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Profiling of major biochemical compounds for identification of nutritionally rich genotypes in mango

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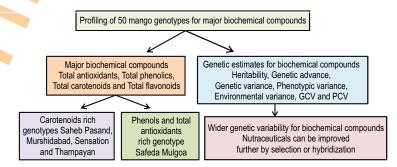
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Abstract

Aim: Mango is a major fruit crop grown in India with wide variability for fruit colour, size, shape and fruit quality. The main objective of this study was profiling of mango genotypes and their genetic estimates for major biochemical compoun<mark>ds</mark> to identify superior genotypes as parents for developing nutritionally rich hybrids in future.

Methodology: Fifty mango varieties were analysed for biochemical compounds such as total antioxidants, total phenols, total flavonoids and total carotenoids. The L*, a*, b* values were recorded for pulp colour, and genetic estimates were also worked for genetic parameters such as genotypic variance, phenotypic variance, GCV, PCV, heritability and genetic advance. Correlation matrix for major biochemical compounds and L*, a*, b* values were also calculated.

Results: The present study revealed the presence of wide genetic variability in mango gene pool for biochemical compounds and pulp colour. The total phenolics ranged from 24.44 to 148.33 mg 100g⁻¹ of pulp, total flavonoids 10.33-49 mg 100g⁻¹, total carotenoids 0.48-7.50 mg 100g⁻¹ and total antioxidants 0.14-1.59 umol Trolox 100g¹ of pulp. The future genetic estimates showed that the phenotypic coefficient of variation (PCV) for all the characters was slightly higher than genotypic coefficient of variation (GCV), indicating the presence of environmental influence to some degree in the phenotypic expression of characters. However, the difference between PCV and GCV for all the traits was narrow suggesting that the traits were less influenced by environment. The broad sense heritability (H²_b) for all the compounds was high which indicates the phenotype of that trait strongly reflects its genotype.



Interpretation: High values of PCV, GCV and heritability for total antioxidants, total phenols, total flavonoids and total carotenoids indicated high genetic variability and better scope for selection and improvement through hybridization.

Key words: Biochemical compounds, Genetic advance, Heritability, Mangifera indica, Superior parents

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Introduction

Mango (Mangifera indica L.) is one of the major fruit crop in the world and native to India. The wide genetic diversity of mango available in India is very rich, and presently more than one thousand vegetatively propagated varieties exist in our country. Previously, characterization of mango germplasm was mainly based on morphological traits (Ram and Rajan, 2003) and the main objectives of hybridization are to develop varieties which are high yielding, attractive colour, less fibre and resistance to pest and diseases. But, now priorities have changed and people are more conscious about health due to frequent occurrence of deadly diseases like cancer, heart attack and obesity problems. Recent developments in epidemiological studies have indicated that frequent consumption of natural antioxidants is associated with lower risk of cardiovascular disease and cancer (Renaud et al., 1998; Temple, 2000). Hence, currently there is a lot of demand for nutritionally rich genotypes due to its health benefits.

Mango is also well known for its good source of dietary antioxidants such as carotenoids and phenolic compounds. There is a necessity to combine production efficiency traits with improved fruit quality, including the new concept of improved fruit sensorial, nutritional and biochemical compounds (Wang and Lewers, 2007; Cheng et al., 2004). Correlation studies will also help in finding out the degree of inter relationship among various biochemical characters, which will further help in selection criteria for improvement. In consequence, present breeding programmes in fruits and vegetables are focused more on improvement of bioactive compounds (Diamanti et al., 2010). Improvement in quality of fruits can be achieved by selecting of genotype with increased content of bioactive compounds or by developing new hybrids. It has already been proved that wide variability exists for fruit physical parameters like weight, length, breadth, colour, size and shape, fruit quality (Majumder et al., 2012; Bhowmick and Banik, 2008) and chemical and nutritional traits such as TSS, sugar, acidity, phenols, carotenes and ascorbic acid content among different varieties (Himabindu et al., 2016).

Therefore, there is an urgent need for screening of mango gene pool for nutritional components to identify superior parents to develop nutritionally rich hybrids in future. In this context, the present study was carried out to see the genetic variability in different genotypes of mango for important biochemical compounds such as total antioxidants, total phenolics, total flavonoids and total carotenoids. This study provides information related to nutritional status of different cultivars for the identification of superior parents and genetic estimates for developing nutritionally rich hybrids in future.

