

PARTICIPATORY SURVEY OF FRUIT AND QUALITY TRAITS OF PUMMELO (*CITRUS MAXIMA* (BURM.) MERR.) ACCESSIONS FROM SAMASTIPUR AND MUZAFFARPUR DISTRICTS OF BIHAR

Hemlata Singh¹, Jyostnarani Pradhan^{*1}, Kumari Sunita¹, Yash Bhardwaj¹, Rajat Singh¹, Varun Kashyap¹, Geeta Kumari¹, Devendra Kumar¹, Roshni Agnihotri^{1*}

¹Dr. Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar, India-848125

*Email of Corresponding Author: roshni@rpcau.ac.in/jyotsna.pradhan@rpcau.ac.in

ABSTRACT

Pummelo (*Citrus maxima* (Burm.) Merr.) is grown in north Bihar mostly as scattered homestead trees, and its variation has not been described. A participatory survey was carried out in 2020 and 2021 in 16 villages of Samastipur and Muzaffarpur districts, covering 60 farmer-managed accessions (29 and 31 per district). Fruit weight, segment number, peel thickness, fruits per tree, total soluble solids (TSS) and titratable acidity were recorded in three replicates, and the TSS/acidity ratio was calculated. Eating quality (sweet or sour), fruit shape and segment colour were scored as classes. The data were analysed by one-way ANOVA and principal component analysis (PCA). Accessions differed significantly ($P < 0.001$) for all seven traits. Accession means ranged from 0.65 to 3.00 kg for fruit weight, 0.83 to 3.90 cm for peel thickness and 50 to 400 for fruits per tree. TSS averaged 8.62 °Brix, acidity 0.53% and the TSS/acidity ratio 17.07. The two districts were similar for all traits, and most variation lay among individual trees. Of the 60 accessions, 90% were rated sweet. Sour accessions had a TSS of 7.2 °Brix or less and a ratio below 11, and the ratio separated the two classes more clearly than TSS did. Three principal components explained 64.5% of the variation, but the KMO value was low (0.49), so the PCA is descriptive only. Eight accessions combined TSS of at least 8.9 °Brix, acidity of 0.50% or less and fruit weight of at least 1 kg, and accessions 34, 41, 5 and 33 were the most promising. These are candidates for vegetative multiplication and multi-season evaluation under uniform conditions, and the survey gives a baseline for conserving and selecting pummelo in Bihar.

KEYWORDS: *Citrus maxima*, pummelo, germplasm survey, fruit quality, TSS/acidity ratio, principal component analysis, Bihar

INTRODUCTION

Pummelo is the largest fruit in the genus *Citrus* and one of its ancestral species. Genomic work has shown that it contributed to the origin of many cultivated citrus types, including sweet orange and grapefruit (Wu *et al.*, 2018). The fruit is borne on spreading evergreen trees in the humid tropics and subtropics of South and South-East Asia. Typical fruits are large and round to pear-shaped with a thick, spongy peel and big juice sacs inside firm segments, and the flavour ranges from sweet and faintly bitter to distinctly acidic (Morton, 1987; Saunt, 2000). The fruit has 16-18 segmented flesh (Ladaniya, 2008). The color of flesh varies from white, pink to red-colored. The juice recovery percentage is around 55–60 %. The total soluble solids (TSS) content of fruit juice ranges between 8 and 12 °Brix. The fruit tastes sweet and is slightly acidic with a hint of bitterness. It is a rich source of ascorbic acid and calories (Singh *et al.*, 2022). In north Bihar the pummelo, known locally as *Gagar Nimbu*, is a familiar fruit around homesteads and field margins. Samastipur and Muzaffarpur districts lie in the alluvial plain north of the Ganga, and trees in both districts are mostly individual plants or small groups maintained by farm families rather than commercial orchards. Named cultivars are not in general use, and planting material appears to come from neighbours and local nurseries. Fruit weight, peel thickness and segment number determine how much edible portion a consumer or processor gets from a fruit, and the number of fruits per tree sets the return to the grower. For eating quality, the balance of sugars and acids matters at least as much as sugar content. The ratio of total soluble solids to titratable acidity is a routine index of palatability and maturity in citrus (Ranganna, 1986; Cheong *et al.*, 2012). Pummelo populations in other countries have shown wide variation for such traits, and participatory surveys that record what farmers grow and prefer have been used to pick strains for further improvement (Paudyal and Haq, 2008). For Bihar no comparable description is available, and without it, conservation, selection and farmer advice have little to build on. This participatory survey therefore set out to describe and test variation in fruit and quality traits among 60 pummelo accessions from two districts of north Bihar, to compare the districts and shortlist accessions that combine good size with good eating quality.

2. MATERIALS AND METHODS

Participatory survey, survey area and accessions

A participatory survey was conducted during 2020 and 2021 using a structured questionnaire administered to farmers (Table 1). Villages were selected randomly from two districts of Bihar, Samastipur and Muzaffarpur. The survey covered 60 pummelo accessions in 16 villages, 29 in Samastipur and 31 in Muzaffarpur. Each accession was a separate farmer-managed tree or planting, numbered 1 to 60 in the field record.

Table 1. Structure of the questionnaire used in the participatory survey.

Section	Content	Information recorded	Response type
A	Identification	Accession number, village, district, date of survey	Entry
B	Fruit characters	Fruit weight, number of segments, peel thickness, fruit shape, segment colour	Measured (three replicates) and coded classes
C	Tree yield	Number of fruits per tree	Count (Current year and previous two-year count)
D	Fruit chemistry	TSS, titratable acidity, TSS/acidity ratio	Measured (three replicates) and calculated
E	Eating quality	Taste of the fruit (sweet or sour)	Coded class
F	Farmers' preferences	Preferred size, taste, segment colour, peel thickness and shape; preference for many fruits; trait ranking; use of fruit; suitability as planting material	Single choice, yes or no, and ranking

Items are listed in full in Appendix 1.

