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RESEARCH ARTICLE

Diversity in morphological and some chemical traits of *Aegle marmelos* (L.) Correa germplasm explored from Achanakmar-Amarkantak Biosphere Reserve, Chhattisgarh, India

Pankaj Kumar Kannaujia · Sunil Gomashe · Amarkant Kushawaha · Rakesh Bhardwaj · Eldho Varghese · Sakharam Kale · Pavan Kumar Malav · Ravi Kishore Pamarthi · Kailash Chandra Bhatt · Sudhir Pal Ahlawat · Praveen Kumar Singh

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Abstract Bael is an important indigenous minor fruit crop of India. Considering its importance, exploring diversity and germplasm collection was conducted in the Achanakmar-Amarkantak Biosphere Reserve (ABR), spreading to three districts of Chhattisgarh state. A total of 35 diverse samples of bael germplasm were collected, of which 21 distinct types were analyzed for various quantitative and qualitative traits. A large amount of variability was recorded in the shape, size, shell thickness, and content of health-promoting bioactive compounds. The highest fruit weight (2.54 kg) was observed for accession IC0645501 followed by accession IC0645463 (1.40 kg), while accession IC0645480 exhibited the lowest fruit weight (77.65 g). IC0645487 showed the highest total soluble solids (TSS) content (54.16°Brix) and IC0645499 had the lowest TSS content (26.81°Brix). Maximum carotenoid content was recorded in IC0645485 (6.21 µg/ g) and lowest in IC0645501 (0.62 µg/ g) showing 9.5 fold variation. FRAP antioxidant activity ranged from 0.095 to 0.202 mg/ g GAE and total phenolics varied from 1.22 (IC0645501) to 2.27 mg/g GAE (IC0645480). Distinct accessions for better fruit quality traits like large size, high pulp weight ratio, low seeds etc. have great potential for breeding and improvement in bael.

Keywords Bael · Carotenoid content · Conservation · Indigenous collection number · Genebank · Shell thickness · Total soluble solids

P. K. Kannaujia · R. Bhardwaj · P. K. Malav · R. K. Pamarthi · K. C. Bhatt · S. P. Ahlawat · P. K. Singh (✉)
ICAR, National Bureau of Plant Genetic Resources, New Delhi, Delhi 110012, India
e-mail: praveen.singh1@icar.gov.in

S. Gomashe
ICAR, National Bureau of Plant Genetic Resources, Akola, Maharashtra 444104, India

A. Kushawaha
ICAR, Central Institute for Subtropical Horticulture, Lucknow, Uttar Pradesh 226101, India

E. Varghese
ICAR, Central Marine Fisheries Research Institute, Kochi,, Kerala 682018, India

S. Kale
ICAR, National Institute of Secondary Agriculture, Ranchi, Jharkhand 834010, India

Introduction

Bael [*Aegle marmelos* (L.) Correa] is one of India's major native minor fruit crops. It is a member of the citrus (Rutaceae) family. It is also known as a Belgiri, Beli, Bengal quince, Bel Kham, Biliva phal, and Vilwa in various languages and regions (Purohit and Vyas 2004). It is distributed in India, Bangladesh, China, Cambodia, Fiji, Indonesia, Laos, Myanmar, Malaysia, Nepal, Pakistan, Philippines, Sri Lanka,

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Thailand, Tibet and Vietnam, etc., and other south-eastern Asian countries (Neeraj and Johar 2017; Sarkar et al. 2020a, b). In India, it mainly occurs in Indo-Gangetic plains of northern states, Sub-Himalayan tracts, North-Eastern states, arid and deciduous forests of the southern and central peninsular regions up to 1300 msl (Singh et al. 2011; Pradheep et al. 2021; Debbarma and Hazarika 2023). Although as a sub-tropical tree, it is remarkably adapted and thrives very well under tropical, dry, and semi arid conditions (Singh et al. 2018). It is cultivated mainly in orchards, field bunds, and home gardens. The Bael tree grows slowly, reaching a height of 12–15 m. It has a slender trunk and occasionally spreads spiky branches and is deciduous (Panda et al. 2014). Bael fruits often have globose shapes with hard, woody skin shells and grey or yellowish fruit skin colors. Fruit maturity starts from March onwards up to June in various regions of the country (Baliga et al. 2013; Hazra et al. 2019). The fruit's mucilaginous yellow or orange color pulp has numerous seeds. Variations in the size and shape of fruits provide adequate possibilities for selecting genotypes for genetic improvement (Hiwale 2015). Therefore, identifying suitable genotypes was crucial to enhancing bael products' yield, productivity, and post-harvest quality. The ancient Sanskrit medical literature Charaka-Samhita thoroughly describes its various therapeutic qualities (Aiyer 1956). Almost every part of bael tree can be used in different ayurvedic and ethnic medicines viz., antidiarrheal, antidysentery, antipyretic, antidiabetes, antibacterial, antiviral, anti-fungal, anti-cancer, analgesic, anti-microbial and anti-helminthic. Mostly fruit pulp and leaves contain several bioactive chemicals, including total carotenoids, phenolics, tannins, coumarins, flavonoids, and terpenoids, which are primarily responsible for their therapeutic qualities (Maity et al. 2009; Lambole et al. 2010). Several compounds have also been isolated from the various parts of the tree and these are mainly aegelin, cineole, citral, citronellol, dictamnine, lupeol, luvangetin, skimmianine, marmelosin, psoralen, rutin, scopoletin, tembamide and xanthotoxin (Maity et al. 2009; Sarkar et al. 2020a). Its fruits are also an excellent source of protein, carbohydrates, minerals and vitamins (Manandhar et al. 2018; Rana 2023). Bael is an abundant source of vitamin B2, which is also known as riboflavin (Pathirana et al. 2020). The pulp of ripened fruit has a higher amount of marmelosin compound as compared to other portions of the bael tree (Gurjar et al. 2019).

In the Indian sub-continent, mature bael fruits are primarily consumed as fresh, ripened pulp in the form of sharbat or shake. By adding traditional sweets, pulp is also commercially processed to make various products, such as jam, syrup, and pudding. Various processed items, viz., candy, nectar, pulp powder, slabs, squash, and toffee, can be made from bael fruit (Singh and Chaurasiya 2014). Due to its capacity to treat constipation and its fragrant, calming, and laxative qualities, it is highly liked in tropical and desert regions. The most recent technological advancement, dehydrated bael powder was created primarily for its superior ability to retain pharmacological ingredients and longer storage than fresh fruit pulp (Gurjar et al. 2019). These days, a number of processing businesses have developed and marketed RTS (ready to serve) beverages prepared from bael fruit pulp as a component of mixed fruit juice. The processing industry requires bael cultivars that can provide pleasing fruit quality characteristics like higher pulp recovery, few numbers of seeds, thinner shells, high TSS, and higher medicinal and nutritional contents for commercial purposes.

This exploration was conducted to study the variability in situ, characterize and quantify the variation available in different traits and identify distinct accessions. Additionally, fruit quality characteristics were also assessed to identify potential accessions possessing important attributes. Hence, the morphological and biochemical properties of fruits from different bael accessions were analyzed.

