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Seed Parameters, Oil Yield and Fatty Acid Profile of *Garcinia gummi-gutta* from Central Western Ghats, India

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***Abstract*:** The study conducted on *G. gummi-gutta* seeds collected from nine distinct locations across the Uttara Kannada district in Karnataka, a region situated within the central Western Ghats. The study employed the Soxhlet extraction method to determine the percentage of oil yield and utilized GC-MS analysis to assess the fatty acid composition. There were minimal statistical differences in seed characteristics and fatty acid profiles among the nine sites. The differences in oil yield across these sites did not demonstrate statistical significance. The study revealed highest of 46.64 percent oil yield from individual tree with an average of 39.32 percent overall. The seed oil extracted exhibited characteristics of being solid at room temperature and had an unpleasant flavour and fragrance, colours ranging from brown to creamy tones. GC-MS analysis identified the eight distinct fatty acids in the oil samples. Predominantly, oleic acid and stearic acid were found in all samples. Oleic acid ranged between 54.56 to 59.79 percent while stearic acid varied from 33.57 to 40.33 percent. The collective proportions of saturated and unsaturated fatty acids were at 60.15 percent and 39.85 percent, respectively.

***Keywords*:** *Garcinia gummi-gutta,* Seed parameters, Oil yield, Fatty acid profile, GC-MS

Garcinia gummi-gutta (L) Rob. (syn. *Garcinia cambogia*) known as Malabar Tamarind. It's a medium sized evergreen tree with average height of 20 m. Flowering starts January to March and fruits start to mature in June in India (Karthik and Ramana 2023). People usually collect the naturally dropped mature fruits, sometimes and also harvest the same manually from the tree. The seeds are not utilized most cases. The sharp sour tasting fruit rind traditionally used as flavoring agent in rural part of southern India and also as remedy to digestive disorders and intestinal parasites, rheumatism, bowel complaints (Semwal et al 2015). Apart from rind seeds are also used for various traditional purposes viz. moisturizer in winter, treating cracks, rashes and sometimes burn wounds, dehydration and diarrhea treatments (Mohammed et al 2017). Seeds oil is edible and usually ranges between 35 to 45 percent on average. The seed oil extracted has very good potential to be used in bio-diesel industries as a raw material (Subramani et al 2018). Various climatic and environmental factors can influence the growth and establishment of trees that could show great variance (Rangwala and Miller 2012). The study was aimed at recording the different parameters viz. seed parameters, oil yield and respective fatty acid composition from nine locations with different climatic and environmental conditions from central Western Ghats of Karnataka, India.

MATERIAL AND METHODS

Study area and collection of samples: The study was conducted in Uttara Kannada district of Karnataka, a part of Central Western Ghats, India. Nine sites were randomly selected. In each site 5 trees were chosen as replications. To maintain the homogeneity, trees in the 50–80 cm girth class were picked. Tree heights ranged from 13.7 to 16.8 m, and crown diameters ranged from 7.50 to 9.25 m (Table 1). Annual rainfall varied from 2300 to 4722 mm, and altitude ranged from 418.7 to 646.3 m. Samples were collected between July and August. The samples were immediately preserved in polythene bags and were taken to the experimental lab for further examinations.

Physical characteristics-Seed parameters: The fruit material was cut open to record number of seeds per fruit. The seed, in terms of the principal axial dimensions, that is (in cm and mm): length, thickness and width is measured using a Vernier caliper with an accuracy of 0.01 mm. These measurements were replicated three times to get mean values.

Extraction and Quantification Oil from *G. gummi-gutta* Seed Kernel

Oil extraction: The seeds were dried; dried kernels of 100 g from each tree was powdered using grinder. The sample were stored in an air-tight jar. Oil was extracted by Soxhlet extraction method using non-polar solvent (petroleum ether) in 40 to 60^{o}C temperatures.

Percentage yield of oil (w/w %): The amount of extracted oil was determined and percentage yield of the oil from each sample was calculated on the basis of weight by using following formula, expressed in w/w %:

$$\text{Percentage of oil yield} = \frac{\text{Weight of the oil}}{\text{Weight of the kernel}} \times 100$$

Chemical Characterization of Seed Oil

Methylation of sample: The reaction was done using a 50 mL shaking flask under 50°C on a reciprocal shaker. An equivalent quantity of 10 g of Garcinia oil was mixed with TL linked Fe_3O_4 nanoparticle, and thrice the quantity of methanol was added to the solution of 0.5 g. After the completion of the reaction process, the residual methanol content was distilled off completely with the help of an evaporator at 65°C under vacuum condition. The resulting *Garcinia gummi-gutta* methyl ester was then subjected to the GCMS analysis. (Subramani et al 2018).