Observations

Four physical and yield traits were recorded in three replicates for every accession. Fruit weight was measured on a balance and expressed in kilograms. Segments were counted after the fruit was opened. Peel thickness was measured in centimetres, and yield was recorded as the number of fruits per tree. Total soluble solids were read on a hand refractometer and expressed in °Brix, and titratable acidity was determined by titration against standard sodium hydroxide with phenolphthalein as indicator and expressed as per cent citric acid (Kumar et al., 2025). Both were determined in three replicates for every accession. The TSS/acidity ratio was calculated for each replicate. Eating quality was scored as sweet or sour, fruit shape as round or elongated, and segment colour as white, light pink, pink, reddish pink or dark pink.

Data analysis

All seven traits were measured in three replicates, so differences among the 60 accessions were tested by one-way analysis of variance (ANOVA) with accession as the factor (Panse and Sukhatme, 1967). Principal component analysis (PCA) was run on the six measured traits (fruit weight, segments, peel thickness, fruits per tree, TSS and acidity). Each trait was standardised first so that every trait carried equal weight. The TSS/acidity ratio was left out because it is calculated from two traits already included. Components with eigenvalues above 1 were retained (Kaiser, 1960), and loadings of 0.5 or more in absolute value were treated as meaningful (Jolliffe, 2002). Suitability of the data for PCA was checked with the Kaiser–Meyer–Olkin (KMO) measure and Bartlett's test of sphericity. All calculations were done in Python 3 with the NumPy and SciPy libraries.

RESULTS

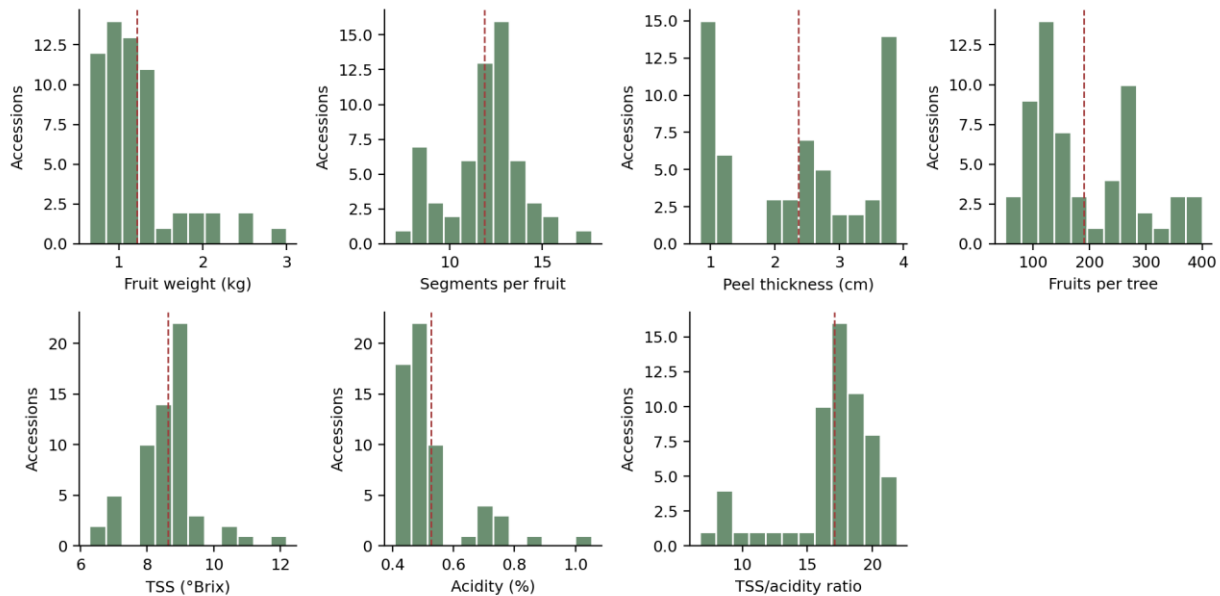
Variation in fruit and yield traits

The accessions varied widely in all four physical and yield traits (Table 2, Fig. 1). Mean fruit weight was 1.22 kg with a median of 1.12 kg. Accession means ran from 0.65 kg to 3.00 kg, the heaviest being accession 34 from Dwarikapur, and only six accessions averaged 2 kg or more. Most accessions were therefore medium-sized, and a few large-fruited trees pulled the mean above the median. Segment number averaged 11.9 and ranged from 7.0 to 17.7, the highest count belonging to accession 41, also from Dwarikapur. Peel thickness varied almost fourfold, from 0.83 to 3.90 cm, with a CV of 47%. Fruits per tree ranged from 50 to 400 with a mean of 190 and a CV of 50%, the largest of any trait. The highest counts were recorded for accession 12 from Chakhaji (400), accession 19 from Muhdda (390) and accession 8 from Rampur Samthu (383).

Table 2. Descriptive statistics for seven traits of 60 pummelo accessions (accession means).

Trait	Mean $\pm$ SD	Range	CV (%)
Fruit weight (kg)	1.22 $\pm$ 0.48	0.65–3.00	39.1
Segments per fruit	11.87 $\pm$ 2.06	7.00–17.67	17.3
Peel thickness (cm)	2.36 $\pm$ 1.11	0.83–3.90	47.0
Fruits per tree	190.0 $\pm$ 94.9	50.0–400.0	50.0
TSS ($^{\circ}$ Brix)	8.62 $\pm$ 0.99	6.27–12.18	11.5
Acidity (%)	0.53 $\pm$ 0.12	0.41–1.05	23.4
TSS/acidity ratio	17.07 $\pm$ 3.47	6.72–21.90	20.3

SD, standard deviation; CV, coefficient of variation of accession means.

**Fig. 1.** Frequency distributions of accession means for seven traits in 60 pummelo accessions. Dashed lines mark the mean.

Accessions differed significantly for all seven traits (Table 3). The *F* values were largest for acidity (214.4), peel thickness (150.4), TSS (141.4) and the TSS/acidity ratio (116.1), and smallest for fruits per tree (45.1), segment number (10.6) and fruit weight (5.6), but every *P* value was below 0.001. In plain terms, accessions differed from one another far more than the three replicates of one accession differed among themselves. The contrast was weakest for fruit weight, because fruits from the same tree varied considerably in size, and strongest for peel thickness and the chemical traits, for which replicate readings agreed closely.

Table 3. Analysis of variance for seven traits of 60 pummelo accessions, each measured in three replicates.