Materials and methods

Conservation status and gaps

The ICAR-NBPGR, New Delhi, is a nodal institute for managing plant genetic resources (PGR) of food, fodder and agri-horticultural crops in India and conducting explorations across the country based on gaps in the gene banks through the National Exploration Plan (NEP). Prior to planning explorations and germplasm collections for any crop, gap analysis is essential to prevent duplication in the gene bank and wastage of resources. The perusal of the database revealed that so far, a sum of 562 accessions of

Bael genotypes has been collected, and Indigenous Collection (IC) numbers have been issued at ICAR-NBPGR. These accessions have been conserved in the form of live plants in the field genebanks (FGB) at different national active germplasm sites, institutes, and centres, and their seeds in cryopreservation facilities. Bael accessions conserved in the genebank are from 23 states, viz., Andaman & Nicobar Island (15), Andhra Pradesh (06), Bihar (25), Chhattisgarh (71), Delhi (04), Gujarat (28), Haryana (02), Himachal Pradesh (12), Jharkhand (179), Karnataka (05), Kerala (07), Madhya Pradesh (13), Maharashtra (15), Odisha (34), Rajasthan (26), Tamilnadu (05), Telangana (10), Uttar Pradesh (60), Uttarakhand (06), West Bengal (25) etc. Maximum accessions are from Jharkhand state, and the least are from Arunachal Pradesh, Assam, and Meghalaya (one accession each). A perusal of the literature and these facts revealed that the states of Gujarat, Haryana, Himachal Pradesh, Karnataka, Kerala, Madhya Pradesh, Maharashtra, Odisha, Tamilnadu, Telangana and north-eastern states are under-represented and need to be explored in consultation with the state horticultural department and agricultural universities and institutions.

Collection sites and sampling strategy

Exploration was conducted in the Achanakmar-Amarkantak Biosphere Reserve (ABR), spreading to three districts of Chhattisgarh (Bilaspur, Mungeli, and Gaurela-Pendra-Marwahi). The ABR also extends to Anuppur and Dindori districts of Madhya Pradesh. Since Biosphere Reserves are designated as protected places under the Wildlife Protection Act of 1972, advance permission was obtained from the Forest Department for the exploration and collection of germplasm. This exploration was conducted in three districts of Chhattisgarh from April to May 2022, with the support of the Biosphere Reserve staff. A standard procedure of sampling and germplasm collection was followed as per the NBPGR guidelines (2016). Ripened fruits were harvested to extract mature seeds for conservation in the genebank and conduct quality tests of pulp. A total of 35 sites/trees were located in the surveyed area, and their passport data consisting of information on the location (latitude, longitude, and altitude), habitat, biological status, diseases, pests, soil and land characteristics, etc., was recorded. The collection sites are shown in Fig. 1 to collect distinct genotypes with a variety of shapes and sizes of fruits (Fig.2) in the buffer and transition zone.

Parameters evaluated

Irrespective of the mode of pollination, generally, fruit trees are highly variable; therefore, they are clonally propagated and treated as individual genotypes. In the present study, out of 35 accessions, 21 distinct accessions were taken as different genotypes and analyzed for various postharvest fruit quality traits i.e. fruit weight, fruit dimensions, seed count, pulp recovery, TSS, ascorbic acid, carotenoid content, total flavonoids, total phenolics, FRAP antioxidant capacity and total sugars. Digital vernier callipers were used to measure the bael fruits' length, width, and shell thickness and the results were recorded in millimetres (mm). Weight (g) of five mature and ripened fruits was recorded by an electronic weighing balance and the average was calculated. To determine the average number of seeds per fruit, the total number of seeds in each of the five fruits was counted. Dry matter content was determined as per the method of AOAC, 934–01 (AOAC 2016). Bael pulp was extracted by first splitting the fruit into half and then scooping out the entire contents. The seeds and fibrous material were separated from the pulp using forceps.

To estimate the TSS content, scooped pulp was macerated with the help of a pestle and mortar, and the bael juice was extracted in a beaker. Four to five droplets of pulp extract were added to the digital refractometer's prism surface, and the result was recorded in °Brix. The Folin phenol reagents method of Jagota and Dani (1982) was used to assess bael fruit pulp ascorbic acid (AA) content. 1 g fresh and homogenized pulp was mixed with 2 ml of 4.5% m-Phosphoric acid, and then volume was made up to 5 ml with 3% m-Phosphoric acid. Sample was centrifuged at 10,000 rpm for 10 min, supernatant was collected and residue was re-extracted with 5 ml of 3% m-phosphoric acid, centrifuged and supernatant were pooled. Vitamin C estimation was performed using the Folin-phenol reagent method. The absorbance at 630 nm was measured using a UV–vis spectrophotometer and the results were represented as mg/g of fresh weight. Total carotenoids were determined as

