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Fruit Based Morphological Characterization of Vegetable Marrow (*Cucurbita Pepo* L.) for Diversity Assessment and Germplasm Identification

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ABSTRACT

Vegetable marrow, a cultivar group of *Cucurbita pepo* L., is widely grown across Pakistan, yet the fruit based morphological diversity of its local germplasm has rarely been documented in a systematic, quantitative way. This study characterized 15 vegetable marrow accessions (VSQ1-VSQ15) using 13 fruit descriptors recommended by Bioversity International (formerly IBPGR/IPGRI) and the European Cooperative Programme for Plant Genetic Resources (ECPGR) Cucurbits Working Group, comprising nine quantitative traits (fruit length, width, shape index, fresh weight, flesh thickness, cavity diameter, ribbing intensity, stem-end diameter and blossom-end diameter) and 3 qualitative traits (shape class, surface texture and fruit colour). Descriptive statistics revealed diverse variation among accessions, with fruit length (coefficient of variation, CV = 48.1%), shape index (CV = 63.0%) and fresh weight (CV = 41.0%) showing the greatest relative variability, while flesh thickness (CV = 13.4%) was comparatively stable. Pearson correlation analysis showed fresh weight to be the most strongly associated with flesh thickness ($r = 0.70$) and fruit length ($r = 0.59$), while width-related traits (cavity, stem-end and blossom-end diameter) were tightly inter-correlated ($r > 0.7$), reflecting a shared girth dimension. Principal component analysis reduced the nine quantitative traits to two components explaining 82.5% of total variance: PC1 (51.3%) represented an overall fruit-girth axis, and PC2 (31.2%) represented an elongation-mass axis. Ward's hierarchical clustering grouped the 15 accessions into 3 morphologically distinct clusters that broadly corresponded to small pyriform/round types, intermediate blocky types, and elongated bell/long types. One-

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way analysis of variance confirmed significant differences in shape index ($p < 0.001$) and fresh weight ($p < 0.01$) among shape classes with adequate replication. These results demonstrate substantial, structured morphological diversity within a small set of local vegetable marrow accessions and provide a quantitative descriptor foundation that can support germplasm identification, core collection development, and parental selection in future breeding programmes.

Introduction

Cucurbits are economically important vegetable crops cultivated worldwide because of their nutritional, medicinal and commercial value. The family Cucurbitaceae includes several widely cultivated species, among which cucumber (*Cucumis sativus* L.), melon (*Cucumis melo* L.), watermelon (*Citrullus lanatus*) and the three cultivated pumpkin and squash species (*Cucurbita pepo* L., *Cucurbita moschata* Duchesne, and *Cucurbita maxima* Duchesne) are the most important (Janick & Paull, 2008). Native to the Americas, *C. pepo* was introduced to Europe, Asia, and Africa during the fifteenth and sixteenth centuries through transoceanic voyages, after which it became established as an important vegetable crop worldwide (Whitaker, 1947). Although cucurbit crops exhibit considerable fruit diversity, pumpkins and squash are particularly remarkable for their extensive variation in fruit size, shape, surface topography, colour and colour pattern (Paris, 2015). Among the cultivated forms of *C. pepo*, vegetable marrow is one of the most widely grown and consumed vegetables in many countries, including Pakistan.

The annual worldwide value of summer squash generates several billion dollars, making it one of the most commercially important vegetable crops grown today (Paris, 2008). World squash and gourd production reached approximately 22 million metric tons across 1.5 million hectares in 2022, led by Asia (11.5 million tons), Europe (4.8 million tons), the Americas (3.5 million tons) and Africa (2.7 million tons) (FAO, 2024). The largest individual producers that year included Russia, the United States, Spain, Turkey and Mexico, alongside China and India (Science of Agriculture, 2024). The relatively high nutritive and medicinal value of squash, compared with other vegetable crops, has made squash fruit a traditional favorite in many countries. Its constituents include vitamins A and C, niacin, folate and dietary fibre, making the crop a suitable dietary source that may help prevent cancers, heart disease, and inflammatory conditions such as asthma and arthritis; as a low-calorie food, summer squash is also beneficial in weight-management diets (Balluz et al., 2005; Hedstorm & Yerxa, 2008; USDA, 2018).

The vegetables of cucurbitacea family have an important place in the vegetable sector of Pakistan. They are grown extensively in the Punjab and other provinces as a part of the overall horticultural economy of the country. The horticultural economy not only meets the domestic consumption but also contributes to the foreign exchange earnings. However, systematic, trait-level documentation that supports modern breeding and conservation programs elsewhere has not been applied to Pakistan's vegetable marrow holdings, resulting in a substantial gap between the local importance of the crop and the data needed to manage it (Khokhar, 2014).

This same variability has always made the crop difficult to pin down taxonomically. Faced with a species that refuses to hold a single shape, botanists eventually organised its edible-fruited cultivars into eight morphotypes of fruit shape, or cultivar-groups, four within each of the two subspecies of the crop. One of the four morpho types defined within subsp. *pepo* is the Vegetable Marrow Group, defined by a short, tapered, cylindrical fruit, narrow at the peduncle end and broad at the stylar end (Paris, 1986). Vegetable marrow and its close relatives, particularly pumpkin and zucchini, continue to blur into one another, sharing the same herbaceous habit, simple leaves, and yellow-to-orange monoecious flowers (Hora, 2018; Platt, 2013).

Identification of species cultivar-group within *Cucurbita pepo* L. is a persistent problem due to the high external morphological similarity between cultivar groups (especially pumpkin and zucchini). Both share vegetative features of herbaceous habit, simple leaves and yellow-to-orange monoecious flowers. Fruit morphology, although providing a basis for differentiation (pumpkins are generally oval to pyriform with a central cavity, while zucchinis are elongated and cylindrical and lack such a cavity), does not always provide sufficient macro-morphological resolution for accurate differentiation of specimens. This constraint highlights the need for more accurate quantitative and diagnostic methods; for example, anatomical and histochemical analyses have been demonstrated to reveal differentiating characteristics not apparent from gross morphology alone (da Silva et al., 2024).