Materials and Methods

Fifty mango genotypes *viz.*, Angoordana, Chausa, Rajawala, Merkeara, Thampayan, Hushnara, Kasi Ratnalu, Jawahar, Lal Mani, Surkha Thakurbag, Menaka, Allahabadi Chausa, Serehayat, Eldon, Neeleshwari, NR-122, Kunjlata,

Amin, Samar Bahist Rampur, Arka Aruna, Mithuwa, Khader, Brindabani, Bathuie Katikee, Rataul, Gulabi, Chandrakaran, Nekkare, Vellaikolumban, Safeda Mulgoa, Neelgoa, Saheb Pasand, Konkan Ruchi, Murshidabad, Santra, Chilta Khas, Mylepelian, Rumani, Bhogal Shah, Janardan Pasand, Kalapahar, Sadaphal, Ratna, Benazir, Gootee, MadhavaRao Pasand, EC-95862, PFN-11, Manjeera and Sensation were used for the study. The fully matured fruits were randomly collected from mango orchards located at institute farm at Rehmankhera, Lucknow, India. The fruits were washed with water to remove field heat, as well as dirt deposited on fruits. The fruits were stored at room temperature for uniform ripening by considering softness and colour. The pulp of mango genotypes was used for analysis of different biochemical compounds, such as total antioxidants, total phenols, total flavonoids and total carotenoids. The colour reading was recorded immediately after cutting the fruits.

Estimation of biochemical compounds: Total phenolics content in the extracts were estimated by the Folin-Ciocalteau reagent method (Singleton et al., 1999). The absorbance was recorded at 750 nm by using a UV–VIS spectrophotometer. Gallic acid was used as standard and total phenolic content was determined on the basis of calibration equations, expressed as gallic acid equivalents (GAE) in mg per 100 g of pulp. The analysis of total flavonoid content was determined according to the colorimetric method (Dewanto et al., 2002). The absorbance was recorded at 510 nm against a reagent blank. The results were expressed as mg quercetin equivalents per 100 g fresh weight. Total carotenoid content was determined following the modified method of Ranganna (1997) using acetone and petroleum ether as extracting solvents and measured the absorbance at 452 nm. The results were expressed as mg per 100 g of pulp. The total antioxidant activity of fruits was determined by using CUPRAC (Cupric Reducing Antioxidant Capacity) assay (Apak et al., 2004). The absorbance was recorded at 450 nm against a reagent blank in the UV-VIS spectrophotometer. The units were expressed in trolox - Trolox per 100g pulp.

Colour parameters: Pulp colour was determined longitudinally at three equidistant points on freshly cut fruits by using ColorLite Sph 850 spectrophotometer (ColorLite GmbH, Katlenburg-Lindau, Germany), $\ln L^*$, a^* , b^* scale, whereas L^* is the measure of lightness, the positive values of a^* are in direction of redness and positive values of b^* are the vector of yellowness. The negative values of a^* indicates colour towards greenness and that of b^* depicts blueness. Each fruit pulp was measured at three different places and mean values were obtained.

Experimental design and genotypic analysis: Data were expressed as means standard deviation of three replications. ANOVA (SPSS version 16.0) and *Turkey's post hoc* test were used to determine the mean difference of different biochemical compounds, and L*, a*, b* values and Pearson's correlation test was used to determine the correlation. Genetic parameters were estimated as suggested by Johnson *et al.* (1955), Panse (1957) and Gomez and Gomez (1984). Heritability in broad sense (H²,)

and genetic advance were also calculated because these are the only useful measure of heritability for out crossing crops (Wricke and Weber, 1986). The genotypic and phenotypic coefficient of variation were calculated as per Burton and De Vane (1953), heritability by following the method of Johnson *et al.* (1955) and genetic advance by that of Allard (1960).

Results and Discussion

Fifty mango genotypes were evaluated for major biochemical compounds such as total phenols, total flavonoids, total carotenoids along with total antioxidants activity whereas colorlite values were recorded to show the pigmentation pattern in the pulp (Table 1). Among the genotypes, significant variability was observed from 0.14-1.59 µmol Trolox 100g¹ for total antioxidants. The genotype Safeda Mulgoa (1.59 µmol Trolox 100g¹) had higher antioxidants, followed by Nekkare (1.33 µmol Trolox 100g¹) and minimum was in Rajawala (0.14 µmol Trolox 100g¹). The genotype Safeda Mulgoa had ten times more total antioxidants compared to Rajawala which bears big sized fruits (1.0 to 2 kg per fruit) with irregular shape.

