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Studies on biochemical profiling of 72 avocado (*Persea americana* Mill.) accessions

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Abstract
The important biochemical traits were analyzed from pulp of 72 accessions was analyzed and recorded wide variability among the accessions. The accession, CHES-PA-X-2 had the highest dry matter content of 34.40 percent followed by CHES-PA-VIII-11 (33.30%) and CHES-PA-VIII-13 (30.24%), the lowest dry matter content was noted in CHES-PA-VIII-10 (15.00%). The fat content in fresh pulp among 72 accessions was ranged between 6.36 to 18.73 percent. The accession, CHES-PA-III-2 recorded the maximum fat content followed by CHES-PA-X-2 (16.37%), CHES-PA-X-5 (13.56%), CHES-PA-X-6 (13.11%) and CHES-PA-XIII-8 (12.45%), while the highest phenolic content was noted in CHES-PA-IV-11 (103.24 mg GAE/100 g). The moisture content had strong significant and negative correlation with fat content (-0.081) while positive correlation with phenol content (0.030). The dry matter content of pulp showed highly positive correlation for fat content (0.278) carotenoids content (0.122). The fat content expressed positive correlation with dry matter content of pulp (0.211) and negative correlation with moisture content (-0.121). The distinct avocado accessions exhibiting important horticultural traits such as fruit size, shape, pulp quality, peel colour, peel thickness etc. could be important in selecting the superior accessions.

Keywords: Biochemical, avocado, phenols and fat content

Introduction

Avocado, *Persea americana* (Mill.), is one of oldest cultivated fruit crop with rich history and belongs to the family Lauraceae and (Galindo-Tovar *et al.*, 2007) [11]. It is often regarded as most nutritive fruit owing to its creamy texture, unique taste and immense nutritional composition. Avocados originated in Mexico and Central America, possibly from more than one wild species (Dreher and Davenport, 2013) [9]. Avocado is subtropical fruit tree and is now commercially grown in more than 50 countries worldwide of which Mexico, Dominican Republic, Peru, Colombia, Indonesia, Kenya, Brazil, Haiti, Chile, and Israel are major Avocado producing countries (Anonymous, 2021) [2]. Avocados have unique nutritional profile, which provides sustainable amount of monounsaturated fatty acids and contains lots of fibre, proteins, carbohydrates, minerals and vitamins like B-vitamins, vitamin K, potassium, copper, magnesium, manganese, vitamin E, vitamin C, and folate (Dreher and Davenport, 2013) [9].

In India Avocado is not grown commercially, though the climatic conditions and soil requirements in the region are ideal for their large scale cultivation. Its cultivation is mainly confined to South Tropical States like Tamil Nadu (Lower Palni Hills, Yercaud, Kodaikanal, Ooty, etc.), Kerala, Karnataka (Kodagu) and North Eastern Himalayan State of Sikkim with the area of around few thousand acres. In Kodagu, avocado is grown as one of the mixed crops in coffee-based cropping system. Almost each house is maintaining few plants of avocado and lots of variability exits in Kodagu and adjoining areas (Tripathi *et al.*, 2016) [21]. One of the major limitation of Avocado cultivation in India is lack of consumer preference owing to its nature of taste. The multi-nutrient rich Avocados have huge potential in domestic markets, as the fruits having highest energy value compared to other fruit crops, to achieve household nutritional security. Nevertheless, due to copious nutritional benefits and increasing consciousness among consumers about health concern, avocados may find equitable place in Indian markets in due course.

During recent past its cultivation is gaining momentum as cash crop, owing to its high nutritional value, high market potential and less resource demanding nature of crop. The Avocadoes were mainly grown through seed for over 100 years and has adapted to a wide range of topography, habitats and climates. As a result, a large diversity has accumulated in this germplasm. So far, only few studies have been conducted to assess the diversity of this germplasm from Karnataka and Tamil Nadu region. Therefore, the present study was undertaken to uncover the genetic diversity of Avocado germplasm through biochemical profiling.

Materials and Methods

Extensive surveys were made in Southern tropical states like Tamil Nadu, Kerala, Karnataka (Coorg), and North-Eastern Himalayan state of Sikkim to meet the above objective, as irregularity in bearing is major hindrance in avocado cultivation. These collected accessions are being maintained at experimental farms of ICAR – IIHR – Central Horticultural Experiment Station, Chettalli with regular orchard management practices. A total adds of 72 accessions were characterized for biochemical composition.