Plant morphology remains the first and most accessible language of plant identification, the one every breeder, farmer and gene-bank curator can read without a microscope. Yet in regions such as Benin, limited attention has been given to documenting the morphological diversity of local pumpkin varieties. A study evaluating agro-morphological variability among pumpkin accessions collected in Benin revealed considerable variation in fruit-related traits among landraces, including fruit weight, fruit length, and fruit diameter, all of which showed highly significant differences ($p < 0.001$). Fruit shape and colour also displayed notable phenotypic variability across the landraces studied. A positive correlation ($r = 0.76$; $p < 0.05$) was found

between average fruit weight and average seed number per fruit, suggesting a link between fruit size and seed productivity, and high heritability estimates were reported for fruit diameter (96.73%), average fruit weight (96.46%) and fruit length (94.64%), traits that were further associated with high genetic advance, indicating strong potential for trait-based selection. Such findings emphasize the extent of fruit morphological diversity present within local pumpkin landraces and highlight the value of systematic morphological characterization for varietal identification, germplasm conservation, and breeding (Ezin et al., 2022).

Fruit-based characterization of *Cucurbita pepo* has attracted growing attention worldwide as a fast, low-cost complement to molecular profiling. In Turkey, morphological survey of twenty-nine local pumpkin genotypes from Erzincan province revealed wide variability in plant, flower, fruit and leaf traits that was subsequently confirmed by SSR-based clustering into four major groups (Ozturk et al., 2022). In Iran, quantitative fruit and seed traits recorded on twenty-one *Cucurbita pepo* accessions were reduced by principal component analysis to five components explaining nearly 90% of total variance and were used to resolve six divergent clusters, several of which were recommended as parental material for future breeding (Darrudi et al., 2018).

A comparable agro-morphological and genetic-parameter study of squash genotypes in Bangladesh, where the crop is known locally as marrow, found the highest phenotypic coefficients of variation and heritability for fruit weight, length and diameter, echoing the traits identified as most discriminating in other *Cucurbita* surveys (Sajid et al., 2022). At the molecular level, a wide-based collection of Spanish *Cucurbita pepo* landraces and commercial cultivars was resolved by AFLP and SRAP markers into clusters that broadly tracked the two recognised subspecies and, within them, the classical fruit-shape cultivar-groups, including Vegetable Marrow (Ferriol et al., 2003; Formisano et al., 2012). More recent marker-based studies using AFLP, ISSR, SSR and high-frequency oligonucleotide-targeting active gene (HFO-TAG) polymorphisms have likewise shown that accessions of the Vegetable Marrow, Cocozelle, Zucchini and Pumpkin cultivar-groups form partially distinct sub-clusters that nonetheless overlap at their margins, underscoring both the taxonomic reality and the practical difficulty of separating these morphotypes on collection data alone (Paris et al., 2003; Paris et al., 2015).

Most recently, genotyping-by-sequencing of the entire USDA *Cucurbita pepo* germplasm collection confirmed that genetic structure in the species aligns closely with geographic origin and market class/morphotype, reinforcing the value of morphotype-referenced characterization, such as that undertaken here, as a first step toward efficient germplasm management (Hernandez et al., 2023). Despite this substantial international literature, none of these studies has focused on vegetable marrow germplasm maintained in Pakistan, leaving local material effectively un-characterized by comparison.

The choice of traits used in the present study is not arbitrary. Standardized descriptor lists for *Cucurbita*, originally developed under the International Board for Plant Genetic Resources (IBPGR) framework (Esquinas-Alcazar & Gulick, 1983) and refined by the ECPGR Working Group on Cucurbits (2008), define this set of fruit-level characters (shape, dimensions, cavity, ribbing, surface texture, and colour) as the minimum standard for characterizing cultivated *Cucurbita* germplasm. Morphological descriptors are considered essential for the preservation, use, and management of germplasm collections, and characterization built on them helps gene-bank staff refine conservation strategies while supporting genetic-improvement work. Although the crop has a long history of cultivation and is popular in Pakistan, systematically compiled morphological data of this type are lacking for local vegetable marrow germplasm. Such data are necessary to identify truly distinct accessions from redundant ones and to make informed decisions regarding future breeding and conservation efforts.

This study sets out to close that gap by characterizing vegetable marrow fruit by shape class, length, width, shape index, fresh weight, flesh thickness, cavity diameter, ribbing, stem-end and blossom-end diameter, surface texture and colour, and by applying multivariate statistical analysis in the R software environment (R Core Team, 2024) to (i) quantify the magnitude and pattern of morphological variability among fifteen local accessions, (ii) identify the trait combinations that best discriminate accessions, and (iii) group accessions into natural clusters, thereby assessing the diversity present in local germplasm and building the descriptor foundation needed for future germplasm identification, conservation, and breeding work.

Materials and Methods

Plant material

Fifteen accessions of vegetable marrow (*Cucurbita pepo* L., Vegetable Marrow Group), coded VSQ1 to VSQ15, were drawn from a local germplasm working collection representative of the material grown in Pakistan's Punjab province. One physiologically mature, fully developed and disease-free fruit was selected to represent each accession, harvested at the marketable-to-physiological maturity stage typically used for descriptor recording in *Cucurbita*, laid out on a uniform background and photographed together for visual record (Figure 1).

Morphological data collection

Thirteen fruit descriptors were recorded for each accession following the minimum descriptor list for cultivated Cucurbita spp. jointly derived from the IBPGR global report on Cucurbitaceae genetic resources (Esquinas-Alcazar & Gulick, 1983) and its subsequent revision by the ECPGR Working Group on Cucurbits (2008). Nine traits were quantitative: fruit length (cm, measured along the stem-to-blossom axis), fruit width (cm, at the widest point), shape index (length:width ratio), fresh fruit weight (g), flesh thickness (cm, at maximum diameter), cavity diameter (cm), ribbing intensity (scored on a 1-5 ordinal scale from smooth to strongly ribbed), stem-end diameter (cm) and blossom-end diameter (cm). Four traits were qualitative or semi-qualitative: fruit shape class (assigned according to standard Cucurbita fruit-shape categories - long blocky, long bell, round, long, long gourd curvy, disc and pear), surface texture (smooth or ribbed) and fruit colour (recorded descriptively, including bicolour and pattern variants).