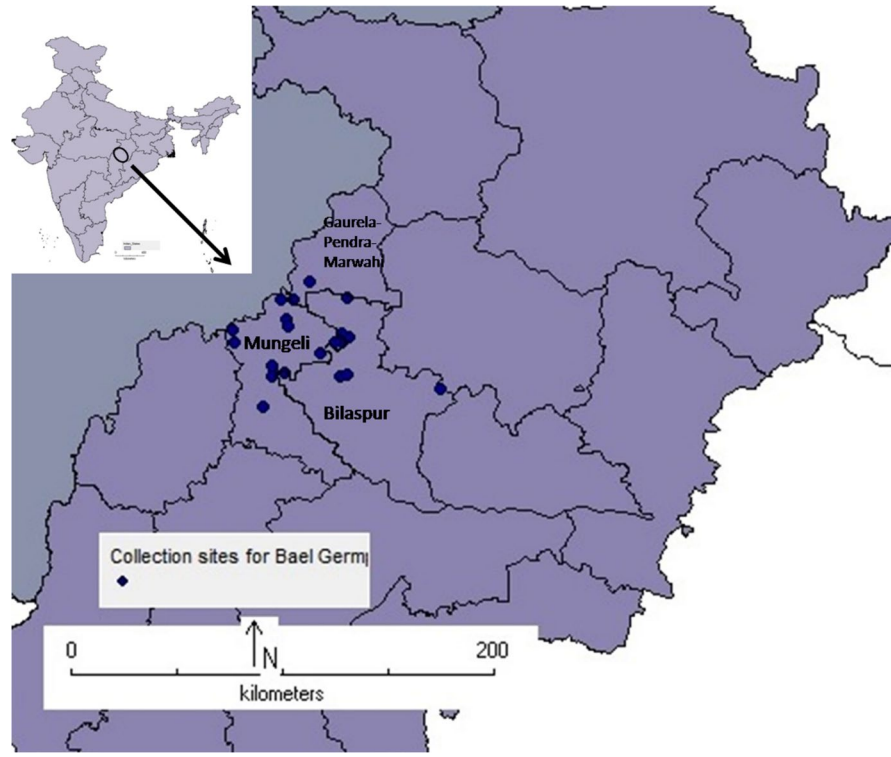

Fig. 1 Germplasm collection sites from Achanakmar- Amarkantak Biosphere Reserve

per the method of Lichtenthaler (1987) with some modifications. For this 2 g of bael fruit pulp was crushed in acetone, and extracted through pressure filter assembly, till the washings became colourless. Then the extracted solution was poured into a separating funnel. To it, petroleum ether and a small amount of sodium sulphate solution were added and shaken vigorously. Then the separating funnel was kept undisturbed to separate the carotenoids from acetone to petroleum ether layer. After that, the coloured solution was separated in a 50 ml volumetric flask and the volume was adjusted with petroleum ether. Finally, the sample absorbance was measured at 452 nm in a spectrophotometer, using petroleum ether as blank. The final results were represented as µg/g of fresh weight basis. For estimation of Total flavonoids**,** a known volume of extract was placed in a 10 ml volumetric flask. Distilled water was added to make 5 ml. and 0.3 ml $NaNO_2$ (1:20) were added. 3 ml $AlCl_3$ (1:10) were added 5 min later. After 6 min, 2 ml 1 mol litre NaOH was added and the total was made up to 10 ml with distilled water. The solution was mixed well again and the absorbance was measured against a blank at 510 nm with a UV–vis spectrophotometer. The flavonoid content was calculated using the linear equation based on the calibration curve and results were reported as mg/g QE (Zhishen et al. 1999).

Sample extraction for total phenolics, FRAP antioxidant activity and total sugar

One gram of fresh, homogenised pulp underwent double extraction in 10 ml 80% ethanol for 30 min at 80 °C. After each extraction cycle, the samples were centrifuged at 10,000 rpm for 10 min, the supernatants were collected and pooled. Extract was dried over boiling water bath and redissolved in 10 ml of double distilled water. This extract in water was used for estimation of sugar, phenol and anti-oxidant activity.

Total phenol was estimated using the Folin–Ciocalteau reagents (FCR), involving a redox reaction. FCR consists of a combination of tungstates and molybdates, where phenolic compounds in the sample undergo reduction, resulting in the visible formation of a blue color. The absorbance at 650 nm was measured using a UV–vis spectrophotometer,

and the results were represented as a mg/g gallic acid equivalents (GAE) (Singleton et al. 1999). Benzie and Strain's (1996) method was used to calculate the ferric-reducing antioxidant power (FRAP) antioxidant capacity with slight modifications. A properly diluted sample (0.2 mL) was mixed with 0.8 mL of freshly prepared FRAP reagent. The FRAP reagent was composed of a 10:1:1 ratio of 0.3 M of acetate buffer (pH 3.6), 20 mM of ferric chloride solution ($FeCl_3.6H_2O$), and aqueous 10 mM of TPTZ in 40 mM of HCl. The absorbance of the solution was measured at 593 nm after 10 min of incubation at room temperature. A freshly prepared standard solution of ferrous sulfate was used for calibration. The final results were represented as a mg/g GAE. Total sugar content was determined using the anthrone reagent involving sugar dehydration by sulfuric acid to generate furfural. The absorbance of the furfural anthrone complex (blue-green) at 630 nm was measured using a UV–vis spectrophotometer, and a calibration curve was established using a D-glucose standard. This technique quantifies the amount of total sugars including mono-, di-, and oligosaccharides, and the results were and results were represented as a percent % (Hedge and Hofreiter 1962).

Statistical analysis

Significant effects ($p < 0.05$) were observed after the obtained data were subjected to a one-way analysis of variance (ANOVA) by using SAS 9.3 PROC GLM programme. Further, pair-wise significant differences among the accessions mean values were calculated by Tukey's Honestly Significant Difference (HSD) test. A correlation study was performed among different traits and generated a correlation matrix that depicts the degree of correlation along with its statistical significance. The correlation analysis was performed using Add-in XLSTAT (version 2014.5.03) software. To identify traits that contribute most to the overall variability were determined by performing principal component analysis (PCA) using Minitab 17 software (Version 2014.5.03). Hierarchical Cluster Analysis (HCA) was carried out to group the all accessions on the basis of similarity between different accessions using Ward's method. The analysis was performed using R-studio (R Core Team 2021).

Results

Generally, the bael tree is common in villages, temples, orchards and on field bunds. It is most common in Hindu temples, since its green leaves are presented to Lord Shankar, the supreme deity of Hindus. However, it occurs sporadically and scantily in forests. Young seedlings, an outcome of the natural regeneration of fallen fruits are observed. Due to the hard shell, its fruits are difficult to break and hence take a longer time to decompose and release the seed. The process starts from June onwards in the rainy season concurrently with the maturity of fruits.

Data from Table 1 reveals that out of the 21 accessions, IC0645501 had the highest average fruit weight (2542.5 g), followed by IC0645463 (1407.78 g) and IC0645509 (1336.84 g). The accessions IC0645480 and IC0645487 exhibited the lowest average fruit weight (77.65 g and 85.2 g, respectively). Maximum fruit length was recorded for IC0645501 (190.34 mm) followed by IC0645463 (136.58 mm) whereas the lowest was in IC0645477 (52.22 mm) followed by IC0645480 (59.51 mm). Accession IC0645501 has highest fruit width (168.21 mm) followed by IC0645509 (164.17 mm) and lowest was in IC0645481 (50.12 mm). Thickest fruit shell was recorded in IC0645506 (3.12 mm) followed by IC0645476 (3.0 mm), and the thinnest was in IC0645509 (1.95 mm). Maximum seeds per fruit were recorded in IC0645489 (151.33) followed by IC0645479 (123.67) and IC0645476 (116.33). The least seed count was in IC0645480 and IC0645481 (12.67 and 13.0, respectively).

Maximum dry matter content was recorded in IC0645477 (50.53%), followed by IC0645487 (48.46%) and IC0645480 (47.35%). Accessions IC0645499 and IC0645501 had low dry matter content (19.5% and 24.7%, respectively). The results showed a wide range of variation (19.5%–50.53%) in dry matter of different accessions. Higher dry matter is preferred due to better processing characteristics coupled with total soluble solid contents. The highest pulp recovery was in IC0645487 (71.22%), followed by IC0645499 (67.59%) and IC0645504 (64.96%). The lowest pulp recovery in ascending order was recorded in IC0645492 and IC0645477 (35.59 and 36.68%, respectively). Results showed a significant variation (> 2 folds) in pulp recovery among the

accessions. Out of 21, nine accessions had > 60% pulp content and 04 accessions had < 40% pulp content.