The various biochemical traits viz. total phenols, total carotenoids, oil content, moisture content, dry matter content, and total soluble solids were analysed from fresh pulp of 72 accessions. The pulp was extracted from mature fruits and homogenized using mixture grinder and the homogenized pulp was used for estimation of all compounds. Phenols were estimated according to the procedure given by Singleton and Rossi, (1965) [20] with slight modifications. Total carotenoids content of 72 accessions were analysed by spectrophotometric method (Lichtenthaler, 1987) [15]. The Cupric Reducing Antioxidant Capacity of 72 avocado accessions was estimated by the procedure standardized by Apak *et al*. (2004) [3]. The Avocado oil extraction was performed using a Soxhlet apparatus according to the method described by AOAC (2007). The moisture content was estimated using the method developed by Ranganna (1986) [18]. The total soluble solids were determined using the Hand Refractometer and the dry matter content of the pulp was estimated after complete drying of fresh pulp in hot air oven for 36-48 hours at 60 ºC. Completely randomized design was used to estimate the critical difference and coefficient of variation using SPSS.16 software.

Results and Discussion

Total phenols

The total phenol content among the studied accessions was differed significantly and it was ranged between 46.02 to 103.24 mg GAE/100 g. The highest phenolic content was noted in CHES-PA-IV-11 (103.24 mg GAE/100 g), followed by CHES-PA-VI-2 (102.65 mg GAE/100 g), CHES-PA-VIII-10 (98.24 mg GAE/100 g) and CHES-PA-XI-10 (95.59 mg GAE/100 g) (Table 1). However, it was significantly lower in CHES-PA-IX-2 (46.02 mg GAE/100 g) followed by CHES-PA-III-2 (47.58 mg GAE/100 g), CHES-PA-VII-7 (49.05 mg GAE/100 g), CHES-PA-X-10 (53.62 mg GAE/100 g). From above data, it was clearly noted that the maximum number of accessions had phenolic content above 50 mg GAE/100 g. The similar variation for phenol content (1.30 to 38.82 mg GAE g-1) in different accessions of avocado was reported by Amado *et al*. (2019) [1] and noticed wider range (3.75 to 8.92 mg/100 g) than the Chinese accessions (Liu *et al*., 2020) [16].

Total antioxidant activity

Total antioxidant activity by CUPRAC method was estimated for 72 accessions and observed significant differences among the studied accessions. The accession, CHES-PA-VIII-3 recorded the highest antioxidant activity (0.81 (µmol Trolox/100 g) among the accessions followed by CHES-PA-VIII-2 (0.79 µmol Trolox/100 g), CHES-PA-X-8 (0.66 µmol Trolox/100 g) and CHES-PA-X-7 (0.61 µmol Trolox/100 g) (Table 1). Further, significantly lower antioxidant activity was noticed in CHES-PA-VII-2 (0.22 µmol Trolox/100 g) followed by CHES-PA-III-7 (0.23 µmol Trolox/100 g), CHES-PA-III-8 (0.24 µmol Trolox/100 g) and CHES-PA-XI-8 (0.27 µmol Trolox/100 g). Similarly, Rodriguez-Carpena *et al*. (2011) [19] reported the highest 2.48 mmol Trolox/g f.w. CUPRAC activity in avocado variety Furete as compare to other accessions studied, which is higher than the accessions in the present study. Further, the similar results were also noted by Wang *et al*. (2010) [23] for FRAP activity in avocado pulp (2.93 µmol Fe2+ /g FW), which are in agreement with the results of present study.

Total carotenoid content

The total carotenoids among the studied accessions was varied significantly from 1.46 to 4.89 mg / 100 g. The maximum carotenoid content was observed in CHES-PA-VI-4 (4.89 mg /100 g) which was on par with CHES-PA-VI-12 (4.46 mg /100 g) and CHES-PA-IX-12 (4.35 mg /100 g). However, it was significantly lowest in CHES-PA-II-10 (1.46 mg /100 g) which was on par with CHES-PA-III-9 (1.50 mg /100 g), CHES-PA-I-12 (1.76 mg /100 g) and CHES-PA-VI-15 (1.80 mg /100 g) (Table 1). From the above results, it can be noted that the maximum number of studied accessions had the total carotenoid content above 2.00 mg /100 g. The accession CHES-PA-VI-4 had higher amount of carotenoids than Breba (4.63 mg β-Carotene kg^{-1}) and Algarvian Hass variety (0.815±0.201 mg/100 g) as reported by Krumreich *et al*. (2018) [14] and Vinha *et al.* (2013) [22] respectively.

