleptic characterization data. Descriptive statistics were used to explain qualitative approaches whereby percentages of each trait in each agro ecological zones were recorded. One-way ANOVA was used to evaluate significant differences of leaf

bitterness, colour of leaf blade, leaf waxiness, colour of the main stem and stem pubescence in six (agro ecological zones) and a post hoc analysis to determine least significance between the variables.

A correlation analysis was done using Pearson correlation chi-square method to estimate relationship between levels of bitterness (dependent variable) and colour of leaf blade, leaf waxiness, colour of the main stem and stem pubescence (predictors).

Hierarchical clustering analysis based on levels of leaf bitterness was performed using SPSS Software version 21. UPGMA (unweighted pair-group method using averages) was used which distributes the accessions into a reasonable number of groups. It calculates differences between clusters as the average of all the point-to-point distances between a point in one cluster and a point in the other. The clusters and relationships were displayed on a dendogram. In cluster analysis, two (2) most similar accessions will be clustered together in a group and similarities of this group calculated. Two closest groups are combined until a single group remains. The results were expressed in a dendogram or a 2-dimensional hierarchical tree diagram which represent multivariate relationships among accessions

3. RESULTS AND DISCUSSION

3.1 Propagation of spider plant

A total of 56 accessions of spider plant seeds were collected from the field mission as shown in table 2 in the methodology. Of the 56 accessions, only 40 accessions (71.4%) germinated and grew to maturity. Accessions that did not grow included; KF-02, KF-03, KF-04A, KF-04B, KF-06, KF-08, KF-10, KF-12, KW-01, KF-13, KIR-1707, WPK-2007B, WPK-2007C, BGM-2107B, KIS-2407A, KRC-2507C.

Lack of germination for the 16 accessions could be attributed to the fact that spider plant seeds experience seed dormancy (27) which is broken after storage for 6 months. In this case, the seeds were planted after two (2) months of harvesting which may have led to germination failure.

The seeds may not have been also mature enough when harvested due to variation in fruit set and development in different accessions [24,10]. The prevailing agro ecology may not have been favourable for them to grow especially for accessions from coastal lowlands of Kwale and Kilifi.

3.2 Variation in qualitative traits

Analysis of variance indicated that leaf bitterness and colour of lead blade varied significantly across the agro ecological zones at p<0.005 (see table 4). However, leaf waxiness, stem pubescence and colour of main stem did not show significant variation in six (6) agro ecological zones.

Table:4 Analysis of variance for level of significance in leaf colour of main stem, stem pubescence, colour of leaf blade, leaf bitterness and leaf waxiness

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
avc_Leaf bitterness	Between Groups	9.122	5	1.824	3.453	.012
	Within Groups	17.962	34	.528		
	Total	27.084	39			
Leaf waxiness	Between Groups	7.007	5	1.401	1.050	.405
	Within Groups	45.393	34	1.335		
	Total	52.400	39			
Stem hairiness	Between Groups	2.896	5	.579	.737	.601
	Within Groups	26.704	34	.785		
	Total	29.600	39			
Colour of the main stem	Between Groups	15.471	5	3.094	.900	.492
	Within Groups	116.904	34	3.438		
	Total	132.375	39			
Colour of leaf blade	Between Groups	14.580	5	2.916	12.720	.000
	Within Groups	7.795	34	.229		
	Total	22.375	39			

3.2.1 Variation in leaf bitterness

Leaf bitterness varied significantly across the agro ecological zones (see table 4) with significant differences noted in upper midlands, lower midlands and coastal lowlands. Upper highlands, lower