Trait	Mean square, accessions	Mean square, error	<i>F</i>	<i>P</i>
Fruit weight (kg)	0.685	0.122	5.63***	<0.001
Segments per fruit	12.69	1.194	10.62***	<0.001
Peel thickness (cm)	3.704	0.0246	150.43***	<0.001
Fruits per tree	27037.0	600.1	45.05***	<0.001
TSS ($^{\circ}$ Brix)	2.946	0.0208	141.44***	<0.001
Acidity (%)	0.0456	0.0002	214.37***	<0.001
TSS/acidity ratio	36.13	0.311	116.07***	<0.001

One-way ANOVA with accession as the factor; degrees of freedom were 59 for accessions and 120 for error. *** *P* < 0.001.

Biochemical quality

TSS averaged 8.62 $^{\circ}$ Brix and was the least variable trait (CV 11.5%). Accessions ranged from 6.27 to 12.18 $^{\circ}$ Brix, and only four reached 10 $^{\circ}$ Brix: accession 10 from Rampur Samthu (12.18), accession 18 from Muhdda (11.15), accession 2 from Chakhaji (10.61) and accession 36 from Dwariapur (10.47). Titratable acidity had a mean of 0.53% and a median of 0.49%. A little over half of the accessions (33 of 60) had acidity of 0.50% or less, and a handful of sour accessions pulled the upper end out to 1.05%. The TSS/acidity ratio averaged 17.07 with a range

of 6.72 to 21.90. Fifty-one accessions had a ratio of 15 or more and 25 had a ratio of 18 or more. The highest ratio, 21.90, belonged to accession 30 from Rewari (9.22 °Brix, 0.42% acidity), closely followed by accession 42 from Nawada (21.85) and accession 34 from Dwarikapur (21.62).

Comparison of districts and villages

Samastipur and Muzaffarpur accessions were similar for every trait (Table 4). Muzaffarpur accessions had slightly heavier fruit (1.31 against 1.13 kg) and more segments (12.3 against 11.4), whereas Samastipur accessions had more fruits per tree (211 against 170) and slightly higher TSS. These differences were small compared with the spread among accessions within each district, and Fig. 2 shows that the ranges of the two districts overlapped almost completely for every trait.

Table 4. Mean $\pm$ SD of accession means for Samastipur (n = 29) and Muzaffarpur (n = 31) accessions.

Trait	Samastipur	Muzaffarpur
Fruit weight (kg)	1.13 $\pm$ 0.34	1.31 $\pm$ 0.57
Segments per fruit	11.39 $\pm$ 1.70	12.32 $\pm$ 2.28
Peel thickness (cm)	2.25 $\pm$ 1.06	2.48 $\pm$ 1.17
Fruits per tree	211.4 $\pm$ 98.3	170.0 $\pm$ 88.6
TSS (°Brix)	8.85 $\pm$ 1.06	8.41 $\pm$ 0.89
Acidity (%)	0.55 $\pm$ 0.14	0.51 $\pm$ 0.10
TSS/acidity ratio	16.85 $\pm$ 3.35	17.27 $\pm$ 3.62

SD, standard deviation.

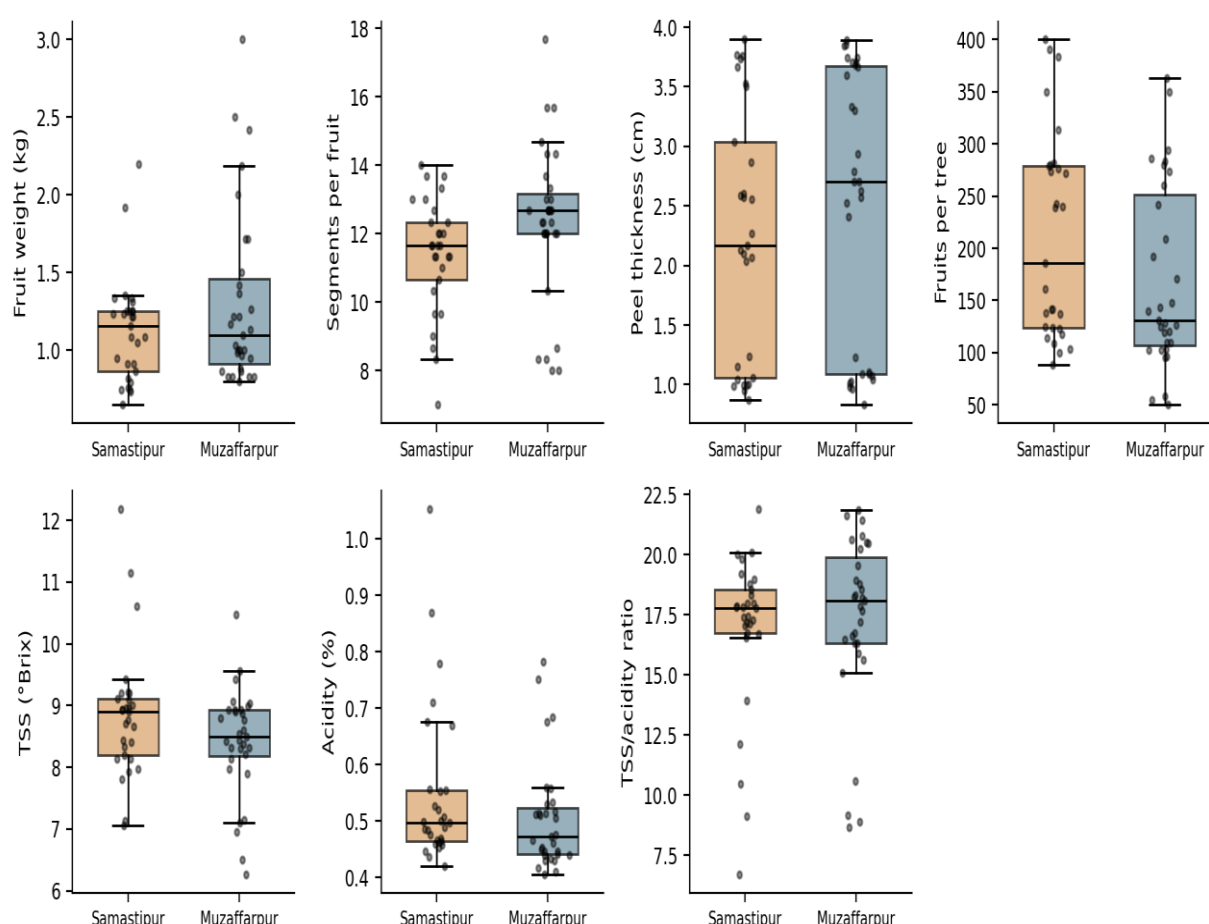


Fig. 2. Distribution of seven traits in pummelo accessions from Samastipur and Muzaffarpur districts. Boxes show the middle half of the accessions and the median, and dots are individual accessions.