The fat content

The fat content in fresh pulp among 72 accessions was ranged between 6.36 to 18.73 percent. The accession, CHES-PA-III-2 recorded the maximum fat content followed by CHES-PA-X-2 (16.37%), CHES-PA-X-5 (13.56%), CHES-PA-X-6 (13.11%) and CHES-PA-XIII-8 (12.45%), while it was significantly lower in CHES-PA-VII-7 (6.36%) which was on par with CHES-PA-II-2 (7.98%) and CHES-PA-VI-2 (7.30%) (Table 1). Among the studied accessions, the majority of the accessions had fat content above 10 percent. Avocado is very well acclaimed for its oil due to the presence of high mono and poly unsaturated fatty acids and is one of the important parameters to indicate the maturity of fruit (Donadio, 1995) [7]. The similar trend for fat content (2.19 to 26.12%) were also reported by Oliveira *et al*. (2013) [17] in accessions of Brazilian avocados and Bayram *et al*. (2012) [5] in Turkey avocado accessions (6.41-19.1%)

Dry matter content

The dry matter content among 72 accessions was differed significantly and ranged between 15.00 to 34.40 percent. The accession, CHES-PA-X-2 had the highest dry matter content of 34.40 percent followed by CHES-PA-VIII-11 (33.30%) and CHES-PA-VIII-13 (30.24%), the lowest dry matter content was noted in CHES-PA-VIII-10 (15.00%) which was on par with CHES-PA-IX-7 (15.38%) and CHES-PA-XI-10 (15.41%), followed by CHES-PA-VIII-6 (16.06%) and CHES-PA-VIII-5 (16.11%) (Table 1). Among the studied accessions, the maximum number of accessions had the dry matter content above 20%. The wide variation for dry matter content was (28.23 to 36.43%) also reported by Espinosa Alonso *et al*. (2017) [10] in avocado. Bayram *et al*. (2012) [5] also noted the wide variability (17.84-29.93%) for dry matter content in 29 varieties of avocado which are grown in Turkey.

Moisture content
The significant differences were noted for moisture content among 72 accessions and it was ranged between 66.70 to 85.38 percent. The highest moisture content was recorded in CHES-PA-VI-12 (85.58%), which was on par with CHES-PA-XIII-5 (85.38%), CHES-PA-X-11 (85.08%) and CHES-PA-VII-7 (85.08%). However, it was significantly lower in CHES-PA-II-9 (66.70%) followed by CHES-PA-XIII-8 (67.47%) and CHES-PA-X-12 (67.52%) (Table 1). The accessions representing higher moisture content are inferior in pulp quality and also moisture content is negatively correlated with oil content. Rodriguez-Carpena *et al*. (2011) [19] also reported 80.25% moisture content in pulp of Fuerte variety and Liu *et al*. (2020) [16] in avocado.

Total soluble solids
The total soluble solids content among the studied accessions showed the significant differences. The maximum TSS was observed in CHES-PA-VI-4 (8.89 0 Brix) which is on par with CHES-PA-X-11 (8.56 0 Brix) and CHES-PA-II-10 (8.26 0 Brix), however significantly lower TSS was noted in CHES-PA-VI-15 (4.62 followed by CHES-PA-IV-7 (5.87 0 Brix) and CHES-PA-III-9 (5.97 ^{0} Brix) (Table 1). Among the accessions under the study, the maximum number of accessions had TSS content above 5 0 Brix. TSS of collected avocados was lower than mango (Gomez *et al*., 2015) [13] and Jack fruit (Aseef *et al*., 2017) [4].

Table 1: Variability for carotenoids, TSS, moisture, fat content and dry matter content of pulp in avocado accessions