Gas Chromatographic-Mass spectrometry analysis of oil: The Fatty acid composition of oil was determined by Gas Chromatography-Mass Spectrometry (GC-MS) technique using Shimadzu-GCMS (QP2020 NX) instrument. The GC-MS was equipped with a split injector and an ion-trap mass spectrometer detector with a fused-silica capillary column having a thickness of 1.00 µm, dimensions of 30m x 0.25mm (Agilent DB-5MS) and temperature ranges of 60 °C and 325 °C. The column temperature was programmed between 60 ºC and 250 ºC at a flow rate of 3.0 mL/min. The temperature of the injector and detector were at 250 °C and 200 °C respectively. Helium gas was used as a carrier gas at a flow rate of 46.3 cm/ sec. Components were identified by computer-aided matching of their spectra with spectra of known compounds from the NIST (National Institute of Standards and Technology, USA) libraries mass spectral databases. The compounds were quantified by the area normalization method without considering response factors. Relative amounts of individual components were calculated based on GC peak areas (Choppa et al 2015).

Data analysis: The data was analyzed using 'R'. Correlation matrix was visualized using "ggcorr" package.

RESULTS AND DISCUSSION

Seed parameters: Seeds are of brown to dark brown in colour and almost stretched bean shape of about 1.5 to 2 cm length and 0.5 to 0.7 cm breadth. With non-significant difference between the sites among majority of seed related parameters (Table 2), the average number of seeds per fruit of 5.87, fresh seed weight of 3.78 g, dry seed weight of 0.64 g, kernel weight of 0.33 g, seed length of 22.03 mm, seed breadth of 6.63 mm and seed thickness of 3.23 mm was observed in the study. Minor variations between seed traits viz. seed length and breadth along with weight of seed among different wild accessions of *G. gummi-gutta* are prevalent (Kavya et al 2016). The changes in the environment caused by latitude, altitude, rainfall, temperature, moisture, and other external factors to the soil and climate of the region where the seeds were grown, are taken into consideration as essential factors influencing the seed qualities (Singh et al 2019). Morphological variation in seed characters among the wild accession may also be because of broad adaptations of species to range of edaphic conditions (Jenner et al 2003).

Oil yield: Sites did not influence the Oil yield of *G. gummi-gutta* seeds (Table 2). The oil colour was brown to creamy white and has disagreeable odour and taste, oil becomes solid in room temperature. Ramesh and Sharanappa (2014) and George et al (2018) and Parthasarathy and Nandakishore (2014) also reported the similar findings on *G. gummi-gutta* seed oil colour and physical parameters. The average oil yield in all the sites combined was 39.32 per cent (w/w), whereas the highest of 46.64 per cent was observed in tree number thirty-eight from site S_8 (Table 3). Parthasarathy

Table 1. Geo-climatic information and tree parameters of study area

Site	Altitude (m)	Locations	Latitude	Longitude	Mean annual rainfall (mm)	Annual rainy days	Mean annual temperature (°C)	Tree height (m)	Girth (cm)	Crown diameter (m)
S_1	646.3	Huthgar	14° 42'14"	74° 75'97"	3800 - 4722	120	24	13.7	61.6	8.35
S_2	610.7	Nettikai	14° 41'85"	74° 76'05"	3800 - 4722	120	23	14.5	70.8	9.25
S_3	623.3	Halegadde	14° 45'22"	74° 70'55"	3800 - 4722	120	24	16.8	68.3	8.55
S_4	516.2	Ambadgar	14° 80'71"	74° 73'63"	2950 - 3667	100	25	17.7	73.2	8.40
S_5	508.9	Kotikoppa	14° 73'35"	74° 67'51"	2950 - 3667	100	25	16.7	72.9	8.20
S_6	522.7	Kaggundi	14° 72'66"	74° 70'23"	2950 - 3667	100	25	15.5	69.6	8.35
S_7	460.8	Nagalagar	14° 54'21"	74° 69'39"	2300 - 3263	95	27	14.4	68.5	7.55
S_8	437.8	Havinakodlu	14° 52'71"	74° 69'22"	2300 - 3263	95	27	14.4	62.0	7.50
S_9	418.7	Thotadakasige	14° 52'71"	74° 64'53"	2300 - 3263	95	27	15.9	68.8	8.25

et al. (2013) reported the highest seed oil yield of 47 per cent. Rahangdale et al (2014) stated the variability within the oil yield content is probably because of the variation in exclusive ecological conditions.