Nine villages had three or more accessions (Table 5). Dwarikapur had the highest mean number of segments (13.7), the heaviest fruit (1.54 kg) and the highest mean ratio (18.93), while Rampur Samthi had the fewest segments (10.5) and the lowest mean fruit weight (1.02 kg). Muhdda and Chakhaji had the highest mean number of fruits per tree (258 and 242) and Malpur Agrail the lowest (136), while Malpur Agrail and Simra had the lowest TSS (8.0 °Brix). Because most villages contributed only three to nine trees, these village means are descriptive and should not be taken as fixed village characteristics.

Table 5. Village means for accessions in villages with at least three accessions.

Village	n	Weight (kg)	Segments	Peel (cm)	Fruits/tree	TSS	Acidity	Ratio	District
Dwarikapur	9	1.54	13.70	1.86	182.7	8.79	0.47	18.93	M
Rampur Samthu	9	1.02	10.48	1.88	200.8	8.96	0.57	16.21	S
Malpur Agrail	8	1.15	10.88	2.48	136.4	7.96	0.53	15.91	M
Simra	6	1.17	12.67	2.57	217.4	7.97	0.55	15.47	M
Chakhaji	5	1.20	11.20	2.13	242.1	8.89	0.52	17.26	S
Rewari	5	1.06	12.60	2.86	161.2	8.60	0.48	18.12	S
Lakhanipur	3	1.10	12.67	2.51	215.8	8.83	0.48	18.39	S/M
Morsand	3	1.11	12.00	3.32	175.9	9.01	0.51	17.78	S
Muhdda	3	1.15	11.22	1.40	257.6	9.06	0.69	15.54	S

S, Samastipur; M, Muzaffarpur. TSS in °Brix and acidity in per cent; segment and Fruit/tree in number.

Sweet and sour accessions

Fifty-four accessions (90.0%) were rated sweet and six (10.0%) sour. Sweet and sour accessions differed in chemistry but not in physical traits (Table 6). Sweet accessions averaged 8.82 °Brix against 6.84 °Brix for sour ones, and 0.50% acidity against 0.79%. The ratio showed the widest contrast, 17.98 against 8.86. Fruit weight, segment number, peel thickness and fruits per tree were similar in the two classes. TSS divided the two classes almost cleanly. All six sour accessions had 7.2 °Brix or less (6.27 to 7.15), and the sweet accessions had 7.1 °Brix or more, so the only overlap was accession 40, which was rated sweet at 7.1 °Brix with 0.43% acidity. The ratio was a clearer guide: every sour accession had a ratio below 11 (6.72 to 10.58), while only three sweet accessions did not reach 15. Accession 6 (ratio 10.5, acidity 0.87%) and accession 9 (12.1) were rated sweet at ratios close to the sour group, and accession 11 was rated sweet at 13.9.

Table 6. Mean $\pm$ SD of accession means for sweet (n = 54) and sour (n = 6) accessions.

Trait	Sweet	Sour
TSS (°Brix)	8.82 $\pm$ 0.83	6.84 $\pm$ 0.37
Acidity (%)	0.50 $\pm$ 0.08	0.79 $\pm$ 0.14
TSS/acidity ratio	17.98 $\pm$ 2.19	8.86 $\pm$ 1.25
Fruit weight (kg)	1.23 $\pm$ 0.48	1.15 $\pm$ 0.53
Segments per fruit	11.88 $\pm$ 2.16	11.78 $\pm$ 0.78
Peel thickness (cm)	2.37 $\pm$ 1.14	2.33 $\pm$ 0.85
Fruits per tree	187.8 $\pm$ 95.8	209.8 $\pm$ 92.5

SD, standard deviation. The sour group is small, so its means are only approximate.

Principal component analysis

Most of the six traits were only weakly related to each other, and this showed in the PCA. The KMO value was 0.49, just below the level of 0.50 usually taken as the minimum, and Bartlett's test was not significant ($\chi^2 = 19.3$, 15 df, $P = 0.201$). The PCA is therefore best read as a descriptive summary and not as evidence of strong underlying structure. Three components had eigenvalues above 1 (1.49, 1.34 and 1.04) and together explained 64.5% of the variation (Table 7, Fig. 3). The first two explained 24.8% and 22.4%, so the variation was spread almost evenly and no single component dominated. PC1 set fruit size and sweetness against acidity. Fruit weight (0.70), segment number (0.63) and TSS (0.54) loaded positively and acidity (−0.45) negatively, so accessions with high PC1 scores bore large, many-segmented, sweet and less acidic fruit. PC2 was a tree-level axis, with peel thickness (0.79) opposed to fruits per tree (−0.76): trees with thick-peeled fruit tended to carry fewer fruits. PC3 was mainly a chemistry axis, in which TSS (0.61) stood against acidity (−0.46) and segment number (−0.52).

Table 7. Eigenvalues, variation explained and trait loadings for the first three principal components of six traits in 60 pummelo accessions.

Trait	PC1	PC2	PC3
Fruit weight	0.70	0.02	-0.38
Segments per fruit	0.63	0.02	-0.52
Peel thickness	-0.06	0.79	-0.12
Fruits per tree	0.30	-0.76	0.18
TSS	0.54	0.16	0.61
Acidity	-0.45	-0.34	-0.46
Eigenvalue	1.49	1.34	1.04
Variation explained (%)	24.8	22.4	17.4
Cumulative variation (%)	24.8	47.1	64.5

PCA on standardised traits. Loadings show how strongly each trait contributes to a component. The TSS/acidity ratio was excluded because it is derived from TSS and acidity.