Acc. No.	Moisture content (%)	Dry matter content (%)	TSS (^{0}B)	Fat (%)	Total phenol content (mg GAE/100 g)	Total carotenoid content (mg /100 g)	Total antioxidants (µmol Trolox/100 g)
CHES-PA-III-2	81.86	27.38	7.79	18.73	47.58	3.15	0.32
CHES-PA-III-6	84.08	18.53	7.44	10.63	53.18	2.96	0.29
CHES-PA-III-7	83.08	22.19	7.23	11.37	55.25	2.66	0.23
CHES-PA-I-12	83.89	17.53	6.30	11.04	54.83	1.76	0.37
CHES-PA-III-8	83.95	20.74	6.21	8.60	64.89	1.50	0.24
CHES-PA-III-9	79.50	21.33	5.97	13.61	49.05	2.07	0.62
CHES-PA-II-2	78.18	20.50	6.71	7.98	52.91	4.60	0.51
CHES-PA-II-4	85.00	19.76	7.12	11.36	82.61	2.89	0.31
CHES-PA-II-9	66.70	20.68	7.59	11.30	58.70	1.96	0.28
CHES-PA-II-6	69.77	22.78	7.99	9.72	53.62	3.15	0.33
CHES-PA-II-10	78.04	14.20	8.26	10.99	61.88	1.46	0.41
CHES-PA-II-11	79.10	19.51	7.40	9.00	51.37	2.18	0.36
CHES-PA-IV-1	71.23	20.25	7.55	11.02	67.87	2.85	0.31
CHES-PA-IV-5	76.84	22.19	6.91	11.01	79.34	2.71	0.42
CHES-PA-IV-7	79.05	17.23	5.87	10.26	95.59	2.05	0.44
CHES-PA-IV-8	78.17	20.74	4.66	10.72	58.88	3.06	0.47
CHES-PA-IV-11	84.59	21.33	5.31	8.87	103.24	3.66	0.52
CHES-PA-IV-13	75.77	20.50	5.91	8.05	67.80	3.03	0.25
CHES-PA-VI-1	80.13	19.76	6.47	9.47	28.80	3.43	0.34
CHES-PA-VI-2	78.70	20.68	8.25	7.30	102.65	4.35	0.39
CHES-PA-VI-4	78.52	22.78	8.89	10.22	73.25	4.89	0.46
CHES-PA-VI-5	84.15	14.20	7.85	12.46	72.67	3.48	0.43
CHES-PA-VI-7	78.11	19.51	6.97	10.35	109.75	2.85	0.32
CHES-PA-VI-12	85.58	20.25	6.81	11.73	81.82	4.46	0.52
CHES-PA-VI-15	77.48	22.19	4.61	16.37	80.07	1.80	0.49
CHES-PA-VII-1	79.76	18.21	7.01	9.96	61.50	2.86	0.35
CHES-PA-VII-2	81.36	20.74	6.40	13.56	55.64	2.15	0.22
CHES-PA-VII-4	81.67	21.33	6.45	13.11	54.83	2.07	0.31
CHES-PA-VII-6	81.86	20.50	6.37	6.36	64.89	4.60	0.30
CHES-PA-VII-7	84.08	19.76	6.36	11.01	49.05	2.89	0.28
CHES-PA-VII-8	83.08	20.68	6.31	11.36	52.91	1.96	0.44
CHES-PA-VII-10	83.89	18.64	6.67	10.95	82.61	3.15	0.63
CHES-PA-VIII-1	83.95	18.34	8.57	10.22	58.70	1.76	0.71
CHES-PA-VIII-2	77.69	18.14	8.59	9.57	53.62	2.18	0.79
CHES-PA-VIII-3	81.65	15.93	8.73	8.34	61.88	2.85	0.81
CHES-PA-VIII-4	79.78	16.93	5.57	8.81	51.37	2.71	0.33
CHES-PA-VIII-5	65.61	16.11	8.06	8.47	67.87	2.05	0.24
CHES-PA-VIII-6	71.97	16.06	5.67	7.42	79.34	3.06	0.33
CHES-PA-VIII-8	81.40	20.51	5.24	10.68	95.59	3.66	0.25
CHES-PA-VIII-9	75.82	21.83	5.66	10.83	58.88	3.03	0.31
CHES-PA-VIII-10	71.37	15.00	5.29	12.41	98.24	3.43	0.34
CHES-PA-VIII-11	78.46	33.30	5.64	7.20	87.58	3.75	0.29
CHES-PA-VIII-13	79.51	30.24	7.07	13.89	86.55	2.41	0.42
CHES-PA-IX-1	79.36	21.97	7.47	10.05	95.85	2.79	0.38