Statistical analysis

All statistical analyses were performed in the R statistical computing environment, version 4.x (R Core Team, 2024), using the base stats package together with the psych, corrplot, FactoMineR, factoextra, cluster and tidyverse/ggplot2 packages for descriptive, multivariate and graphical analysis; the complete analysis script is reproduced in the Supplementary Material. Descriptive statistics (mean, standard deviation, minimum, maximum and coefficient of variation, CV%) were computed for the nine quantitative traits to summarise the magnitude of trait variability, following the interpretive convention that a CV above roughly 20% denotes high relative variability (Mohammadi & Prasanna, 2003). Pearson product-moment correlation coefficients were calculated among all pairs of quantitative traits and visualised as a correlation heat map.

Principal component analysis (PCA) was performed on the standardized (mean-centred, unit-variance) quantitative trait matrix to reduce dimensionality and identify the trait combinations accounting for the largest share of morphological variance; component scores and trait loadings were extracted and plotted as a biplot. Agglomerative hierarchical cluster analysis was performed on the standardized trait matrix using squared Euclidean distance and Ward's minimum-variance linkage criterion (Ward, 1963), and the resulting dendrogram was cut to define natural groupings among accessions. Differences in shape index and fresh weight among shape classes represented by more than one accession (Long Blocky, Long Bell, Round, Long and Pear; $n = 11$ accessions in five classes) were tested by one-way analysis of variance (ANOVA); the two singleton classes (Disc, Long Gourd Curvy) were excluded from this specific test because a class of $n = 1$ provides no within-group variance estimate, though they were retained in all descriptive, correlation, PCA and cluster analyses.

Results

Fruit material and diversity overview

Raw morphological data recorded for all fifteen accessions across the thirteen descriptors are presented in Table 1.

Table 1. Fruit morphological descriptor data recorded for 15 vegetable marrow accessions

Sr. No	Fruit ID	Shape Class	Length (cm)	Width (cm)	L/W	Fresh Wt (g)	Flesh Thickness (cm)	Cavity Diam. (cm)	Ribbing	Stem End (cm)	Blossom End (cm)	Texture	Fruit Colour
1	VSQ1	Long Blocky	26.92	11.82	2.28	1470	2.36	9.59	4	8.30	11.92	Ribbed	Dark Green with Orange Lines
2	VSQ2	Long Bell	31.75	10.86	2.92	1570	2.66	6.87	4	8.04	11.20	Ribbed	Green with Yellow Stripes & Patchy
3	VSQ3	Long Blocky	23.11	11.13	2.08	1110	2.34	6.19	5	8.32	10.56	Ribbed	Dark Green
4	VSQ4	Round	12.95	12.26	1.06	715	2.19	8.48	2	9.51	9.82	Ribbed	Dark Green, Orange Stripes, Patchy

5	VSQ5	Long	36.57	9.94	3.68	1935	2.63	5.09	3	8.36	9.98	Rib bed	Lime
6	VSQ 6	Long Gourd Curvy	49.00	8.08	6.06	1520	2.01	4.30	3	5.94	9.11	Rib bed	Lime
7	VSQ7	Disc	12.95	20.02	0.65	2220	3.11	14.41	3	16.51	17.78	Rib bed	Lime
8	VSQ 8	Pear	16.51	14.35	1.15	1055	2.31	9.40	2	6.58	13.14	Rib bed	Yellow
9	VSQ 9	Pear	10.16	9.33	1.09	415	2.41	4.00	1	6.27	8.90	Smo oth	Yellow
10	VSQ1 0	Pear	10.66	11.04	0.97	515	2.04	7.02	2	8.75	10.06	Rib bed	Yellow
11	VSQ1 1	Long Bell	34.03	9.87	3.45	1755	2.48	5.91	3	7.79	10.80	Rib bed	Yellow
12	VSQ1 2	Long Blocky	21.33	12.62	1.69	1545	2.12	8.39	4	10.08	10.76	Rib bed	Yellow
13	VSQ1 3	Long	34.54	12.51	2.76	2400	3.08	7.65	4	9.01	11.06	Rib bed	Yellow
14	VSQ1 4	Round	13.46	10.50	1.28	1260	2.42	10.52	3	11.19	11.88	Rib bed	Lime
15	VSQ1 5	Long Bell	34.03	10.53	3.23	1760	2.40	6.02	4	7.22	9.97	Rib bed	Yellow

The 15 accessions displayed conspicuous, readily visible diversity in fruit size, shape, and colour when arranged together for photographic recording (Figure 1), ranging from small, near-spherical fruits of roughly 400-700 g to large, elongated fruits exceeding 2 kg with pronounced ribbing and curvature. This visual range foreshadows the quantitative variation captured by the descriptor data (Table 1).



Fig. 1. The 15 vegetable marrow (*Cucurbita pepo* L.) accessions (VSQ1-VSQ15) evaluated in this study, illustrating the range of fruit size, shape and colour recorded.

Descriptive statistics and trait variability

Descriptive statistics for the 9 quantitative traits are summarized in Table 2. Mean fruit length was 24.53 cm (range 10.16-49.00 cm) and mean fruit width was 11.66 cm (range 8.08-20.02 cm), giving a mean shape index of 2.29 (range 0.65-6.06). Mean fresh weight was 1416.3 g (range 415-2400 g). The coefficient of variation was highest for shape index (63.0%), fruit length (48.1%) and fresh weight (41.0%), indicating that elongation and fruit mass were the most variable characters among

accessions, whereas flesh thickness (CV = 13.4%) and blossom-end diameter (CV = 19.3%) were comparatively conservative traits, varying relatively little regardless of overall fruit size or shape.

Table 2. Descriptive statistics of nine quantitative fruit traits (n = 15 accessions).