Total phenol contents were higher in genotype Safeda Mulgoa (148.33 mg 100g⁻¹) followed by Brindabani (137.50 mg 100g⁻¹) and lowest in Rajawala (24.44 mg 100g⁻¹). Ribeiro *et al.* (2007) reported that Safeda Mulgoa had higher phenolics among the varieties but lower than the pulp of cultivar Uba (200 mg 100g⁻¹). Higher antioxidants and phenolics content in Safeda Mulgoa indicate that, the phenols contribute more to antioxidant activity compared to flavonoids and carotenoids. The results also indicate that total antioxidants have positive relationship with total phenol contents. The genotype Safeda Mulgoa had six times higher amount of phenolics compared to Rajawala. Kjersti *et al.* (2012) reported wider variability for phenols i.e., 57 to 133 mg per 100 g of fruit weight. in 27 genotypes of strawberry, which was lower than the Safeda Mulgoa.

Similar to phenols the highly significant differences were found for total flavonoids content which varied from 10.33-49.00 mg 100g⁻¹ among different genotypes. The polyembryonic genotype Nekkare had highest total flavonoids content (49.0 mg 100g⁻¹) followed by Santra (34.0 mg 100g⁻¹), while lowest was found in genotype NR-122 (10.3 mg 100g⁻¹). Monaco *et al.* (2014) reported high amount of total flavonoids content (14.16 mg mangiferin per 100g) in the pulp of Palmer variety, which was almost 3-4 times lower than genotype Nekkare.

Mango is one of the fruits having high amount of carotenoids and more than 70 per cent of β -carotene. Carotenoids contain α -carotene, β -carotene and lycopene, and among them β -carotene is the most prevalent one. A wide range of variability (0.48-7.50 mg 100g¹¹) exists for total carotenoids among genotypes. The genotype Saheb Pasand possessed high amount of total carotenoids (7.50 mg 100g¹¹) followed by Murshidabad (6.93 mg 100g¹¹), Thampayan (6.47 mg 100g¹¹) and Sensation (6.37 mg 100g¹¹), while carotenoids content was

observed lowest in Kasi Ratnalu (0.48 mg 100g¹). Saheb Pasand had higher amount of carotenoids compared to Palmer variety (2.6 mg 100g¹) as reported earlier by Ribeiro *et al.* (2007) and five times (1.2 mg per 100g) higher than papaya (Yahia *et al.*, 2009). The results clearly indicate that a wide range of variability exists for biochemical compounds in mango gene pool and it varies from genotype to genotype. Hence, it is important to study them in all the commercial and noncommercial varieties for identification of superior parents. The morphological and molecular diversity was worked for East and North Indian heirloom varieties of mango (Anju Bajpai, 2015; Srivastava, *et al.*, 2012).

The L*, a*, b* values were recorded for pulp of all the genotypes by using ColorLlite Sph 850 spectrophotometer and values are represented in Table.1. The L* values indicated lightness of the pulp where it varied from 36.41 to 66.11 and a* values varied from negative to positive (-0.5 to 21.74). But all the b* values were positive and ranged from 19.76 to 56.10. It indicated the dominance of yellowness in pulp colour over the redness. Highest a* value (21.74) was found in the genotype sensation, followed by Saheb Pasand (21.54) and Chandrakaran (20.45). The b* values were found highest in genotype Chandrakaran (56.11) followed by Rataul (54.44) and Murshidabad (53.31). The results indicated that total carotenoids have positive relationship with a* and b* values. Ribeiro et al. (2007) have also reported that positive values of a* and b* are attributed to the carotenoids present in the mango pulp.

The knowledge of genetic characters such as heritability, PCV, GCV among the traits under selection is very useful for assessing the genetic progress in breeding programme and developing effective breeding strategies (Falconer and Mackay, 1996). The estimates of genetic parameters for biochemical compounds were worked out and are presented in Table 2. The knowledge of genetic variation among different fruit characters and their heritability is most important in highly out crossing species like mango (Rajan et al., 2009).

In the present study, genetic estimates for biochemical compounds revealed the magnitude of phenotypic variance, and phenotypic co-efficient of variance (PCV) was higher than the genotypic variance and genotypic co-efficient of variance (GCV) for all the characters indicating the presence of environmental effect on expression of traits. However, the difference between PCV and GCV for all the traits was narrow meaning that the traits were less influenced by environment. The high values of PCV and GCV recorded for total antioxidants, phenols, flavonoids and carotenoids indicated the presence of high genetic variability and better scope for selection and improvement. Similar results were observed for morphological traits such as fruit weight (20.84), fruit length (11.57), fruit peel (19.61) by Bhowmick and Banik (2008) and for bio-chemical components such as ascorbic acid (40.55), βcarotene (27.59) and total phenols (45.98) by Himabindu et al. (2016) in mango for PCV.