highlands and inland lowlands showed least significant differences in leaf bitterness. Variation in bitterness levels in spider plant accessions is attributed to difference in concentration of condensed tannins due to genetic variability in quantity of condensed tannins as bitter taste increases with increase in quantity of condensed tannins as earlier shown by Kutsukutsa's study [11]. Accessions from coastal lowlands and lower midlands exhibited the highest bitterness levels followed by those from upper midlands (see table 5). On the other hand, accessions from upper highlands, lower highlands and inland lowlands had the lowest bitterness levels. Bitterness levels in lower highlands ranged from 1.7-2.4 (mild taste) while upper highlands were 1.0-2.4 (not bitter to mild in taste). In coastal lowlands, bitterness level ranged from 1.4-4.0 (not bitter to very bitter taste) while those in lower midlands ranged from 1.6-4.2 (mild to very bitter taste). Inland lowland accessions were considered as mild to bitter in taste as the bitterness level range was 2.0-3.5. Accessions with the highest level of bitterness were KF-05, KI-01, KW-02, GA-01 and TT-01 which were termed as very bitter to extremely bitter (4-5) while the accessions which had the lowest level of bitterness were UAG1907A, UAG1907B, UAG1907C, KRC2507A, HBY2307B and KF-07 which were termed as not bitter to mild (1-2).

Spider plant is highly commercialized in the lower highlands areas where a lot of selection work is done to achieve the preferential criteria of the consumers and producers [3]. The producers in the lower highlands provide spider plant for the large market town centers in the region and thus tend to select and propagate spider plant types with low or no bitterness. On the other hand, spider plant types in the coastal lowlands were mostly bitter. This may be due to the effects of high temperatures occurring in the lowlands which cause increase in concentration of condensed tannins making the plants bitter, a strategy which protects the plant against herbivores and pests for survival purposes [30]. Local communities in coastal lowlands have a rich culture of consuming wild vegetables. the spider plant included [13]. They conserve both bitter and non-bitter landraces of spider plant but are accustomed to and appreciate the flavor of the bitter landraces which they associate with high medicinal benefits [6]. They also have well-developed traditional vegetable recipes for preparing the bitter types which tone down the bitterness hence making the vegetable palatable [6]. Spider plant types from lower midlands had a wide range of bitterness probably because sites sampled in lower midlands had varying humidity levels e.g. Homabay is more humid than Kitui and therefore accessions are less bitter compared to those from Kitui [13]. The upper midlands'

accessions had bitterness which was highly varied from mild, bitter and very bitter (1.7-4.1) with very bitter types from Taita. This is attributed to humidity differences within the agro ecological zone e.g. Kakamega is more humid than Taita which causes differences in concentration of condensed tannins. Accessions KF-07 and GA-01 did not express bitterness levels like other accessions in their respective AEZ because they were cultivated types sourced from other regions probably through seed trade.

Table 5: Range in leaf bitterness in six (6) agro ecological zones based on the perception of the ten selected tasters (see table 3)

		Mode	Maximum	Minimum
Agro-ecological zones	Coastal lowlands	3.1	4.0	1.4
	Inland lowlands	2.1[a]	3.5	2.0
	Lower highlands	1.8	2.4	1.7
	Lower midlands	2.7	4.2	1.6
	Upper highlands	1.0	2.4	1.0
	Upper midlands	2.9, 2.0[a]	4.1	1.7

a. Multiple modes exist. The smallest value is shown

3.2.2. Colour of the main stem

Spider plant accessions varied in the colour of the main stem from purely green to dark purple but did not show significant variation in six (6) agro ecological zones (see table 4). Green coloured stems dominated the accessions with 30% purely green and 25% being green with a tinge of purple. Only 5% had dark purple stems while those with purple and pink (light purple) stems were both 12.5% of the accessions. Some populations had their main stems green and some purple (mixed) which made 5% of the accessions. Colour variation displayed in spider plant tissues such as stems varied from pink to purple is due to accumulation of plant pigment; anthocyanins in plant tissues which are environmentally controlled by factors such as stress, nutrients and temperature [10]. Anthocyanins are flavonoids responsible for plant-animal interactions which include attraction of pollinators and chemical repellence of herbivores and pests (plant defense mechanisms [25]. Anthocyanins also camouflage plant parts against their background protecting them from predation and protect plant tissues against UV light especially those

growing in the highlands [32]. Accumulation of anthocyanins in plant tissues (stems and petioles) therefore increases the ability of purple coloured spider plant to grow and thrive under diverse environmental conditions [10]. Anthocyanins being flavonoid in nature, have anti-bacterial, anti-viral, antifungal [25], anti-tumor, anti-oxidant [24] properties which are powerful health promoting compounds in the body when consumed [18].