Trait	Mean	SD	Min	Max	CV (%)
Fruit length (cm)	24.53	11.80	10.16	49.00	48.09
Fruit width (cm)	11.66	2.77	8.08	20.02	23.77
Shape index (L/W)	2.29	1.44	0.65	6.06	63.04
Fresh weight (g)	1416.33	580.94	415.00	2400.00	41.02
Flesh thickness (cm)	2.44	0.33	2.01	3.11	13.39
Cavity diameter (cm)	7.59	2.70	4.00	14.41	35.54
Ribbing (1-5)	3.13	1.06	1.00	5.00	33.83
Stem-end diameter (cm)	8.79	2.56	5.94	16.51	29.10
Blossom-end diameter (cm)	11.13	2.15	8.90	17.78	19.30

Relationships among quantitative traits

Fruit length and width followed clearly divergent trajectories across accessions (Figure 2): several accessions (e.g., VSQ6, VSQ5, VSQ13) combined long fruit length with comparatively narrow width, producing markedly elongated shapes, whereas others (e.g., VSQ7, VSQ4) combined short length with large width, producing flattened, disc-like or round fruit. This inverse patterning between length and width across accessions is consistent with the wide range of shape index values reported in Table 2.

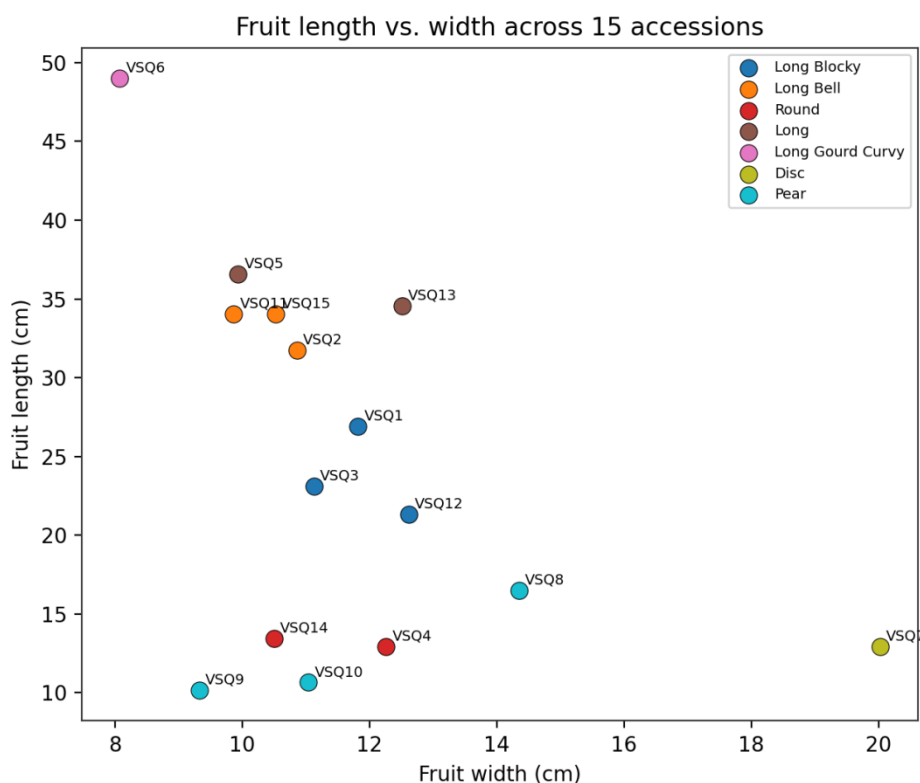


Figure 2. Relationship between fruit length and fruit width across the 15 accessions, coloured by fruit shape class.

The Pearson correlation matrix (Figure 3) showed that fresh weight was most strongly associated with flesh thickness ($r = 0.70$) and fruit length ($r = 0.59$) and to a lesser extent with ribbing intensity ($r = 0.58$) and shape index ($r = 0.42$), confirming that larger, more elongated, more heavily ribbed fruit tended to be heavier. A separate cluster of strong positive correlations ($r = 0.6$ - 0.9) linked fruit width, cavity diameter, stem-end diameter and blossom-end diameter, indicating that these four traits largely track a single underlying girth dimension of the fruit. Fruit length and shape index were, by contrast, weakly or negatively correlated with these girth traits, confirming that elongation and girth behave as largely independent axes of fruit form in this germplasm set.