 Table 1 : Variability in mango genotypes for major biochemical compounds and colour characteristics

Genotypes	Total antioxidants (µmol Trolox 100g⁻¹)	Total phenols (mg 100g ⁻¹)	Total flavonoids (mg 100g ⁻¹)	Total carotenoids (mg 100g ⁻¹)	L*	a*	b*
Angoordana	0.69	76.94	19.00	4.30	44.08	7.88	35.24
Chausa	0.54	52.92	17.00	1.03	57.25	7.04	41.15
Rajawala	0.14	24.44	14.00	2.50	54.58	10.28	43.37
Merkeara	0.44	43.19	12.67	0.91	51.76	-0.03	19.76
Thampayan	0.37	30.69	15.00	6.47	47.54	13.89	37.60
Hushnara	0.73	65.97	32.33	0.85	45.02	2.82	25.94
Kasi Ratnalu	0.68	70.56	14.67	0.48	66.11	-0.06	12.56
Jawahar	0.42	44.03	19.00	2.99	60.69	10.76	42.47
Lal Mani	0.66	88.61	19.00	2.25	54.18	10.43	46.54
Surkha Thakurbag	0.39	42.78	18.33	5.66	56.69	10.99	49.96
Menaka	0.26	37.92	12.67	1.21	54.60	6.48	38.02
Allahabadi Chausa	0.35	34.72	24.00	3.80	52.78	16.36	52.48
Serehayat	0.55	74.31	18.67	1.41	58.65	7.05	43.86
Eldon	0.44	52.92	15.33	3.53	52.91	14.36	45.74
Neeleshwari	0.98	99.03	22.00	3.77	59.94	10.34	48.14
NR-122	0.22	24.72	10.33	0.79	57.79	1.90	25.32
Kunjlata	0.29	39.44	12.67	1.45	55.96	9.26	47.71
Amin	0.22	35.97	13.00	0.86	53.51	17.43	49.17
Samar Bahist Rampur	0.45	38.19	18.67	2.74	54.33	9.18	41.28
Arka Aruna	0.24	30.69	17.00	0.88	51.47	5.62	37.90
Mithuwa	0.51	54.03	33.00	3.50	41.29	11.02	32.46
Khader	0.85	78.06	20.00	1.83	55.89	9.00	36.29
Brindabani	0.45	137.50	15.67	2.91	59.59	14.97	44.49
Bathuie Katikee	0.67	60.00	32.00	3.32	45.92	17.43	39.52
Rataul	0.62	65.14	25.33	4.55	55.44	19.45	54.44
Gulabi	0.28	33.47	13.00	2.54	54.28	3.30	37.18
Chandrakaran	1.09	103.06	49.00	5.01	58.78	20.45	56.11
Nekkare	1.33	108.61	23.33	2.13	42.05	15.84	45.15
Vellaikolumban	0.73	61.67	23.67	1.98	44.24	11.92	37.85
Safeda Mulgoa	1.59	148.33	21.00	2.41	54.41	3.99	40.09
Neelgoa	0.51	36.11	18.67	1.55	48.00	10.22	30.34
Saheb Pasand	0.56	56.25	22.67	7.50	52.62	21.54	49.16
Konkan Ruchi	0.73	75.56	13.33	2.64	36.42	10.31	37.99
Murshidabad	0.56	55.97	19.67	6.93	54.52	16.15	53.31
Santra	0.76	61.39	34.00	5.00	48.10	18.69	41.17
Chilta Khas	0.75	35.28	24.33	2.71	38.08	9.64	34.32
Mylepelian	0.44	39.44	20.33	3.05	51.76	8.05	45.68
Rumani	0.34	27.78	14.33	1.58	59.72	6.58	34.63
Bhogal Shah	0.63	44.03	19.00	2.77	45.84	14.71	40.50
Janardan Pasand	0.72	59.03	20.33	1.11	58.38	2.82	32.97
Kalapahar	0.72	35.69	18.33	1.36	43.14	7.90	38.32
•	0.66	49.86	20.00	2.36	50.68	8.89	41.14
Sadaphal	0.85	79.03	29.67	3.43	57.12	13.30	49.01
Ratna	0.65	61.67	30.67	3.87	44.74	12.77	39.38
Benazir							
Gootee Madhaya Pao Pasand	0.53	38.61	21.00	3.63	42.37	17.00 15.31	43.52
MadhavaRao Pasand	0.27	22.64	19.00	3.80	41.19	15.31	38.04
EC-95862	0.74	70.83	26.67	4.43	46.29	16.95	37.26
PFN-11	0.36	52.22	15.00	1.45	49.61	5.50	32.14
Manjeera	0.30	35.69	11.33	1.15	58.36	4.58	35.69
Sensation	0.41	54.31	23.00	6.37	49.20	21.74	38.78
CD @ 5%	0.08	12.40	4.01	0.50	11.98	3.70	11.48
CV (%)	6.964	9.986	9.148	8.12	10.84	15.93	13.38