3.2.3 Stem pubescence

The stems for the spider plant accessions ranged from being slightly hairy to being wooly (densely hairy) and did not show significant variation in six (6) agro ecological zones (see table 4). A total of 77.5% were mainly slightly hairy, 12.5% were hairy, 2.5% were very hairy and 7.5% were wooly.

Stem pubescence variation recorded in this study concurs with Chweya and Mnzava's findings which in addition also recorded glabrous stems [5]. Glabrous character was reported to be rare which concurs with this study as they were no glabrous stems found in the accessions. Stiff and irritating hairs on petioles and stems of plants protect plants against browsing by herbivorous animals and insects [19] hence very hairy spider plants are better protected to enhance chances of survival.

3.2.4 Colour of leaf blade

The study showed that colour of leaf blade varied from light green, green and dark green. Colour of leaf blade also showed significant variation across six (6) agro ecological zones (see table 4). A total of 42.5% of the accessions were green, 35% were dark green while 22.5% were light green in colour.Green colour was most common among the accessions. However, another study recorded brown coloured leaves which were not documented in the study [5]. Variation in colour of leaf blade has not been evaluated before in any other study. Colour of leaf blade was found to negatively correlate with bitterness, whereby bitterness levels decreased with increase in concentration of the green colour. This indicates that colour of leaf blade is a strong indicator of level of bitterness in spider plant.

3.2.5 Leaf waxiness

Leaf waxiness did not show significant variation in six (6) agro ecological zones (see table 4). Levels of leaf waxiness noted were not waxy, fairly waxy, waxy and very waxy. Almost half of the accessions (45%) were very waxy, 40% fairly waxy, 12.5% not waxy and the least were waxy (2.5%).

Most of the spider plant accessions were waxy and this trait insulates plant surfaces by preventing them against excessive loss of water caused by transpiration, protection against pest damage and air pollutants [20].

3.4 Correlation between qualitative characters and level of leaf bitterness

The four (4) predictors; colour of leaf blade, colour of main stem, leaf waxiness and stem pubescence combined together were strongly positively correlated at P=0.05 to leaf bitterness (0.001) as shown in table 6.

Table 6: Relationship between level of bitterness and combined four (4) morphological traits

ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	11.057	4	2.764	6.037	.001[b]
	Residual	16.027	35	.458		
	Total	27.084	39			

a. Dependent Variable: avc_Leaf bitterness

b. Predictors: (Constant), colour of leaf blade, leaf waxiness, stem pubescence, colour of the main stem

However, when the predictors were separated, colour of the main stem, stem pubescence, leaf waxiness showed moderately positive correlation with leaf bitterness as shown in table 7 but was not significant. Colour of the main stem, leaf waxiness and stem pubescence showed 0.102, 0.301 and 0.955 correlation respectively which though positive correlations were not significant. However, colour of leaf blade showed a strong negative correlation at P=0.05 with leaf bitterness (-0.000) (Table 6). This meant that bitterness levels increased as the leaf green colour became lighter (light green) and decreased as leaf green colour became darker. However, GA-01 had dark green leaves yet was bitter probably because it was a cultivated type.

Various reports indicate that farmers determine or distinguish bitter types from non-bitter types of spider plant using colour of the stems [21,24]. This does not concur with this study because colour of the main stem alone did not correlate with bitterness levels in *Cleome gynandra*. However, this study found out that only colour of leaf blade correlated negatively with bitterness levels indicating that as the leaf's green colour decreased the bitterness levels increased.