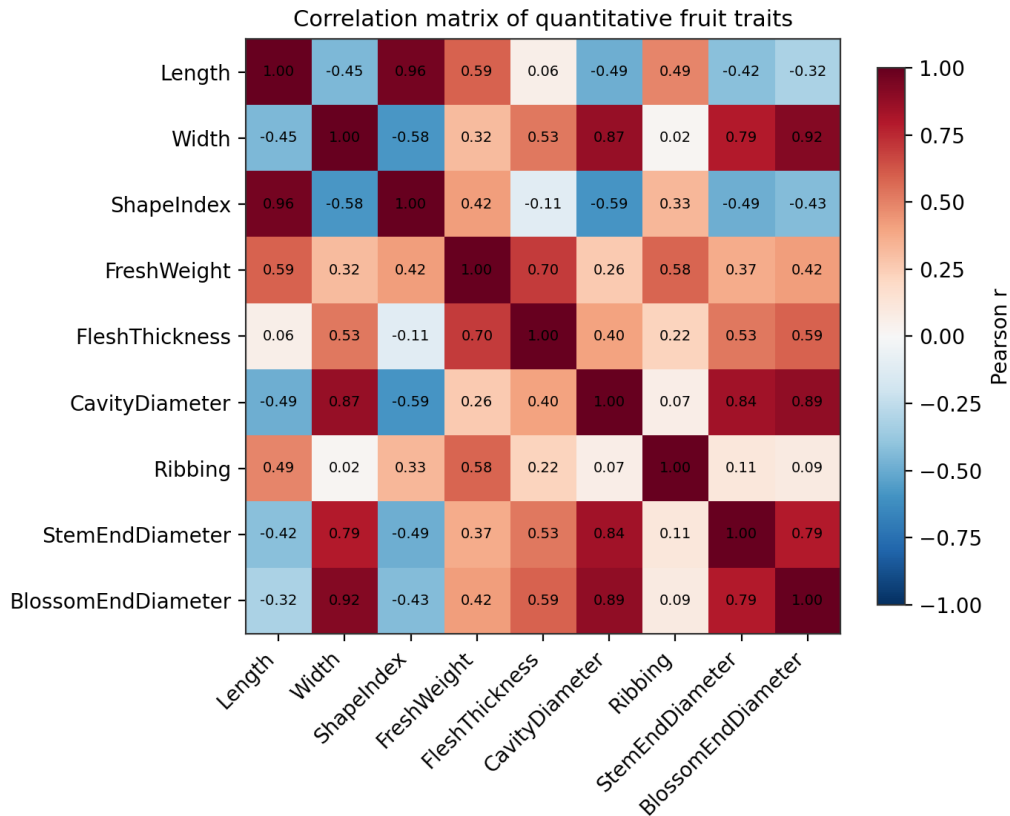


Figure 3. Pearson correlation heat map among nine quantitative fruit traits.

Principal component analysis

Principal component analysis of the nine standardized quantitative traits produced two dominant components together accounting for 82.5% of total variance ($PC_1 = 51.3\%$, $PC_2 = 31.2\%$; Figure 4), with subsequent components each contributing less than 8% (Table 3). PC_1 was defined by strong positive loadings for width, cavity diameter, stem-end diameter and blossom-end diameter, and can be interpreted as an overall fruit-girth axis. PC_2 was defined by strong positive loadings for fresh weight, ribbing and fruit length, together with a negative loading for shape index acting in the opposite sense, and is best interpreted as an elongation-mass axis that separates long, heavy, ribbed fruit from short, light, smooth ones.

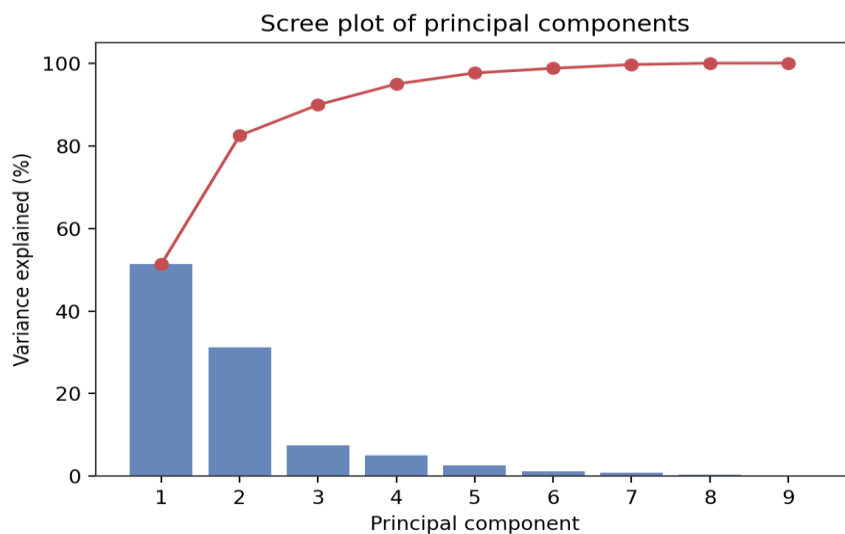


Figure 4. Scree plot showing the percentage of variance explained by each principal component (bars) and cumulative variance (line).

The PCA biplot (Figure 5) separated accessions largely along the two axes identified above. Disc-shaped VSQ7, with its exceptionally wide, shallow fruit, scored at the extreme positive end of PC1 as expected for its very high width and cavity diameter, whereas the curved, narrow-girth VSQ6 (Long Gourd Curvy) scored at the extreme negative end. Along PC2, the heavy, long, well-ribbed VSQ13 (Long) and the elongated VSQ5 (Long) scored highest, while the small pyriform accessions VSQ9 and VSQ10 (Pear) scored lowest, consistent with their low fresh weight and shape index.

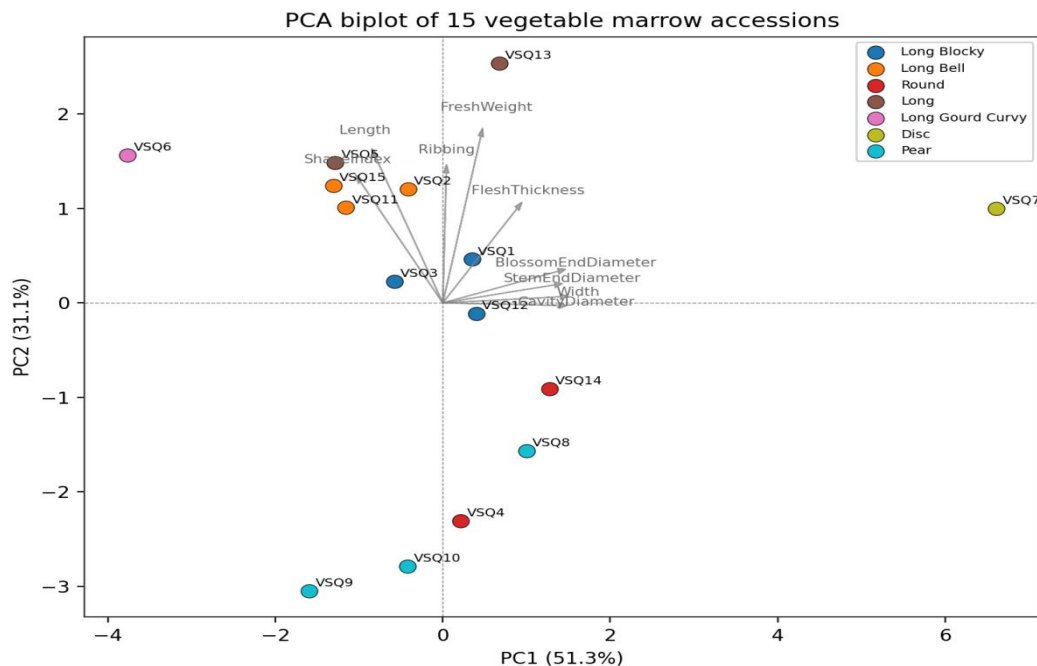


Figure 5. Principal component biplot (PC1 vs PC2) of 15 vegetable marrow accessions, with trait loading vectors overlaid and accessions coloured by fruit shape class.