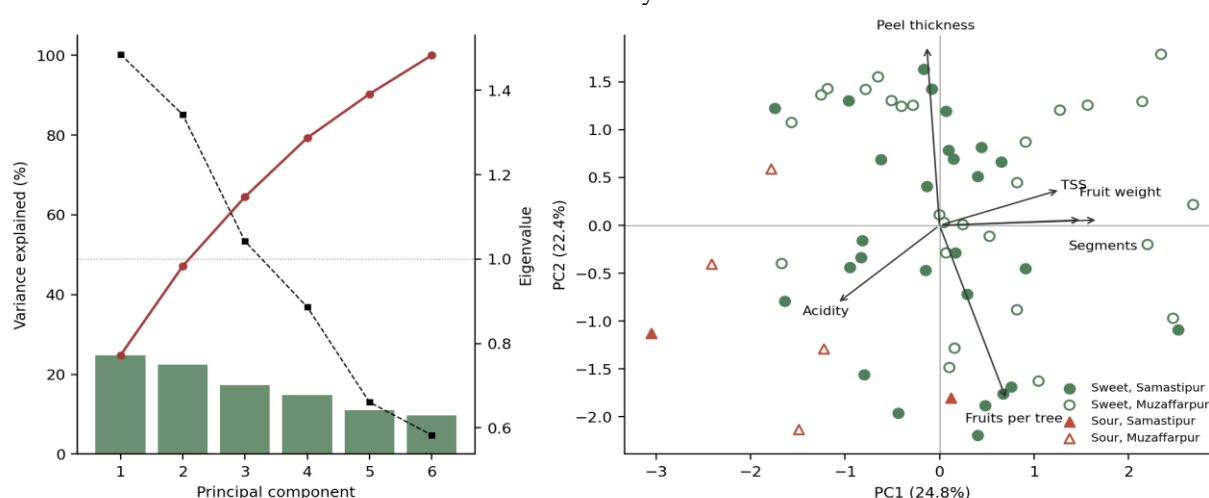


Fig. 3. Principal component analysis of six traits. Left, scree plot with eigenvalues (dashed) and cumulative variation (red); right, biplot of PC1 and PC2 with accessions marked by eating quality and district.

Sweet and sour accessions lay apart on PC1 (mean scores 0.18 and -1.64) and very clearly on PC3 (0.21 and -1.90), and less so on PC2 (0.11 and -1.03). In the biplot the sour accessions lie mostly to the left, in the direction of the acidity arrow, while accessions from the two districts are thoroughly mixed. The highest PC1 scores belonged to accessions 34 (2.69), 18 (2.52) and 41 (2.47), and the lowest to accessions 7 (-3.05), 54 (-2.41) and 31 (-1.78). Accessions 12 and 19, the two heaviest bearers, were among the lowest on PC2 (-2.19 and -1.96).

Fruit shape and segment colour

Fifty-five accessions (91.7%) bore round fruit and five (8.3%) elongated fruit. Pink was the commonest segment colour (23 accessions, 38.3%), followed by light pink (14, 23.3%), reddish pink (10, 16.7%), dark pink (7, 11.7%) and white (6, 10.0%). The two districts had a similar mix of eating quality, fruit shape and segment colour (Table 8, Fig. 4), although the groups are small and the counts should be read as approximate.

Table 8. Number of accessions in each class of three qualitative traits, by district.

Trait and class	Samastipur	Muzaffarpur	Total (%)
Eating quality: sweet	27	27	54 (90.0)
Eating quality: sour	2	4	6 (10.0)
Shape: round	26	29	55 (91.7)
Shape: elongated	3	2	5 (8.3)
Colour: white	3	3	6 (10.0)
Colour: light pink	8	6	14 (23.3)
Colour: pink	9	14	23 (38.3)
Colour: reddish pink	4	6	10 (16.7)
Colour: dark pink	5	2	7 (11.7)

Counts are numbers of accessions.

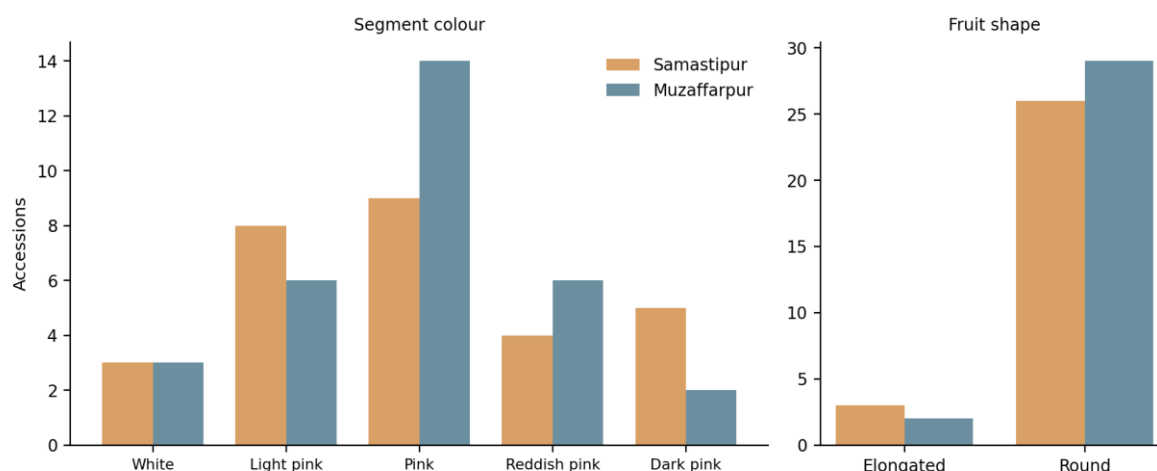


Fig. 4. Number of accessions by segment colour and fruit shape in Samastipur and Muzaffarpur districts.