CHES-PA-IX-2	83.73	20.91	7.14	10.86	46.02	3.17	0.43
CHES-PA-IX-3	83.89	20.00	7.67	11.48	96.01	2.25	0.46
CHES-PA-IX-4	83.95	17.76	6.58	7.55	66.94	2.30	0.36
CHES-PA-IX-5	79.50	18.69	6.45	12.34	68.58	1.87	0.31
CHES-PA-IX-6	78.18	17.60	6.57	7.03	76.57	1.97	0.39
CHES-PA-IX-7	85.00	15.38	5.91	11.94	76.55	3.86	0.45
CHES-PA-IX-8	66.73	16.67	6.46	9.47	85.66	3.83	0.48
CHES-PA-IX-9	69.77	16.47	7.35	7.30	88.76	3.03	0.31
CHES-PA-IX-10	78.04	17.49	6.59	10.22	96.75	3.43	0.33
CHES-PA-IX-12	79.10	22.31	8.33	12.46	81.82	4.35	0.26
CHES-PA-IX-13	78.18	18.36	6.56	10.35	80.07	4.89	0.54
CHES-PA-IX-14	85.00	20.23	5.39	11.73	61.50	3.48	0.47
CHES-PA-X-2	67.71	34.40	5.69	16.37	55.64	2.85	0.36
CHES-PA-X-3	69.77	22.19	6.51	9.96	54.83	4.46	0.39
CHES-PA-X-5	78.04	17.53	6.45	13.56	64.89	1.90	0.43
CHES-PA-X-6	79.10	20.74	6.57	13.11	59.05	2.86	0.56
CHES-PA-X-7	75.35	21.33	5.91	6.36	52.91	2.97	0.61
CHES-PA-X-8	83.95	20.50	6.46	11.01	82.61	2.89	0.66
CHES-PA-X-9	79.50	19.76	7.35	11.36	58.70	2.44	0.29
CHES-PA-X-10	78.18	20.68	7.99	11.01	53.62	1.82	0.28
CHES-PA-X-11	85.08	22.78	8.56	10.26	61.88	1.97	0.36
CHES-PA-X-12	67.52	14.20	8.31	10.72	51.37	2.09	0.31
CHES-PA-X-13	69.77	19.51	6.48	8.87	67.87	5.42	0.38
CHES-PA-XI-8	78.04	22.15	7.81	8.05	79.34	2.14	0.27
CHES-PA- XI-10	79.10	15.41	6.25	9.47	95.59	2.13	0.42
CHES-PA-XIII-1	78.18	24.23	5.64	7.30	58.88	3.15	0.53
CHES-PA-XIII-5	85.38	19.87	5.91	10.22	67.79	1.84	0.47
CHES-PA-XIII-8	67.47	21.31	6.46	12.45	67.57	2.26	0.41
CV	4.96	13.48	6.28	11.32	16.23	3.89	3.22
CD @ 5%	6.241	7.25	4.22	7.11	9.78	1.98	1.78
SE (m) ±	1.09	2.11	0.96	1.48	2.21	0.29	0.32

Correlation coefficients for biochemical characters

The correlation coefficients for biochemical traits revealed significant positive and negative correlation among the traits (Table 2). The moisture content had strong significant and negative correlation with fat content (-0.081) while positive correlation with phenol content (0.030). The dry matter content of pulp showed highly positive correlation for fat content (0.278) carotenoids content (0.122). The fat content expressed positive correlation with dry matter content of pulp (0.211) and negative correlation with moisture content (-0.121). The phenols content of pulp showed highly positive correlation with carotenoid content (0.209). Further, the total soluble solids expressed significantly positive correlation with total antioxidant content (0.046). The similar positive correlation was reported by Wang *et al*. (2010) [23] for total phenols and antioxidants activity (FRAP and DPPH) in avocado pulp. The negative correlation between oil content and moisture content in different varieties of avocado were also reported by Bezuidenhout and Bezuidenhout (2014) [6]. Further, the similar trend for compositions avocado oil was confronted by Galvo *et al.* (2014) [12] in Fortuna, Barker and Collison varieties.

Table 2: Estimation of Correlation Coefficient for Biochemical characters

-	Moisture content	Dry matter content	Total soluble solids	Fat content	Total phenols	Total carotenoids	Total antioxidant
Moisture content	1.000						
Dry matter content	-0.018	1.000					
Total soluble solids	-0.036	-0.129	1.000				
Fat content	-0.081	0.278	-0.053	1.000			
Total phenols	0.003	-0.077	-0.109	0.129	1.000		
Total carotenoids	-0.077	0.122	-0.076	-0.202	0.203	1.000	
Total antioxidant	0.199	-0.117	0.048	-0.022	0.209	0.052	1.000

Values in bold are different from 0 with a significance level alpha=0.05

Conclusion

The assessment of diversity in the form of biochemical traits revealed existence of wider genetic diversity. The implications of present investigation revealed that though India is not the centre for origin of avocado, but a large quantum of genetic diversity exists in the Indian avocadoes. The distinct avocado accessions exhibiting important horticultural traits such as fruit size, shape, pulp quality, peel colour, peel thickness etc could be important in selecting the superior accessions or their use as parent in the avocado breeding program to meet the diversified consumer demands.

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