Table 3. Trait loadings on the first three principal components.

Trait	PC1	PC2	PC3
Fruit length (cm)	-0.253	0.486	-0.071
Fruit width (cm)	0.440	0.021	-0.015
Shape index (L/W)	-0.308	0.401	-0.105
Fresh weight (g)	0.142	0.552	-0.150
Flesh thickness (cm)	0.277	0.313	-0.553
Cavity diameter (cm)	0.435	-0.008	0.221
Ribbing (1-5)	0.013	0.430	0.772
Stem-end diameter (cm)	0.419	0.060	0.098
Blossom-end diameter (cm)	0.431	0.105	-0.038
Eigenvalue (% variance)	51.32%	31.15%	7.44%

Hierarchical cluster analysis

Ward's minimum-variance clustering of the standardized trait matrix (Figure 6) grouped the fifteen accessions into three well-separated clusters. Cluster I comprised the small, low-weight pyriform and round accessions (VSQ9, VSQ10, VSQ4, VSQ8, VSQ14) together with the disc-shaped VSQ7, which joined this branch at a comparatively large distance owing to its unusually wide fruit. Cluster II comprised the intermediate long-blocky and round-to-blocky accessions (VSQ6, VSQ3, VSQ1, VSQ12). Cluster III comprised the elongated, heavier long and long-bell accessions (VSQ13, VSQ5, VSQ11, VSQ2, VSQ15). This three-cluster structure is broadly consistent with the shape-class assignments made in the field and with the separation of accessions observed in the PCA biplot, indicating good agreement between independent morphological classification approaches.

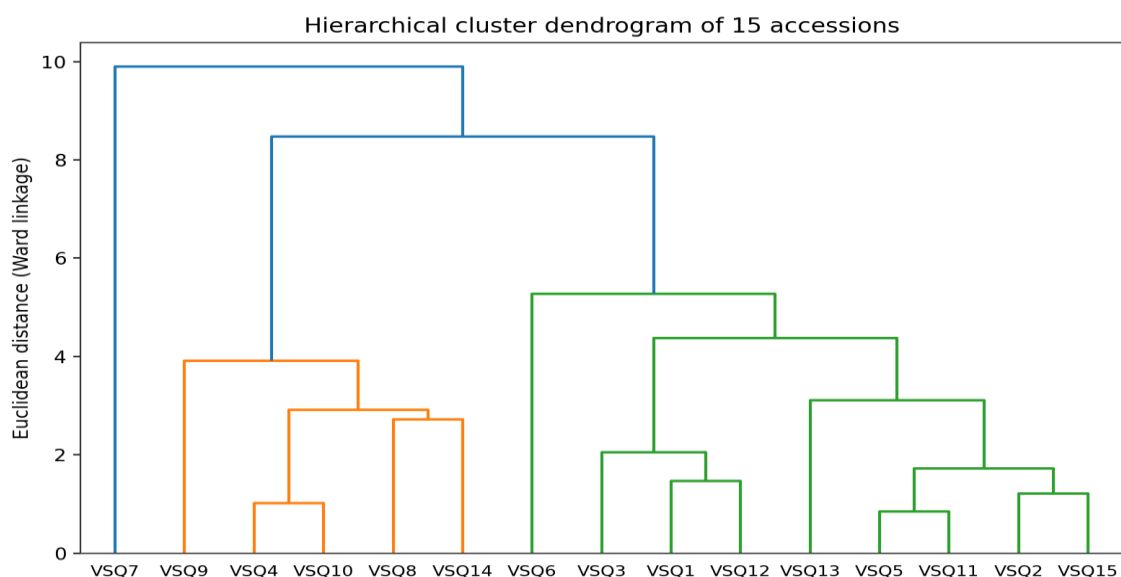


Figure 6. Ward's hierarchical cluster dendrogram of 15 vegetable marrow accessions based on Euclidean distances among standardized quantitative fruit traits.

Qualitative trait diversity

Frequency distributions of the qualitative descriptors (Figure 7) showed that seven distinct fruit shape classes were represented among only fifteen accessions, with Long Blocky, Long Bell and Pear each represented by three accessions and the remaining classes by one or two accessions, reflecting a broad and largely unstructured sampling of the shape spectrum of the Vegetable Marrow Group. Yellow was the single most frequent fruit colour (six accessions), followed by Lime (three accessions), with the remaining accessions showing distinctive bicolour or patchy colour patterns (e.g., dark green with orange lines, green with yellow stripes). Surface texture was overwhelmingly ribbed (fourteen of fifteen accessions), with only one accession (VSQ9) recorded as smooth-skinned. Ribbing intensity scores were concentrated at the intermediate levels of 3 and 4 (five accessions each), with fewer accessions at the extremes of 1 (smooth) and 5 (heavily ribbed).