Promising accessions

To pick accessions worth multiplying, the study screened for TSS of at least 8.9 °Brix, acidity of 0.50% or less and fruit weight of at least 1 kg. Eight accessions met all three conditions (Table 8). Six came from Muzaffarpur and two from Samastipur. Accession 34 combined the heaviest fruit (3.00 kg) with a ratio of 21.62 and a thin peel (1.09 cm), although it bore only about 100 fruits per tree. Accession 41 had the most segments (17.7) with a thin peel, and accessions 33 and 21 bore 363 and 350 fruits per tree, among the highest counts in the survey. Accession 5 offered large fruit (2.19 kg) and a good ratio but a thicker peel (3.33 cm). Accession 30 (ratio 21.90, the highest) missed the list because its fruit weighed only 0.73 kg, and accessions 42, 43 and 55 each narrowly missed one limit (TSS of 8.87 °Brix for 42, acidity of 0.503% for 43 and TSS of 8.80 °Brix for 55). Fig. 5 shows how the shortlisted accessions sit among the others.

Table 9. Accessions with TSS ≥ 8.9 °Brix, acidity $\leq 0.50\%$ and fruit weight ≥ 1 kg, ranked by TSS/acidity ratio.

Acc.	Village	Weight (kg)	Segments	Peel (cm)	Fruits/tree	TSS	Acidity	Ratio	Segment colour
34	Dwarikapur	3.00	12.7	1.09	103.7	9.00	0.42	21.62	Reddish pink
41	Dwarikapur	1.50	17.7	1.01	260.3	8.93	0.43	20.77	Reddish pink
5	Simra	2.19	12.7	3.33	370.0	9.07	0.44	20.62	Light pink
33	Dwarikapur	1.22	12.3	1.23	363.3	9.03	0.44	20.51	Reddish pink
51	Bochha	1.42	13.0	2.62	128.7	9.43	0.46	20.47	Light pink
52	Malpur Agrail	1.03	8.0	2.79	50.0	8.94	0.44	20.24	Pink
21	Lakhanipur	1.22	11.3	1.06	350.0	9.20	0.50	18.53	Dark pink
23	Lakhanipur	1.25	13.0	3.77	239.0	8.97	0.49	18.32	Dark pink

Values are accession means. TSS in °Brix and acidity in per cent. The screening limits were chosen for description and are not formal selection indices.

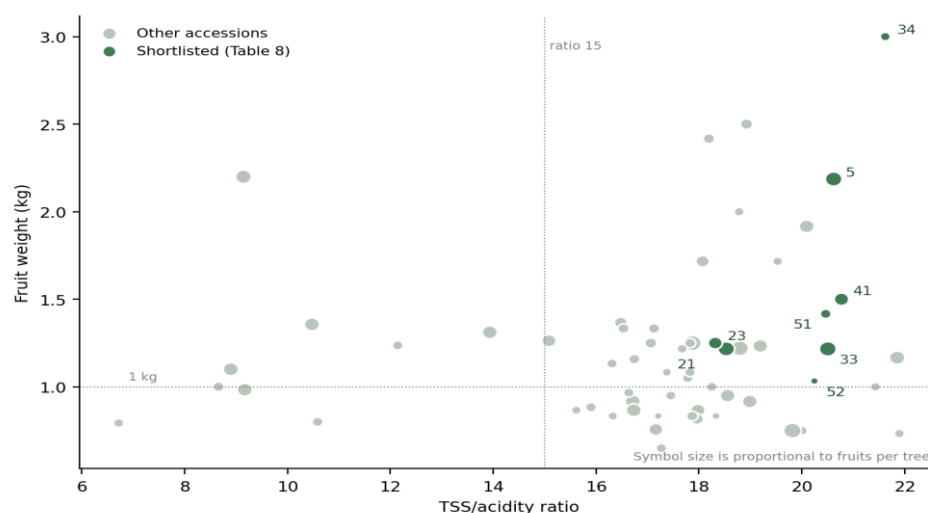


Fig. 5. Fruit weight against TSS/acidity ratio for 60 pummelo accessions. Shortlisted accessions (Table 8) are numbered, and symbol size is proportional to fruits per tree.

DISCUSSION

The pummelo accessions of Samastipur and Muzaffarpur are far from uniform. Fruit weight differed more than fourfold among accessions, peel thickness almost fourfold, and fruits per tree eightfold. The ANOVA confirms that these differences among trees are real and not an effect of measurement scatter, since they exceeded the differences among replicates for every trait. This is the pattern expected where trees are raised mostly from seed and exchanged informally, since pummelo is cross-pollinated and seedlings of heterozygous parents differ from their parents. Wide variation in fruit traits is also reported for pummelo populations elsewhere (Paudyal and Haq, 2008), so the Bihar material is not unusual in this respect, although it has not been described before. Similarly, considerable quantitative variation has been reported among Indian pummelo germplasm, particularly for fruit weight, seed content, juice characteristics and other fruit attributes (Dubey et al., 2019). More recently, Ahmed et al. (2019) demonstrated considerable morphological variation among Indian pummelo cultivars and showed that fruit number per tree and rag percentage made major contributions to overall diversity. Thus, the broad phenotypic differentiation observed in the present Bihar population is consistent with the wider pattern of diversity reported in *Citrus maxima*.

Fruit number is a composite productivity trait influenced not only by genotype but also by tree age, canopy size, flowering intensity, fruit set, crop load, water availability, nutrition and management practices. Consequently, the present values describe the productivity of the individual farmer-managed trees under their existing conditions rather than their intrinsic genetic yield potential. Although fruit weight differed significantly among accessions, the relatively low *F* statistic compared with several other traits suggests greater within-tree variability relative to the between-accession effect. Thus, selection based on only a small number of fruits per tree may be less reliable than selection based on traits showing greater repeatability within a tree. This interpretation is supported by previous pummelo characterization studies in which quantitative fruit characters, including fruit weight, contributed strongly to accession differentiation but were also associated with substantial phenotypic variability (Dubey et al., 2019). In contrast, peel thickness, TSS and titratable acidity showed comparatively greater consistency among replicate measurements. These traits may therefore provide more useful preliminary descriptors for accession characterization, although multi-year and multi-location evaluation would be necessary before making selection decisions. The importance of fruit quality traits in distinguishing pummelo genotypes has also been demonstrated in Indian and Chinese germplasm collections, where TSS, acidity, peel thickness, fruit size and other quality parameters contributed substantially to cultivar differentiation (Ahmed et al., 2019; Chen et al., 2021).