Total soluble solids ranged from 26.81 to 54.16°Brix (Table 2). Accession IC0645487 had the highest TSS content (54.16°Brix) followed by IC0645480 (47.78°Brix). The lowest TSS content was found in IC0645499 and IC0645501 (26.81 and 28.13°Brix, respectively). Eight accessions had > 40°Brix TSS and remaining 13 accessions had < 40°Brix TSS. Accession IC0645481 had the maximum ascorbic acid (0.69 mg/g) subsequently IC0645485 (0.62 mg/g), while the lowest was recorded in IC0645509 and IC0645501 (0.31 and 0.36 mg/g, respectively), showing 2.24 folds variation. Total carotenoid content was maximum in IC0645485 (6.21 µg/ g) followed by IC0645465 (5.90 µg/ g), while it was lowest in IC0645501 and IC0645491 (0.62 and 0.71 µg/ g, respectively), showing difference of 9.5 folds. FRAP antioxidant activity ranged from 0.10 to 0.20 mg/g GAE. Accession IC0645480 had the highest FRAP antioxidant activity (0.20 mg/ g GAE) followed by IC0645465 (0.20 mg/g GAE), while it was lowest in IC0645496 (0.10 mg/ g GAE). The flavonoid content varied from 0.12 mg/ g QE (IC0645501) to 0.61 mg/ g QE (IC0645487), showing five-fold difference among the accessions. Total phenolics varied from 1.22 (IC0645501) to 2.27 mg/ g- GAE (IC0645480). The maximum total sugars was reported in IC0645496 (5.30%) followed by IC0645463 (5.04%) and IC0645465 (5.03%). However, IC0645477 and IC0645506 had the lowest total sugar content (3.10 and 3.24%, respectively).

All fruit characteristics (14) were tested for significant differences among 21 accessions through Tukey's HSD test (Table 1 and 2). The accession means denoted by different letters are significantly different while those with the same letters are non-significantly different. Based on fruit traits all genotypes are differentiated in different letters with superscript form.

Hierarchical cluster and characters association

The correlation between the various bael genotypes was assessed in the current study using a scale ranging from 0 to 15 (Fig. 3). All the accessions were grouped into three major clusters at Euclidean distance of 10. Cluster one has seven members; cluster two is the smallest which has only two bael accessions (IC0645501 and IC0645509) and twelve accessions comes under cluster three. These three clusters were primarily separated on the basis of similarity in morphological traits of fruits in various accessions.

Association among different characteristics of fruits was evaluated through correlation coefficient (Table 3 and Table 4). It is evident from Table 3 that dry matter showed negative correlation with fruit length ($r = -0.59$), fruit width ($r = -0.66$) and fruit weight ($r = -0.57$). Table 4 shows the correlation among chemical characteristics of the bael fruits. Total carotenoid content showed positive correlation with total phenolics ($r = 0.73$) and total flavonoids ($r = 0.59$).

Principal component analysis for physico-chemical traits of different bael genotypes

PCA was applied to morphological and chemical characteristics of bael accessions and results are summarized in Fig. 4 and Fig. 5. It is evident from results (Fig. 4) that the highest explained variance (PC-1) in case of morphological characteristics was associated with length, width and average fruit weight in parallel directional trend, while dry matter in reverse direction. PC-1 explained 50.50% variance, whereas the second factor (PC-2) explained about 20.70% variance. PC-2 was found to be closely linked with shell thickness and seed count. Results (Fig. 5) further explain that the highest explained variance (PC-1) in case of chemical characteristics was associated with TSS, Ascorbic acid, FRAPS, Flavonoid content, total carotenoids and total phenolic content, and explained 47.12% variance, whereas PC-2 explained 19.86% variance and showed association with total sugar content.

It can be interpreted from Fig. 4 & 5 that the variables close to one another and placed in the same quadrant of the biplot are positively associated, whereas variables loaded at a higher distance and in opposite quadrants are negatively associated. Figure 4 explains that the accession IC0645501 was located in first quadrant and showed a close association with length, width and average fruit weight. Likewise, seed count showed association with accessions IC0645476, IC0645489, IC0645479 and IC0645492. Shell thickness showed association with IC0645480 and IC0645481. Pulp content was associated with IC0645509 whereas dry matter content was found to be associated with accessions IC0645480 and

IC0645487. Figure 5 presents the association of different bael accessions with their chemical characteristics. It is evident from Fig. 5 that TSS, Ascorbic acid, FRAP, Flavonoid content, Total carotenoids and Total phenolics showed a grouping, and this group showed association with the accessions IC0645465, IC0645485, IC0645504, IC0645480, IC0645487, IC0645481, IC0645503 and IC0645479. Likewise, Total sugar content showed the association with the accessions IC0645465, IC0645463, IC0645468, IC0645489 and IC0645489.

Discussion

As Fig. 2 and Table 1 demonstrate, there was significant variation in the fruit shape and size among the different accessions. It has been found that different genotypes and cultivars exhibit comparable variances in bael fruit properties, such as fruit weight, fruit length, volume, diameter, and shell thickness (Nath et al. 2003; Pandey et al. 2013). Although the morphological fruit quality traits are genetically governed, but they are also equally affected by environmental and edaphic conditions (Mahmood et al. 2012). The fruit length (52.2–190.3 mm) varied much higher than reported earlier (81.39–173.43 mm), while shell thickness (1.95–3.12 mm) showed lesser variation (Dhakar et al. 2019, 1.43–4.10 mm). A thinner shell is a desirable trait of the bael fruits. A wide range of variations in fruit weight (0.315–2.09 kg) of different genotypes were recorded in the present study and by others (Singh et al. 2000; Ram and Singh 2003; Dhakar et al. 2019)). The difference in fruit weight may be due to fruit size and increased amount of pulp, seeds, and shell. Present findings of these traits also agree with previous work (Pandey et al. 2008, 2013; Mitra et al. 2010).

The difference in seed weight might be attributed to differences in the quantity and dimension of seeds from different genotypes. The findings presented here align with previous research (Pandey