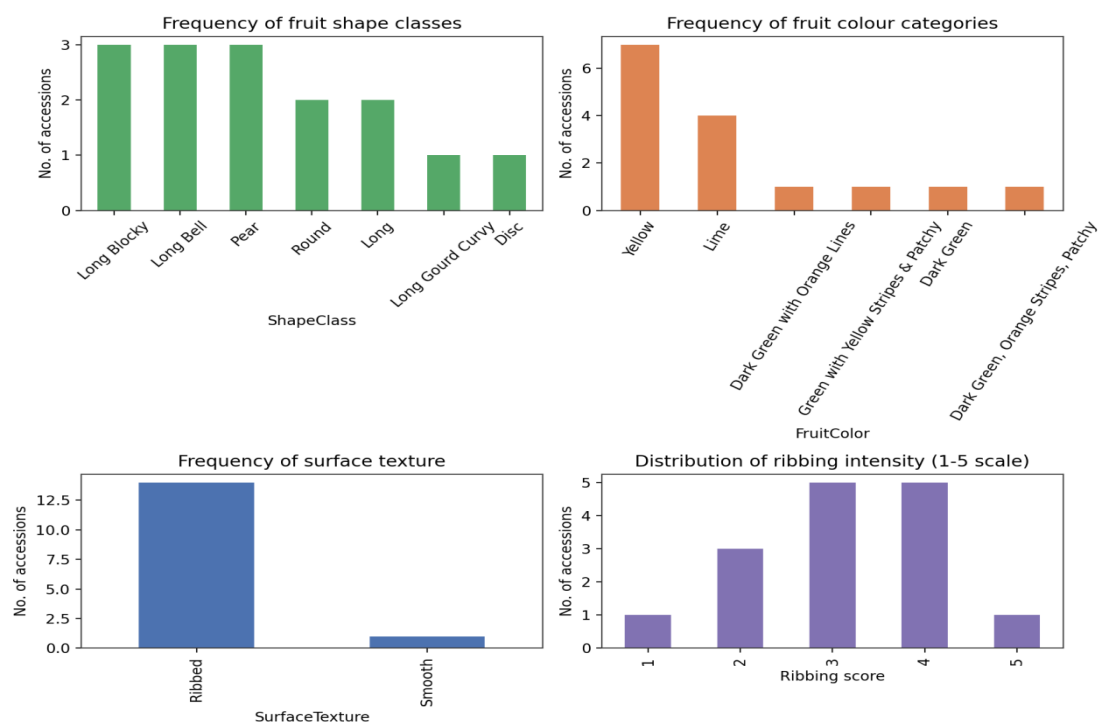


Figure 7. Frequency distribution of (a) fruit shape class, (b) fruit colour, (c) surface texture, and (d) ribbing intensity score across 15 accessions.

Trait variation among fruit shape classes

Boxplots of shape index and fresh weight grouped by shape class (Figure 8) illustrate a clear, monotonic ordering of classes from the compact Pear and Round types through to the strongly elongated Long and Long Gourd Curvy types. Restricting the comparison to the five shape classes represented by more than one accession (Long Blocky, Long Bell, Round, Long and Pear; $n = 11$), one-way ANOVA confirmed that both shape index ($F = 28.38$, $p < 0.001$) and fresh weight ($F = 10.81$, $p = 0.003$) differed significantly among shape classes, indicating that the shape-class assignments made from visual inspection are well supported by the underlying quantitative measurements and are not an arbitrary categorical overlay on continuous variation.

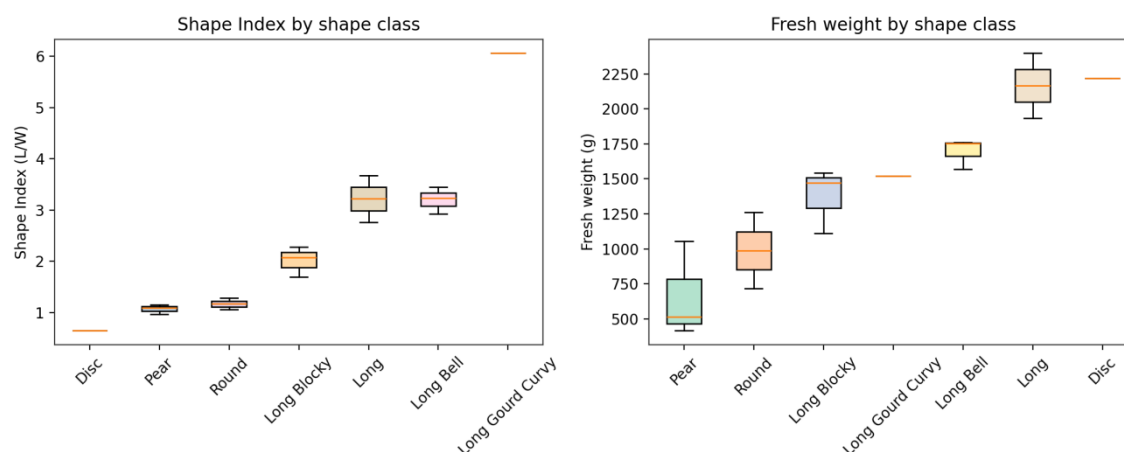


Figure 8. Boxplots of (a) shape index and (b) fresh weight across fruit shape classes, ordered by median value.

Discussion

The fifteen vegetable marrow accessions examined in this study displayed a level of fruit based morphological diversity that is remarkable given the small sample size, spanning nearly the full range of fruit-shape categories recognized within the Vegetable Marrow Group and its neighbouring cultivar groups of *Cucurbita pepo* (Paris, 1986). Coefficients of variation for shape index (63.0%), fruit length (48.1%) and fresh weight (41.0%) fall well within, or above, the range of high variability reported for fruit traits in other *Cucurbita* landrace surveys; in Beninese pumpkin (*Cucurbita moschata*) landraces, for example, fruit weight, length and diameter all differed highly significantly among accessions and fruit-diameter and fruit-weight heritability exceeded 96% (Ezin et al., 2022), underscoring that fruit dimensions are typically the most discriminating and most heritable descriptors available for morphological characterization of cucurbit germplasm. The comparatively low CV recorded here for flesh thickness (13.4%) mirrors a broader pattern in cucurbit descriptor studies, in which internal traits tend to be buffered against the same drivers - genetic segregation at fruit-shape loci and environmental heterogeneity - that produce large swings in external fruit dimensions.