The two districts showed broadly comparable phenotypic patterns. Their means differed only modestly, the ranges overlapped substantially, and similar proportions of sweet, sour, round, elongated and differently coloured fruits were observed. This similarity suggests that the sampled trees from the two districts may belong to a broadly related regional pummelo population rather than representing sharply differentiated geographical groups, this might be due to exchange of plant material between these two neighbouring districts.

The biochemical characteristics revealed a population with moderate soluble-solids content but generally low titratable acidity. The mean TSS of 8.6 °Brix indicates that most accessions were not exceptionally high in soluble solids, with only four accessions reaching 10 °Brix or higher. In contrast, 33 accessions had acidity ≤0.50%, indicating that low acidity was a prominent characteristic of the population. This distinction is important because perceived citrus flavour is determined by the balance between sugars and organic acids rather than by TSS alone. Citrus quality assessment commonly uses the soluble-solids-to-acidity ratio as an indicator of maturity and flavour balance, although appropriate thresholds vary among species, cultivars and production environments (Lado et al., 2018; Goulart et al., 2021). The comparatively greater coefficient of variation for acidity (23%) than for TSS (11.5%) further suggests that acidity contributed more strongly to the differentiation of the accessions for internal fruit quality. The inverse relationship between acidity and the TSS:acidity ratio observed in the present material indicates that variation in perceived sweetness was influenced substantially by changes in acidity. This interpretation is biologically reasonable because the ratio integrates the two principal components contributing to citrus taste. Studies of pummelo cultivars have similarly demonstrated substantial variation in both soluble solids and organic acids and have identified the sugar–acid balance as an important determinant of fruit quality (Chen et al., 2021). The identification of accessions combining relatively high TSS with low acidity is therefore of particular interest for selection. Accession 10 from Rampur Samthi, with 12.18 °Brix and 0.71% acidity, represents a high-TSS phenotype, whereas accessions 30 and 34, each with 0.42% acidity, represent low-acidity phenotypes. More importantly, these examples indicate that the two traits do not necessarily change in parallel. The sensory observations provide additional support for this interpretation. TSS generally differentiated sweet and sour accessions, but the TSS:acidity ratio provided a clearer separation, with sour accessions occurring predominantly at lower ratios and sweet accessions generally showing higher ratios. Accession 40 is particularly informative because it was classified as sweet despite having only 7.1 °Brix; its low acidity of 0.43% resulted in a TSS:acidity ratio of 16.5. This indicates that sensory perception was not governed by soluble solids alone and supports the importance of sugar–acid balance in determining perceived flavour. Comparable relationships between physicochemical characteristics and sensory acceptability have been documented in pummelo and pummelo-derived citrus materials (Ahmed et al., 2019; Singh et al., 2024). The apparently anomalous classification of accessions 6, 9 and 11 as sweet despite relatively low TSS:acidity ratios may have several explanations. Differences in fruit maturity at sampling, individual variation among fruits, cultivar-specific taste characteristics,

and the limitations of a binary sensory classification could all contribute. Citrus maturity is known to affect both soluble solids and acidity, and maturity indices vary considerably among citrus species and cultivars (Lado et al., 2018; Goulart et al., 2021). Therefore, the sensory observations in the present survey should be regarded as preliminary rather than as evidence for definitive TSS or TSS:acidity thresholds. Future evaluation should use fruits harvested at a standardized physiological maturity and a trained or semi-trained sensory panel employing a hedonic scale.

The three components of PCA explained approximately 65% of the total variation, indicating that the observed diversity was distributed across several independent trait combinations rather than being controlled by a single dominant axis. This agrees with previous characterization studies in which multiple principal components were required to describe the morphological diversity of pummelo germplasm (Luro et al., 2025; Ahmed et al., 2019). PC1 grouped large, many-segmented, sweet fruit against acidity and set the sour accessions apart, whereas PC2 isolated the contrast between peel thickness and bearing. The lack of clear separation of Samastipur and Muzaffarpur accessions in the PCA space further supports the observation that geographical origin was not the dominant source of variation in the sampled population. Similar findings have been reported for pummelo germplasm, where PCA has successfully identified combinations of morphological and fruit-quality traits responsible for accession differentiation (Dubey et al., 2019). However, the KMO value of 0.49 indicates weak sampling adequacy or limited common variance among the variables. Therefore, PCA in this study should primarily be regarded as an exploratory tool for visualizing relationships among accessions rather than as evidence of a strong latent trait structure. In particular, the component loadings should not be interpreted as proof that the traits are biologically controlled by common factors. A larger dataset incorporating additional fruit, tree and biochemical descriptors, together with replicated observations over multiple seasons, would provide a stronger basis for developing a selection index. The accessions identified in Table 9 should consequently be regarded as promising candidate selections rather than confirmed superior genotypes. Vegetative propagation through budding, grafting or air layering would be appropriate for conserving selected phenotypes because it allows the desirable characteristics of individual trees to be maintained clonally.

CONCLUSION

The present study demonstrated substantial diversity among pummelo accessions of Samastipur and Muzaffarpur, with significant differences among accessions for fruit weight, segment number, peel thickness, fruits per tree, TSS, acidity and TSS ratio. Most of the observed variation occurred among individual trees rather than between districts or villages, indicating considerable within-population diversity. Approximately 90% of the accessions were classified as sweet, with about 7.2 °Brix TSS or a TSS ratio of around 11 providing a preliminary separation between sweet and sour fruits in the present population. The TSS ratio was influenced largely by variation in acidity, suggesting that acidity and sugar contribute independently to eating quality. PCA further indicated that fruit size, peel and bearing characteristics, and biochemical traits represented largely distinct dimensions of variation. Eight accessions, particularly 34, 41, 5 and 33, showed promising combinations of fruit size and eating quality and therefore merit vegetative multiplication and multi-season evaluation. Pigment profiling of contrasting pulp-colour types and structured sensory evaluation would further strengthen the selection of superior accessions. Although the study provides a useful baseline of pummelo diversity in Bihar, non-standardized fruit maturity, farmer-managed conditions and unequal sampling limited the separation of genetic and environmental effects. Therefore, clonal evaluation under uniform conditions, multi-season validation and molecular characterization are necessary to confirm stable and genetically superior accessions for conservation, breeding and commercial utilization.