Table 7: Relationship between level of bitterness and four (4) morphological traits

Coefficients[a]

Model		**Unstandardized Coefficients**		**Standardized Coefficients**	**t**	**Sig.**
		B	Std. Error	Beta		
1	(Constant)	3.141	.440		7.143	.000
	Leaf waxiness	.108	.103	.150	1.050	.301
	Colour of the main stem	.105	.063	.233	1.681	.102
	Stem pubescence	.007	.131	.008	.056	.955
	Colour of leaf blade	-.586	.144	-.532	-4.067	.000

a. Dependent Variable: avc_Leaf bitterness

1. Model for leaf waxiness

Y=0.108+0.15X

2. Model for colour of main stem

Y=0.105+0.233X

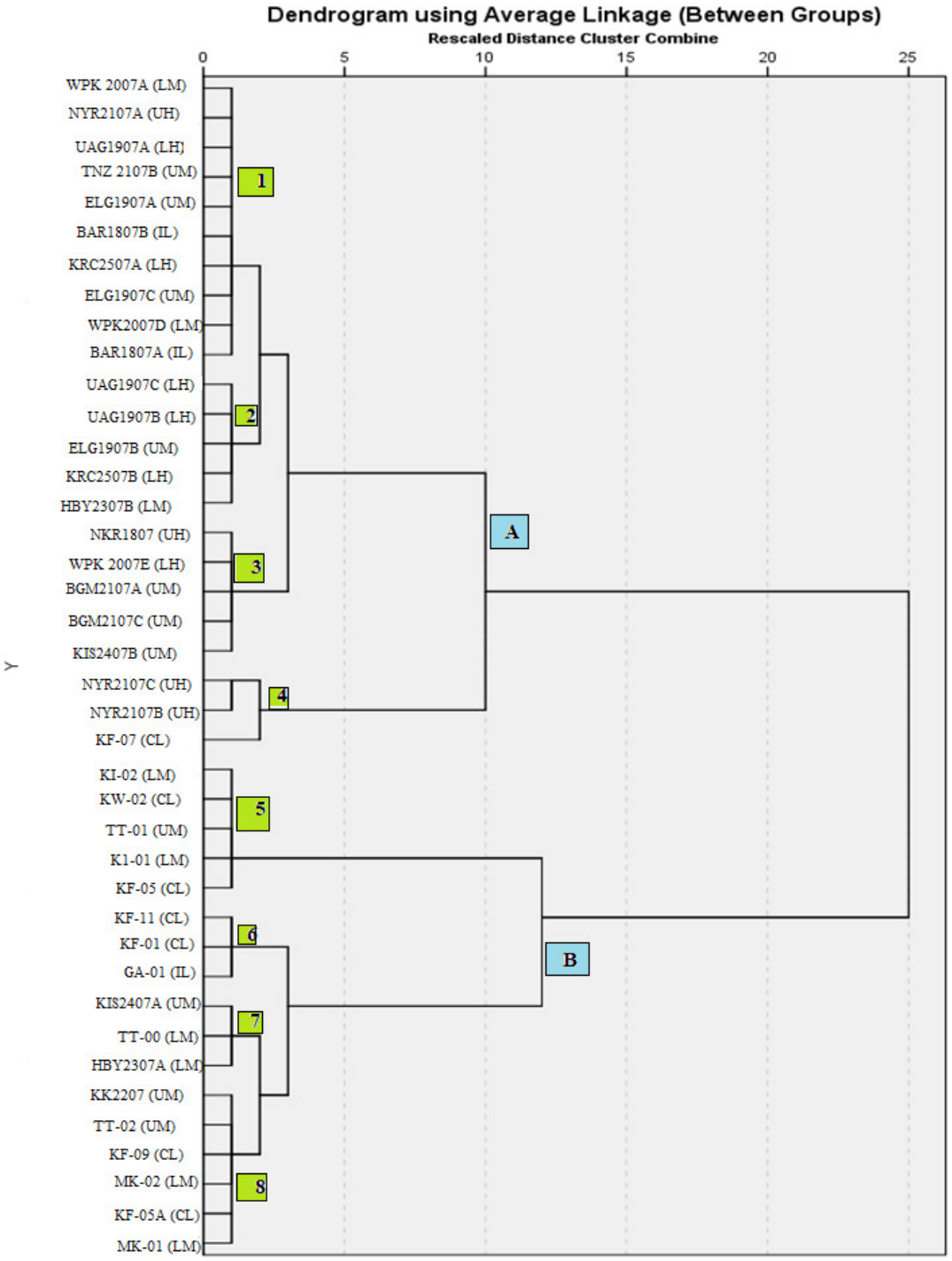
3. Model for stem pubescence

Y=0.007+0.008X

4. Model for colour of leaf blade

Y=-0.586+-0.532X

3.5 Cluster analysis



UH (Upper Highland), UM (Upper Midland), LH (Lower Highland), LM (Lower Midland), IL (Inland Lowland), CL (Coastal Lowland)