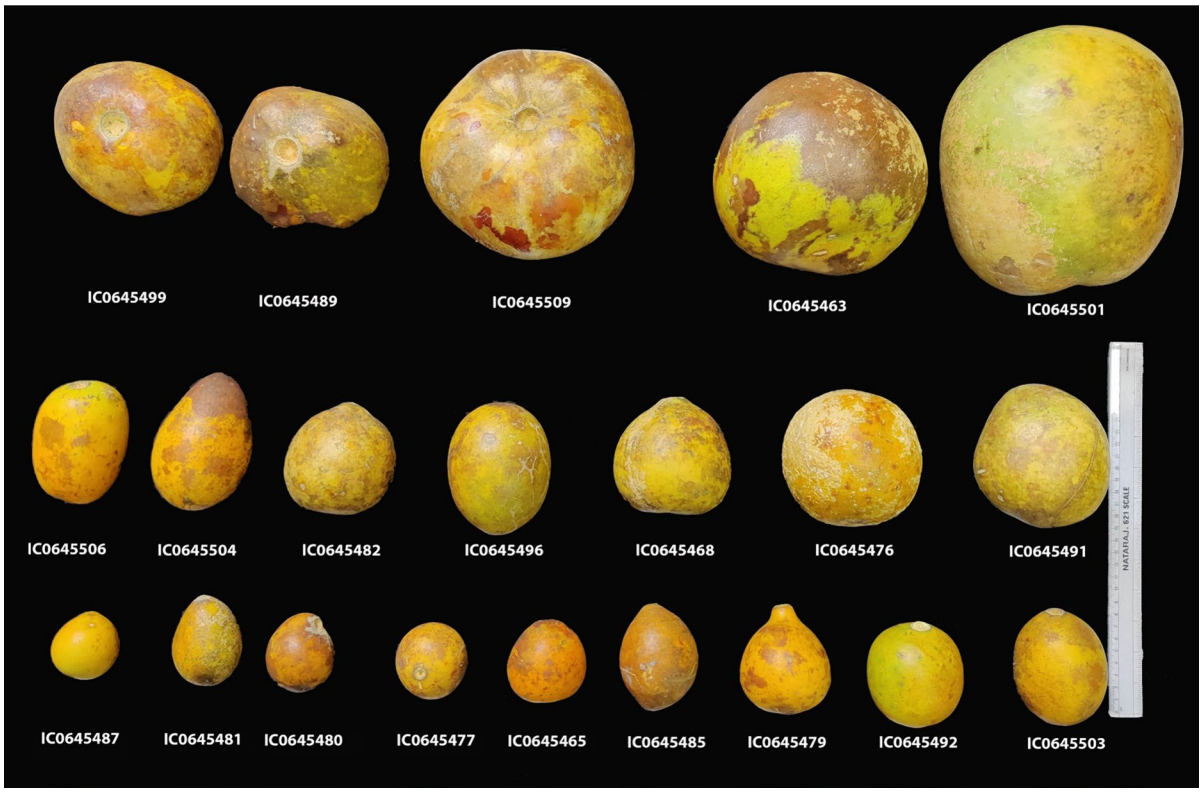

Fig. 2 Variability in shape and size of different bael accessions obtained from Achanakmar-Amarkantak Biosphere Reserve

Table 1 Morphological traits of 21 bael fruits accessions explored from Achanakmar-Amarkantak Biosphere Reserve

Accession No	Length (mm)	Width (mm)	Shell thickness (mm)	Fruit weight (g)	No. of seeds/fruit	Dry matter (%)	Pulp (%)
IC0645463	136.58 ± 21.20^{b}	133.50 ± 4.43^{b}	2.70 ± 0.08^{abcd}	1407.78 ± 23.84^{b}	54.00 ± 1.73^{j}	42.38 ± 0.92^{e}	63.33 ± 2.99^{bc}
IC0645465	81.82 ± 5.55^{fg}	76.86 ± 3.03^{efg}	2.17 ± 0.03^{ef}	145.82 ± 1.58^{lmn}	67.33 ± 1.53^{h}	45.38 ± 0.78^{cd}	37.50 ± 1.61^{ij}
IC0645468	88.93 ± 1.95^{def}	95.58 ± 1.86^{d}	2.44 ± 0.04^{cde}	369.01 ± 9.72^{g}	54.67 ± 0.58^{j}	38.11 ± 0.80^{fg}	54.69 ± 0.92^{fg}
IC0645476	102.46 ± 3.49^{cde}	102.92 ± 4.37^{d}	2.94 ± 0.19^{ab}	583.51 ± 20.69^{e}	116.33 ± 0.58^{c}	31.77 ± 0.30^{k}	60.02 ± 2.87^{cdef}
IC0645477	52.22 ± 1.46^{h}	58.08 ± 2.42^{ijk}	3.00 ± 0.21^{ab}	94.61 ± 1.68^{n}	44.00 ± 1.73^{l}	50.53 ± 0.81^{a}	36.68 ± 1.30^{ij}
IC0645479	83.97 ± 3.46^{fg}	69.80 ± 4.11^{fgh}	2.61 ± 0.05^{bcde}	191.15 ± 5.60^{klm}	123.67 ± 1.15^{b}	35.61 ± 0.73^{hij}	39.26 ± 1.02^{hij}
IC0645480	59.51 ± 1.01^{h}	51.17 ± 2.26^{k}	2.59 ± 0.25^{bcde}	77.65 ± 1.23^{n}	12.67 ± 0.58^{n}	47.35 ± 0.68^{bc}	61.71 ± 2.49^{bcde}
IC0645481	69.68 ± 1.52^{gh}	50.12 ± 1.74^{k}	2.71 ± 0.03^{abcd}	92.38 ± 4.71^{n}	13.00 ± 1.00^{n}	44.26 ± 1.13^{de}	43.08 ± 1.81^{h}
IC0645482	87.06 ± 2.69^{efg}	81.98 ± 6.40^{e}	2.79 ± 0.10^{abcd}	291.05 ± 5.93^{hi}	97.33 ± 1.15^{e}	38.78 ± 0.70^{fg}	44.27 ± 1.49^{h}
IC0645485	79.81 ± 4.30^{fg}	63.59 ± 0.81^{hij}	2.15 ± 0.09^{ef}	144.73 ± 2.64^{mn}	72.33 ± 1.15^{f}	37.52 ± 0.89^{ghi}	40.03 ± 1.88^{hij}
IC0645487	60.78 ± 4.35^{h}	53.00 ± 1.65^{jk}	2.39 ± 0.09^{cdef}	85.20 ± 3.62^{n}	22.00 ± 1.73^{m}	48.46 ± 0.47^{ab}	71.22 ± 3.15^{a}
IC0645489	105.59 ± 2.22^{cd}	119.50 ± 5.49^{c}	2.71 ± 0.27^{abcd}	708.63 ± 34.76^{d}	151.33 ± 1.15^{a}	35.53 ± 0.97^{ij}	60.05 ± 2.57^{cdef}
IC0645491	112.55 ± 0.04^{c}	98.45 ± 5.61^{d}	2.54 ± 0.01^{bcde}	492.13 ± 31.20^{f}	41.67 ± 1.15^{l}	37.92 ± 0.47^{fgh}	56.54 ± 1.44^{defg}
IC0645492	82.22 ± 1.13^{fg}	72.74 ± 2.82^{efgh}	2.83 ± 0.26^{abc}	215.56 ± 8.95^{jkl}	110.33 ± 0.58^{d}	39.95 ± 0.68^{f}	35.59 ± 1.39^{j}
IC0645496	102.07 ± 5.90^{cde}	80.15 ± 2.25^{ef}	2.72 ± 0.22^{abcd}	343.50 ± 3.24^{gh}	71.00 ± 0.00^{fg}	37.62 ± 0.76^{ghi}	52.58 ± 3.02^{g}
IC0645499	97.20 ± 4.29^{cdef}	120.95 ± 2.42^{c}	2.80 ± 0.12^{abcd}	704.28 ± 16.55^{d}	68.33 ± 0.58^{gh}	19.49 ± 0.45^{m}	67.59 ± 0.27^{ab}
IC0645501	190.34 ± 1.48^{a}	168.21 ± 1.73^{a}	2.73 ± 0.15^{abcd}	2542.50 ± 65.84^{a}	59.67 ± 0.58^{i}	24.69 ± 0.49^{l}	60.50 ± 1.51^{cdef}
IC0645503	91.39 ± 2.73^{def}	68.30 ± 2.35^{ghi}	2.60 ± 0.20^{bcde}	221.82 ± 14.99^{ijk}	48.33 ± 0.58^{k}	44.56 ± 1.05^{de}	55.52 ± 3.27^{efg}
IC0645504	110.17 ± 3.65^{c}	70.74 ± 1.33^{fgh}	2.36 ± 0.18^{def}	266.93 ± 15.45^{ij}	42.67 ± 1.15^{l}	43.33 ± 0.81^{de}	64.96 ± 2.66^{abc}
IC0645506	86.09 ± 4.50^{efg}	72.04 ± 3.24^{efgh}	3.12 ± 0.04^{a}	254.54 ± 1.70^{ijk}	24.00 ± 1.73^{m}	33.57 ± 0.39^{k}	40.86 ± 1.55^{hij}
IC0645509	112.47 ± 3.74^{c}	164.17 ± 6.73^{a}	1.95 ± 0.04^{f}	1336.84 ± 47.00^{c}	72.67 ± 1.15^{f}	34.85 ± 0.73^{j}	62.12 ± 1.73^{bcd}