The correlation structure in width, cavity diameter, stem-end diameter and blossom-end diameter form a tightly inter-correlated block while length and shape index vary comparatively independently, was consistent with the widely reported developmental decoupling of fruit elongation from fruit girth in *Cucurbita pepo*, a decoupling that underlies the very existence of distinct 'long' and 'round/disc' morphotypes within the species (Paris, 2015). That fresh weight correlated most strongly with flesh thickness and length, rather than with width alone, suggested that in this germplasm set fruit mass is driven jointly by the elongation axis and by the depth of edible flesh, rather than by girth per se; this is a practically useful finding for breeders selecting for yield-related fruit mass, since it implies that girth alone is a poor single proxy for weight in these materials.

The reduction of nine quantitative traits to two principal components explaining 82.5% of total variance, an unusually high proportion for a morphological dataset, indicates that fruit form in this collection is governed by relatively few underlying developmental axes: an overall girth axis (PC1) and an elongation-mass axis (PC2), echoing the two-dimensional (length x width) conceptualisation of *Cucurbita* fruit shape that underlies standard descriptor systems (Esquinas-Alcázar & Gulick, 1983; ECPGR Working Group on Cucurbits, 2008). Comparable PCA-based reductions of fruit or plant traits to two or three dominant components explaining the bulk of variance are routinely reported in Cucurbitaceae diversity studies and are considered a hallmark of a well-chosen, non-redundant descriptor set (Mohammadi & Prasanna, 2003). The proportion of variance captured here compares favourably with other *Cucurbita pepo* fruit-trait PCAs: Darrudi et al. (2018) needed five components to explain a similar 89.9% of variance across a larger set of fifteen fruit and seed traits in Iranian accessions, while Sajid et al. (2022) likewise identified fruit weight, length and diameter as the traits carrying the greatest phenotypic coefficient of variation and

heritability among Bangladeshi squash genotypes, closely matching the length-, weight- and shape-index-dominated PC2 axis recovered in the present study.

The three-cluster structure recovered by Ward's hierarchical clustering, separating small pyriform/round types, intermediate blocky types, and elongated bell/long types, aligned closely with the a priori shape-class assignments and with the spatial separation of accessions in the PCA biplot. This convergence between an unsupervised numerical method and field-based visual classification provides an internal consistency check on the descriptor set, and is analogous to the concordance between clustering and morphotype classification reported for *Cucurbita pepo* germplasm elsewhere (Paris, 1986, 2015). Marker-based studies of much larger, more heterogeneous *C. pepo* collections have reported a broadly similar pattern at a coarser taxonomic scale: AFLP- and SRAP-based clustering of Spanish landraces separated accessions first by subspecies and then, within subsp. *pepo*, into sub-clusters that only partially resolved the Vegetable Marrow, Cocozelle, Pumpkin and Zucchini cultivar-groups from one another (Ferriol et al., 2003; Formisano et al., 2012), while HFO-TAG marker analysis of a wider international panel found the same four cultivar-groups forming dissimilarity clusters with overlapping margins rather than fully discrete blocks (Paris et al., 2015). The present morphology-only clustering of vegetable marrow accessions achieved cleaner three-way separation than these molecular studies typically achieve for cultivar-group discrimination, which is plausible given that the sample here was drawn from a single cultivar-group and clustered on the very fruit-shape traits that define morphotype boundaries by definition, whereas the molecular studies clustered genome-wide markers that are only indirectly linked to fruit shape. The disc-shaped accession VSQ7 behaved as a partial outlier in both the PCA and cluster analyses, joining Cluster I only at a relatively large distance; such wide, flattened fruit combine an extreme girth phenotype with a short length, and their morphological 'distance' from more typical marrow shapes is exactly the kind of signal that trait-based characterization is designed to detect, since it flags material that may be of particular interest for expanding the phenotypic range of a breeding programme or for further taxonomic scrutiny given the acknowledged difficulty of distinguishing pumpkin, squash and marrow fruit types on gross morphology alone (da Silva et al., 2024).

Qualitative descriptors added complementary diversity information that the quantitative traits alone could not capture: fruit colour ranged from solid yellow and lime through to complex bicolour and striped patterns, and ribbing intensity varied continuously from smooth to heavily ribbed. The predominance of ribbed fruit (fourteen of fifteen accessions) and the concentration of ribbing scores at intermediate levels suggest that smooth-skinned marrow (as in VSQ9) is comparatively rare in this local germplasm pool, a pattern worth confirming in a larger sample, since smooth skin is often a trait of specific market or processing interest.

Two limitations of the present study should be acknowledged. First, the sample of fifteen accessions, while sufficient to demonstrate clear multivariate structure, was small and several shape classes were represented by only one or two accessions; the ANOVA reported here for shape index and fresh weight was accordingly restricted to the five classes with adequate replication and results for singleton classes (Disc, Long Gourd Curvy) should be treated as descriptive rather than statistically tested. Second, each accession was represented by a single fruit; standard germplasm characterization protocols recommend measurements on a minimum of ten fruits per accession (ECPGR Working Group on Cucurbits, 2008) to separate genetic variation among accessions from within-accession and environmental variation, and future work should expand both the number of accessions and the number of fruit sampled per accession, ideally across more than one growing season, to obtain robust heritability and genetic-advance estimates comparable to those reported for other *Cucurbita* germplasm (Ezin et al., 2022).

Conclusion

This study provides the first systematically quantified, multivariate characterization of fruit based morphological diversity in a set of local Pakistani vegetable marrow (*Cucurbita pepo* L.) accessions. Substantial and structured variation was documented across 13 IBPGR/ECPGR-based descriptors, with shape index, fruit length and fresh weight identified as the most variable and most diagnostic quantitative traits. Multivariate analysis condensed this variability into two dominant, biologically interpretable axes of fruit form, girth and elongation-mass, that together explained over 80% of total variance, and hierarchical clustering resolved the fifteen accessions into three coherent morphological groups that agreed closely with field shape-class assignments. These findings confirmed that a compact set of fruit descriptors, analyzed with standard multivariate statistical tools, is sufficient to reveal meaningful genetic diversity structure in vegetable marrow germplasm and they establish a quantitative descriptor baseline that can be extended to larger accession collections to support germplasm identification, redundancy removal, core-collection formation, and the selection of morphologically divergent parents for future vegetable marrow breeding programmes in Pakistan.

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