All the values shown here are average of 3 replicates for each genotype and each biochemical compounds and colour parameter

Table 2: Estimates of genetic parameters for biochemical compounds in mango genotypes

Genetic parameters	Total antioxidants	Total phenolics	Total flavonoids	Total carotenoids	
F-Value	155.46	70.35	44.46	162.70	
Mean	0.56	56.99	20.45	2.90	
Range Minimum	0.14	22.64	10.33	0.48	
Maximum	0.59	148.33	49	7.5	
CV (%)	6.964	9.986	9.148	8.12	
$\sigma^2 g$	0.078	730.193	50.812	2.970	
σ^2 e	0.002	31.588	3.508	0.055	
$\sigma^2 p$	0.080	761.781	54.319	3.025	
PCV	50.285	48.434	36.034	60.054	
GCV	49.803	47.419	34.851	59.505	
H_b^2	98.095	95.853	93.543	98.179	
GA	0.570	54.499	14.202	3.518	
GAM (%)	101.612	95.636	69.437	121.458	

GA = Genetic advance, H_b^2 = Heritability in broad sense; σ^2 g = genotypic variance; σ^2 e = environment variance; σ^2 p = Phenotypic variance; σ^2 coefficient of variability

Table 3: Correlation coefficients for biochemical compounds and colour value of mango genotypes

	Total antioxidants	Total phenols	Total flavonoids	Total carotenoids	L*	a*	b*
Total antioxidants	1.00						
Total phenols	0.82	1.00					
Total flavonoids	0.53	0.35	1.00				
Total carotenoids	0.12	0.09	0.40	1.00			
L*	-0.06	0.13	-0.23	-0.16	1.00		
a*	0.12	0.12	0.49	0.73	-0.26	1.00	
b*	0.12	0.19	0.28	0.52	0.11	0.68	1.00

GCV alone is not sufficient to estimate the extent of heritable variability in population. Therefore estimation of heritability is also important. Heritability estimates were high for all the traits which indicated the phenotype of that trait strongly reflects its genotype. The heritable values ranged from 93.54 to 98.17 among the biochemical compounds. Himabindu et al. (2016) and Gill and Navprem (2015) have also reported similar results for heritability in mango for morphological and biochemical characters such as fruit weight (96.31), fruit length (93.19), pulp weight (98.6) ascorbic acid (97.75), β-carotene (90.96) and total phenols (98.86). According to Jonshon et al. (1955), an estimated heritability associated with genetic advance is more reliable compared to heritability alone for prognosticating the impact of selection. High heritability accompanied by high genetic advance is mainly referred to the action of additive genes (Panse, 1957). So it can be concluded that different biochemical compounds components under the study can be improved though selection and hybridization.

The correlation coefficients among biochemical compounds and L*, a*, b* values studied among different genotypes are presented in Table 3. The results revealed that the total antioxidants had significant positive correlation with total phenols content (0.82) and total flavonoids (0.53). Total

flavonoids showed higher positive correlation values with total carotenoids (0.40), however total carotenoids content were highly dependent on a* (0.73) and b* (0.52) values. Existence of highly positive relationship between the total phenolics content and antioxidant activity has been reported in several plant species (Chen and Yen, 2007). The a* values also showed positive correlation with b* values. The results clearly indicated that yellowness and redness of fruit is due to higher carotenoids content. If a* and b* values are higher, the carotenoids content will be more. The study concludes that wider genetic variability for biochemical compounds was found among genotypes. The genotypes Saheb Pasand, Murshidabad, Sensation and Thampayan were identified as rich source of carotenoids and Safeda Mulgoa as a good source of phenols and total antioxidants. This information will be helpful in identifying superior parents for developing nutritionally rich hybrids in future.

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