et al. 2008, 2013; Singh and Misra 2010). A high degree of difference found in number of seeds/fruit (37.67 to 195.40) agrees with other reports (Dhakar et al. 2019). Higher seeds in a fruit are an undesirable trait; hence, low seeds or seedless fruits are preferred. However, for medicinal properties, the higher seeds may be a desirable trait since mucilaginous content which is adhered to seeds, may be maximum in these fruits. Out of 21, nine accessions had quite high fruit pulp ($\geq 60\%$). Other improved or domesticated genotypes had 53.72–89.33% pulp (Prasad and Singh 2001; Dhakar et al. 2019). A large variation in morphological and biochemical traits may be attributed to genotypic variation and recombinants develop through cross-pollination and seed propagation (Kumar et al. 2008). Among different accessions, IC0645501 (fruit weight 2542.5 g; seed count 59.67) and IC0645463 (fruit weight 1407.78 g; seed count 54) had the lowest seed counts compared to the fruit weight. Because of its low seed-to-fruit ratio, accession IC0645501 is especially suggested for the fresh market and processing uses.

In the present study, a significant variation recorded in biochemical traits like TSS (26.81–54.16°Brix), ascorbic acid (0.31–0.69 mg/g), total carotenoids (0.62–6.21 µg/g), total phenolics (1.22–2.27 mg/g GAE), flavonoid content (0.12–0.61 mg/g QE) and FRAP antioxidant activity (0.10–0.20 mg/g GAE). Similar variation in biochemical traits like ascorbic acid, acidity, TSS, sugar content, phenolics, antioxidants etc. of different accessions were reported and attributed to genotypic, environmental, size and age differences of individuals (Lee et al. 1976; Prasad and Singh 2001; Ram and Singh 2003; Bhat and Kumari 2006; Sarkar et al. 2015; Singh et al. 2018; Dhakar et al. 2019).

To a certain extent, the higher phenolic content ($r = 0.63$) in different accessions might be responsible for their enhanced FRAP antioxidant activity. Biochemical parameters like AA, TSS, pH, etc. are more influenced by environmental conditions. Many researchers stated that environmental factors not only affect the physiological fruit development process but also modify the phytochemical constituents (Mahmood et al. 2012; Sum et al. 2013). The growing season, area, and type of cultivars/genotype components have an equal impact on synthesis of TSS and

Table 2 Biochemical quality traits of 21 bael fruits accessions explored from Achanakmar-Amarkantak Biosphere Reserve

Accession No	TSS	Ascorbic acid	FRAP antioxidant activity	Flavonoid content	Total carotenoids	Total phenolics	Total sugar
	°Brix	mg/g	mg/g GAE	mg/g QE	µg/g	mg/g GAE	%
IC0645463	39.27 ± 0.77^{fgh}	0.54 ± 0.02^{d}	0.15 ± 0.002^{fgh}	0.19 ± 0.004^{ij}	1.27 ± 0.04^{jk}	1.80 ± 0.04^{de}	5.04 ± 0.11^{ab}
IC0645465	44.86 ± 0.80^{cd}	0.62 ± 0.00^{b}	0.20 ± 0.002^{ab}	0.31 ± 0.008^{f}	5.90 ± 0.05^{b}	2.17 ± 0.02^{a}	5.03 ± 0.15^{ab}
IC0645468	37.94 ± 0.22^{hij}	0.50 ± 0.01^{fg}	0.19 ± 0.002^{bc}	0.29 ± 0.006^{g}	1.40 ± 0.03^{ij}	1.49 ± 0.03^{hi}	4.91 ± 0.10^{bc}
IC0645476	34.20 ± 0.26^{kl}	0.58 ± 0.00^{c}	0.15 ± 0.003^{fgh}	0.14 ± 0.003^{m}	1.12 ± 0.03^{k}	1.55 ± 0.03^{gh}	4.44 ± 0.15^{ef}
IC0645477	45.47 ± 1.20^{bc}	0.54 ± 0.01^{de}	0.15 ± 0.002^{fgh}	0.50 ± 0.012^{b}	4.92 ± 0.08^{c}	1.99 ± 0.05^{b}	3.10 ± 0.07^{j}
IC0645479	42.54 ± 1.74^{de}	0.53 ± 0.02^{def}	0.14 ± 0.003^{gh}	0.30 ± 0.001^{fg}	1.52 ± 0.06^{i}	1.72 ± 0.04^{ef}	3.96 ± 0.08^{hi}
IC0645480	47.78 ± 1.60^{b}	0.51 ± 0.00^{ef}	0.20 ± 0.004^{a}	0.38 ± 0.004^{d}	5.01 ± 0.18^{c}	2.27 ± 0.04^{a}	4.90 ± 0.08^{bcd}
IC0645481	41.28 ± 0.57^{ef}	0.69 ± 0.01^{a}	0.10 ± 0.003^{i}	0.46 ± 0.009^{c}	1.89 ± 0.01^{h}	1.69 ± 0.03^{ef}	4.61 ± 0.08^{de}
IC0645482	33.52 ± 0.17^{l}	0.47 ± 0.01^{h}	0.15 ± 0.003^{fgh}	0.21 ± 0.004^{i}	3.51 ± 0.08^{e}	1.68 ± 0.03^{f}	4.40 ± 0.05^{efg}
IC0645485	33.09 ± 0.83^{l}	0.62 ± 0.01^{b}	0.16 ± 0.003^{ef}	0.35 ± 0.007^{e}	6.21 ± 0.05^{a}	1.94 ± 0.05^{bc}	4.79 ± 0.09^{bcd}
IC0645487	54.16 ± 0.94^{a}	0.60 ± 0.01^{bc}	0.16 ± 0.003^{efg}	0.61 ± 0.006^{a}	4.54 ± 0.10^{d}	1.69 ± 0.01^{ef}	4.89 ± 0.06^{bcd}
IC0645489	34.82 ± 0.70^{kl}	0.58 ± 0.00^{c}	0.16 ± 0.002^{efg}	0.16 ± 0.001^{l}	2.45 ± 0.05^{g}	1.62 ± 0.06^{fg}	4.79 ± 0.10^{bcd}
IC-0645491	36.46 ± 0.85^{ijk}	0.47 ± 0.01^{gh}	0.14 ± 0.002^{h}	0.18 ± 0.003^{jk}	0.71 ± 0.01^{mn}	1.48 ± 0.02^{hi}	3.84 ± 0.08^{i}
IC0645492	38.37 ± 1.29^{ghi}	0.46 ± 0.01^{hi}	0.17 ± 0.001^{de}	0.29 ± 0.008^{g}	0.90 ± 0.01^{lm}	1.84 ± 0.02^{cd}	4.37 ± 0.08^{efg}
IC0645496	35.25 ± 0.78^{jkl}	0.51 ± 0.00^{ef}	0.10 ± 0.002^{i}	0.23 ± 0.002^{h}	1.30 ± 0.03^{jk}	1.54 ± 0.04^{gh}	5.30 ± 0.13^{a}
IC0645499	26.81 ± 0.45^{m}	0.62 ± 0.01^{b}	0.17 ± 0.002^{de}	0.17 ± 0.003^{kl}	1.51 ± 0.04^{i}	1.38 ± 0.04^{ij}	4.85 ± 0.14^{bcd}
IC0645501	28.13 ± 0.50^{m}	0.36 ± 0.01^{j}	0.10 ± 0.003^{i}	0.12 ± 0.001^{n}	0.62 ± 0.01^{n}	1.22 ± 0.02^{k}	4.20 ± 0.09^{fgh}
IC0645503	40.80 ± 1.01^{efg}	0.55 ± 0.01^{d}	0.16 ± 0.003^{efg}	0.30 ± 0.003^{fg}	2.67 ± 0.04^{f}	1.50 ± 0.03^{h}	4.12 ± 0.02^{ghi}
IC0645504	46.35 ± 0.55^{bc}	0.50 ± 0.01^{fg}	0.18 ± 0.004^{cd}	0.29 ± 0.006^{g}	4.52 ± 0.14^{d}	2.22 ± 0.07^{a}	4.91 ± 0.10^{bcd}
IC0645506	28.68 ± 0.63^{m}	0.43 ± 0.01^{i}	0.14 ± 0.003^{gh}	0.18 ± 0.005^{jkl}	2.53 ± 1.15^{fg}	1.26 ± 0.02^{k}	3.24 ± 0.09^{j}
IC0645509	34.65 ± 0.80^{kl}	0.31 ± 0.01^{k}	0.11 ± 0.002^{i}	0.14 ± 0.002^{m}	1.10 ± 0.02^{kl}	1.29 ± 0.01^{jk}	4.62 ± 0.09^{cde}