ACKNOWLEDGEMENTS

This research was funded by Dr Rajendra Prasad Central Agricultural University funded project “Assessing phytochemical composition of pummelo, an underutilized fruit crop and its suitability for making value-added products”.

CONFLICT OF INTEREST

The author declares no conflict of interest.

REFERENCES

1. Ahmed, S., Rattanpal, H. S., & Singh, G. (2019). Diversity, characterization and evaluation in pummelo (*Citrus maxima* Merr.) cultivars using SSR markers and quality parameters. *Indian Journal of Genetics and Plant Breeding*, 79(3), 594–605. <https://doi.org/10.31742/IJGPB.79.3.9>
2. Chen, Y., et al. (2021). Fruit physiology and sugar-acid profile of 24 pomelo (*Citrus grandis* (L.) Osbeck) cultivars grown in subtropical region of China. *Agronomy*, 11, 2393.
3. Cheong, M. W., Liu, S. Q., Zhou, W., Curran, P., & Yu, B. (2012). Chemical composition and sensory profile of pomelo (*Citrus grandis* (L.) Osbeck) juice. *Food Chemistry*, 135(4), 2505-2513.
4. Luro, F., Marchi, E., Costantino, G., Paoli, M., & Tomi, F. (2025). Diversity of pummelos (*Citrus maxima* (Burm.) Merr.) and grapefruits (*Citrus x aurantium* var. *paradisi*) inferred by genetic markers, essential oils composition, and phenotypical fruit traits. *Plants*, 14(12), 1824.

5. Dubey, A. K., Sharma, R. M., Awasthi, O. P., Sharma, N., & Kholia, A. (2019). Morphological characterization of Indian pummelo (*Citrus maxima*). *The Indian Journal of Agricultural Sciences*, 89(9).
6. Goulart, R. R., et al. (2021). Emerging approaches to determine maturity of citrus fruit. *Critical Reviews in Food Science and Nutrition*, 62(19).
7. Jolliffe, I. T. (2002). *Principal component analysis* (2nd ed.). Springer.
8. Kaiser, H. F. (1960). The application of electronic computers to factor analysis. *Educational and Psychological Measurement*, 20, 141–151.
9. Kumar, D., Singh, H., Khan, M. G., & Kumari, A. (2025). Enhancing consumer's acceptability of pomelo juice (*Citrus grandis* Linn.) through blending with sweet orange juice. *Agriculture Association of Textile Chemical and Critical Reviews Journal*, 511–516.
10. Ladaniya, M. S. (2008). *Citrus fruit: Biology, technology and evaluation*. Elsevier.
11. Lado, J., et al. (2018). Key determinants of citrus fruit quality: Metabolites and main changes during maturation. *Scientia Horticulturae*, 233, 238–248. <https://doi.org/10.1016/j.scienta.2018.01.055>
12. Morton, J. F. (1987). Pummelo. In *Fruits of warm climates* (pp. 147–151). Julia F. Morton.
13. Panse, V. G., & Sukhatme, P. V. (1967). *Statistical methods for agricultural workers* (2nd ed.). Indian Council of Agricultural Research.
14. Paudyal, K. P., & Haq, N. (2008). Variation of pomelo (*Citrus grandis* (L.) Osbeck) in Nepal and participatory selection of strains for further improvement. *Agroforestry Systems*, 72(3), 195–204. <https://doi.org/10.1007/s10457-007-9088-z>
15. Ranganna, S. (1986). *Handbook of analysis and quality control for fruit and vegetable products* (2nd ed.). Tata McGraw-Hill.
16. Saunt, J. (2000). *Citrus varieties of the world* (2nd ed.). Sinclair International.
17. Singh, H. (2022). Pummelo: A Potential Underutilized Nutraceutical Crop for Multiple Health Benefits. *International Journal of Plant & Soil Science*.
18. Wu, G. A., Terol, J., Ibanez, V., López-García, A., Pérez-Román, E., Borredá, C., ... & Talon, M. (2018). Genomics of the origin and evolution of Citrus. *Nature*, 554(7692), 311–316.

APPENDIX 1. STRUCTURED QUESTIONNAIRE

Pummelo participatory survey, Samastipur and Muzaffarpur districts, Bihar, 2020 and 2021. Sections A to E are the field observation record and section F is the farmer interview. Tick or circle one response unless stated.

No.	Item	Response
A	Identification	
A1	Accession (tree) number	Serial number, 1 to 60
A2	Village	Name of village
A3	District	Samastipur / Muzaffarpur
A4	Date of survey	Day, month, year (2020 or 2021)
B	Fruit characters (record three replicates, R1 to R3)	
B1	Fruit weight	kg, to the nearest 0.01 kg on a balance
B2	Number of segments per fruit	Count after opening the fruit
B3	Peel thickness	cm
B4	Fruit shape	Round / Elongated
B5	Colour of segments	White / Light pink / Pink / Reddish pink / Dark pink
C	Tree yield	
C1	Number of fruits on the tree (Current year count, and last two year count as replicate)	Count
D	Fruit chemistry (three replicates, R1 to R3)	
D1	Total soluble solids (TSS)	°Brix, hand refractometer
D2	Titrateable acidity	% citric acid, titration with NaOH
D3	TSS/acidity ratio	Calculated as D1 divided by D2
E	Eating quality	
E1	Taste of the fruit, as judged on tasting	Sweet / Sour
F	Farmers' preferences	
F1	Which fruit size do you prefer?	Small (under 1 kg) / Medium (1 to 2 kg) / Large (over 2 kg)
F2	Which taste do you prefer?	Sweet / Sweet-sour / Sour
F3	Which colour of segments do you prefer?	White / Light pink / Pink / Reddish pink / Dark pink / No preference
F4	Which peel thickness do you prefer?	Thin (under 1.5 cm) / Medium (1.5 to 3 cm) / Thick (over 3 cm)
F5	Which fruit shape do you prefer?	Round / Elongated / No preference
F6	Would you prefer a tree with many fruits even if the fruits are smaller?	Yes / No