Figure 2: Diversity relationship of leaf bitterness levels

Figure 2 is based on relationship on level of bitterness among accessions from different agro ecological zones. The dendogram separated the 40 accessions into two (2) major clusters by grouping accessions into groups A and B, which share similar bitterness levels. The two (2) clusters represent bitter and non-bitter taste and are further divided into eight (8) clusters, with each cluster having accessions related to each other in terms of bitterness levels.

Group A consists of accessions whose bitterness level range from 'non-bitter to mild' in taste, while group B has accessions whose bitterness level range from 'bitter to extremely bitter'. Clusters one (1), two (2), three (3) and four (4) fall under group A, while clusters five (5), six (6), seven (7) and eight (8) fall under group B. Cluster one (1) is the largest and is made of up of 10 clades, in which the accessions have bitterness range of 1.9-2.1 hence mild in taste. Cluster two (2) is divided into five (5) clades with accessions which are non-bitter to slightly mild in taste (1.6-1.8). Cluster three (3) consists of five (5) clades in which the accessions have a bitterness level of 2.3-2.4 thus considered mild in taste. Cluster four (4) is divided into three (3) clades and has a bitterness range of 1.0-1.4 hence non-bitter in taste. It is a unique cluster because the cluster analysis indicates that accession NYR 2107B is a subgroup/variety of accession NYR 2107C and also accession KF-09 is closely related to accession NYR 2107C.

Cluster five (5) is divided into five (5) clades whose accessions have a bitterness range of 3.8-4.2 hence considered very bitter in taste. Cluster six (6) is divided into three (3) clades whose accessions are considered bitter (3.0-3.5). Cluster seven (7) consists of three (3) clades whose accessions are bitter in taste with a bitterness level of 2.7. Cluster eight (8) has six (6) clades in which the accessions have bitterness range of 2.9-3.1 and therefore bitter in taste.

The clustering analysis clearly showed that accessions were related to each other based on range in level of leaf bitterness. It also showed presence of variation in leaf bitterness within agro ecological zones. Unique similarities were noted in cluster four (4) whereby accession KF-07 exhibited non bitter taste yet it was from coastal lowlands which were in group B made up of bitter to very bitter types. This may be attributed to the fact that it was a cultivated type obtained through seed trade probably sourced from upper highlands. This is the reason why accession KF-07 was clustered together with NYR 2107B and NYR 2107C indicating similar genetic background. Accession GA-01 expressed bitter taste (3) yet it was

a cultivated type and therefore could have been sourced from a lowland region (associated with bitter types) through seed trade.

CONCLUSION

A number of observations and conclusions were made from this study using qualitative characters used; sensory and observational. The study showed that there was significant variation in leaf bitterness levels across six (6) agro ecological zones and correlation analysis proved that there was a significant relationship between level of leaf bitterness and colour of leaf blade in spider plant. This means that colour of leaf blade can be used as indicator to distinguish non-bitter types from bitter types in vegetable species; *Cleome gynandra* This can be used by consumers and farmers for selection of bitter types from non-bitter types depending one's preference. The study therefore recommends;

- Use of colour of leaf blade as an indicator of leaf bitterness in spider plant
- further characterization is recommended to validate variation in leaf bitterness by growing the plants in multiple sites under different environments. This can further be supplemented by use of molecular markers such as simple sequence Repeats (SSRs) to identify polymorphism that is not due to environmental conditions
- further characterization to determine levels of bitterness at different stages of spider plant growth i.e. juvenile and maturity stage
- farmers and consumers be taught how to use colour of leaf blade to discriminate types with low levels of bitterness which will reduce boiling or cooking time and hence save nutrients

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