bioactive compounds (Das et al. 2012; Csambalik et al. 2014; Pandey et al. 2015; Kannaujia et al. 2019).

The most contribution to genetic divergence comes from several fruit quality attributes, such as average fruit weight, fruit dimensions (length), total number of seeds per fruit, seed weight of individual fruits, ascorbic acid concentration, and fibre content (Rai and Misra 2005). Large variations in the economic importance variables, such as fruit yield, fruit weight and TSS were also reported by Singh and Misra (2010), and suggested that there is scope for both selection and use of various parents in hybridization. Hence, in addition to fruit weight, other traits should be given due weightage like pulp recovery, number of seeds per fruit, shell thickness and TSS when choosing a superior bael genotype.

In the current study, various accessions/genotypes of bael showed a positive association in morphological parameters, such as fruit weight, length and width etc. Similarly, biochemical parameters also had a positive correlation almost with each other (viz., TSS content, total phenolics, flavonoids, total carotenoids etc.). Furthermore, positive correlation among the morphological and biochemical parameters were also reported by several researchers in various fruit and vegetable crops. The correlation coefficient among morphological and biochemical traits has also been previously reported in various horticultural crops like mango, carrot, litchi and cherry tomatoes (Barman and Asrey 2014; Koley et al. 2014; Kumari et al. 2018; Oboulbiga et al. 2018; Kannaujia et al. 2019). HCA was performed by several authors to find the similarity among the different cultivars based on evaluated quality parameters in various horticultural crops (Kumari et al. 2018; Kannaujia et al. 2019).

PCA results indicated that the accession IC0645501 showed strong association with length, width and average fruit weight. Likewise, number of seeds was found to be associated with the accessions IC0645476, IC0645489, IC0645479 and IC06

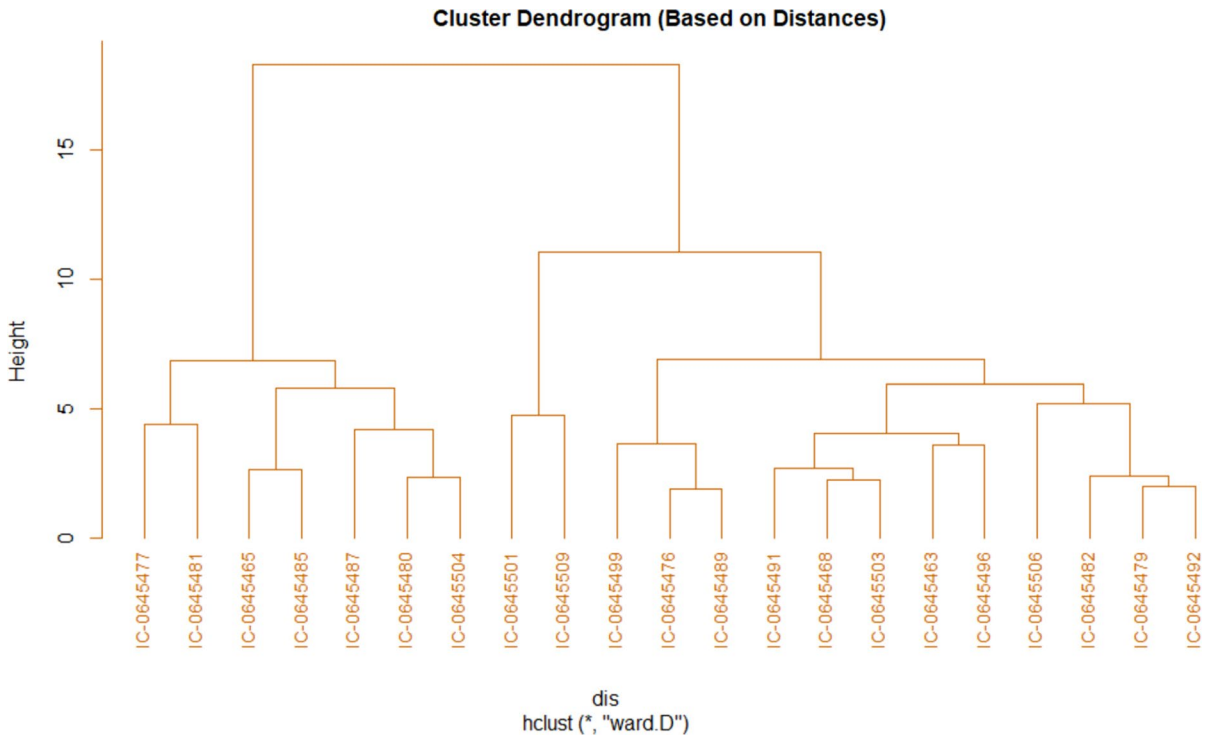

Fig. 3 Analysis of various Bael accessions using hierarchical clustering derived from evaluated parameters

Table 3 Correlation coefficient matrix for morphological traits of different bael accessions

Value shows in bold form are different from 0 with a significance level alpha = 0.05

Variables	L	W	Shell thickness	Fruit weight	Seed count	Dry matter	Pulp
L	**1**						
W	**0.833**	**1**					
Shell thickness	0.050	−0.073	**1**				
Fruit weight	**0.914**	**0.913**	0.008	**1**			
Seed count	0.190	0.314	0.138	0.130	**1**		
Dry matter	**−0.591**	**−0.659**	−0.261	**−0.565**	−0.388	**1**	
Pulp	0.392	**0.447**	−0.169	0.418	−0.160	−0.225	**1**

Table 4 Correlation coefficient matrix of biochemical traits of different bael accessions

Variables	TSS	Ascorbic acid	FRAP	Flavonoid content	Total carotenoids	Total phenolics	Total sugar
TSS	**1**						
Ascorbic acid	0.304	**1**					
FRAP	0.378	0.271	**1**				
Flavonoid content	**0.799**	**0.489**	0.200	**1**			
Total carotenoids	**0.513**	0.388	**0.530**	**0.588**	**1**		
Total phenolics	**0.689**	0.391	**0.613**	**0.521**	**0.731**	**1**	
Total sugar	0.143	0.282	0.172	−0.007	0.084	0.251	**1**

value shows in bold form are different from 0 with a significance level alpha = 0.05

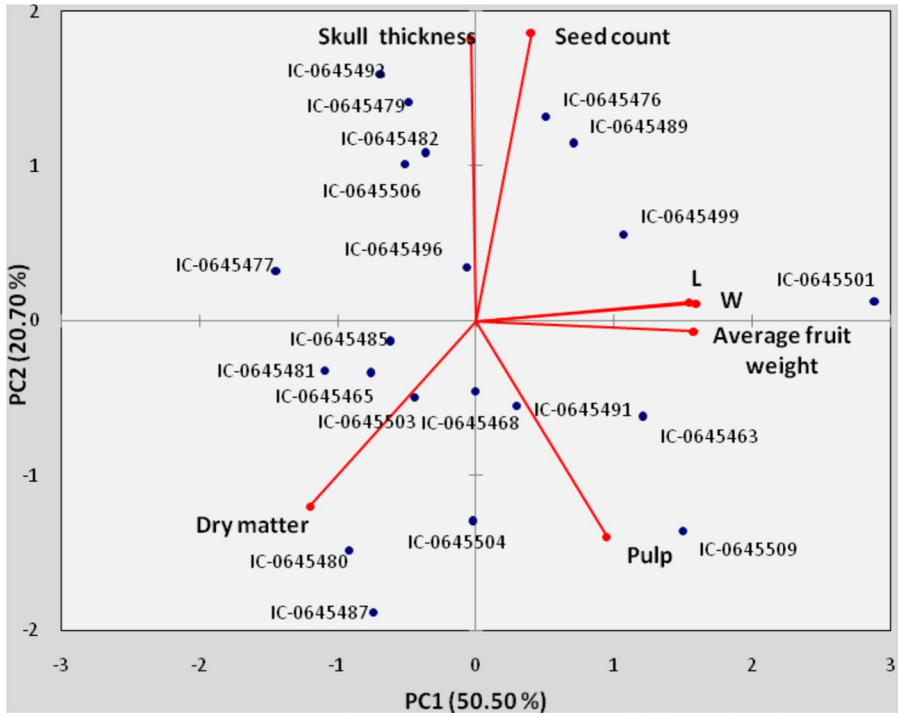

Fig. 4 Plots of first two principal components (PC) obtained for morphological traits of different bael accessions

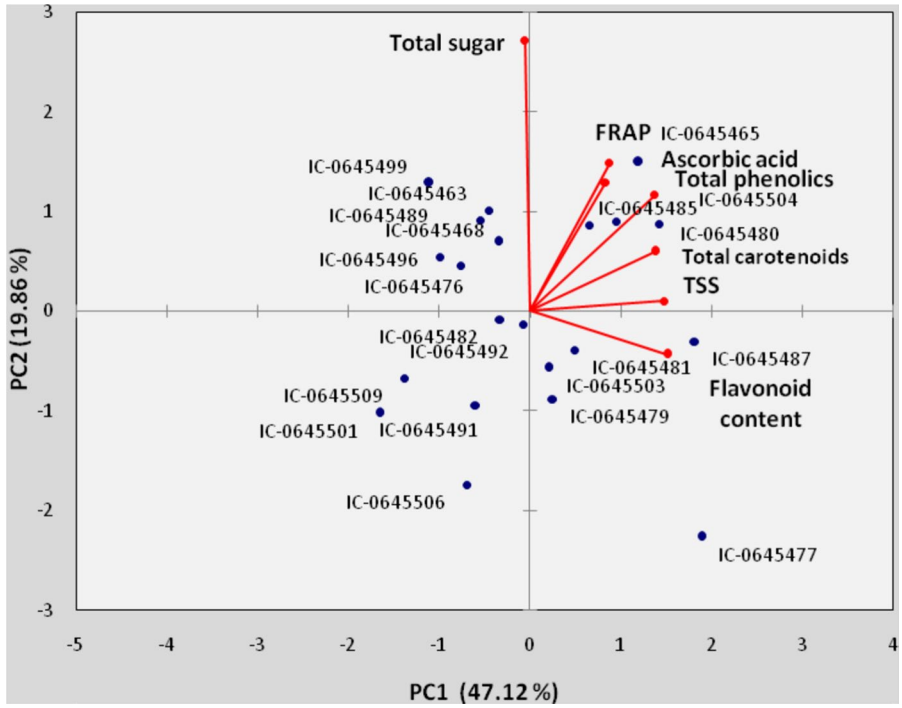

Fig. 5 Plots of first two principal components (PC) obtained for biochemical traits of different bael accessions

45492 whereas shell thickness was associated with the accessions IC0645480 and IC0645481. Overall biochemical composition comprising of TSS, Ascorbic acid, FRAP, Flavonoid content, Total carotenoids and Total phenolics showed a good association with the accessions IC0645465,

IC0645485, IC0645504, IC0645480, IC0645487, IC0645481, IC0645503 and IC0645479 as compared to the remaining accessions.

Conclusion

Present findings revealed a high degree of variability in fruit attributes of bael accessions. The findings have helped in identifying the promising accessions by breeders in crop improvement programmes. The bael trees having bigger fruits, high pulp, TSS, carotenoids, antioxidants etc. can be multiplied for cultivation. The accessions IC0645501, IC0645463, IC0645465 and IC0645509 may be suggested for cultivation to produce good quality raw material for boosting local consumption and processing, develop value-added products and provide phytochemical compounds for nutraceutical use.

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Data availability The data that support this study are available in the article.

Declarations

Conflict of interest It is confirmed that no conflicts of interest among the all authors.

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