Fatty acid profile of *Garcinia gummi-gutta* seed oil: Eight different fatty acids were found through GC-MS analysis (Fig. 1) of oil sample from the nine sites. Fatty acids viz. Saturated fatty acids (Palmitic acid, Stearic acid, and Arachidic and Unsaturated fatty acids (Oleic acid, Linoleic acid, Linolenic acid, Gadoleic acid and 11- eicosanoic acid were found at the concentration of 39.85 and 60.15 percent respectively (Table 4). Oleic acid and stearic acid are found in major quantities, while, palmitic acid and linoleic acid in minor quantities. Arachidic acid, linolenic acid, gadoleic acid and 11-eicosanoic acid found in trace amounts. Patil et al (2016) reported that major fatty acid found was oleic acid in seed oil of *G. gummi-gutta*. Ajayi et al (2007) reported that predominant fatty acids in *Garcinia xanthocymus* was palmitic acid (48.5 %) and oleic acid (35 %). In study

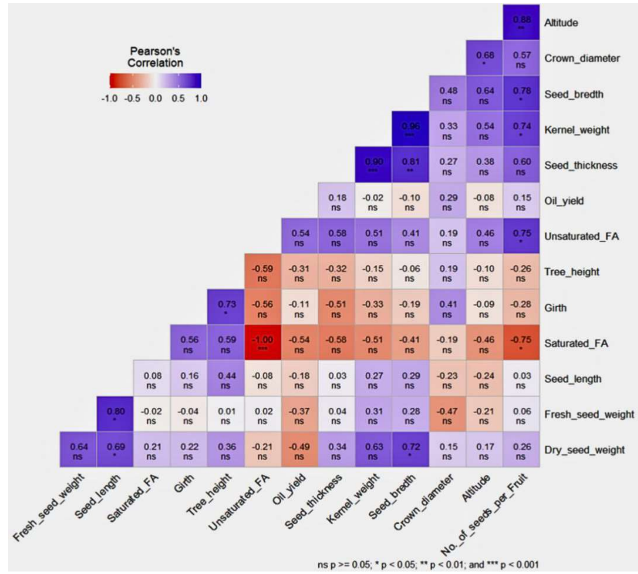

Fig. 2. Correlation heat map of site, tree, seed factors and fatty acid

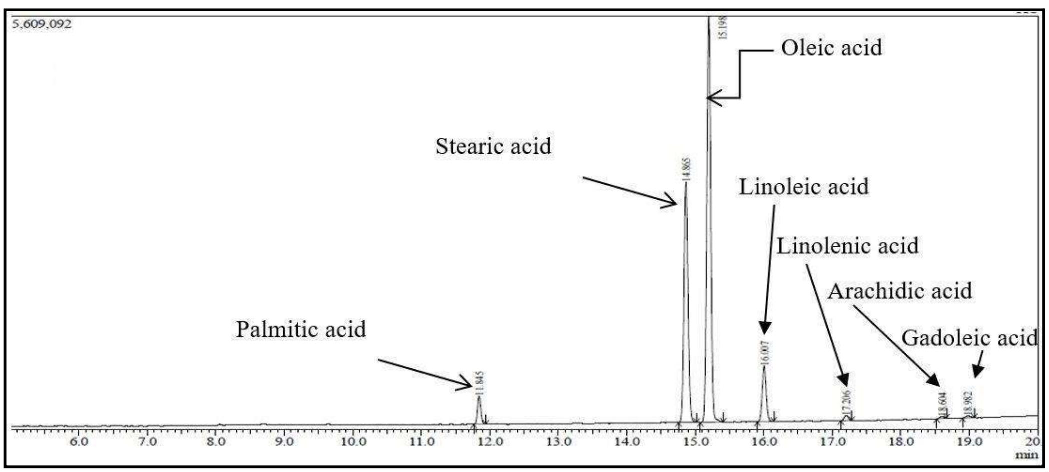

Fig. 1. Gas chromatogram of seed oil from the study site

Table 2. Seed parameters of study sites

Site	No. of seeds per fruit	Fresh seed weight (g)	Dry seed weight (g)	Kernel weight (g)	Seed length (mm)	Seed breadth (mm)	Seed thickness (mm)	Seed Oil Yield (%, w/w)
S_1	7.12 ^{a}	3.61 ^{cde}	0.60 ^{bc}	0.38 ^{a}	20.87 ^{c}	7.08 ^{a}	3.61 ^{a}	38.99 ^{a}
S_2	6.84 ^{ab}	3.14 ^{de}	0.61 ^{bc}	0.38 ^{a}	20.69 ^{c}	7.07 ^{a}	3.54 ^{a}	41.06 ^{a}
S_3	6.58 ^{ab}	3.34 ^{cde}	0.66 ^{bc}	0.43 ^{a}	21.69 ^{bc}	7.19 ^{a}	3.84 ^{a}	39.02 ^{a}
S_4	6.14 ^{bc}	5.14 ^{a}	0.84 ^{a}	0.37 ^{ab}	26.64 ^{a}	7.11 ^{a}	3.04 ^{a}	38.84 ^{a}
S_5	4.90 ^{d}	2.91 ^{e}	0.52 ^{bc}	0.15 ^{d}	20.47 ^{c}	5.47 ^{b}	1.84 ^{b}	38.78 ^{a}
S_6	5.46 ^{cd}	3.98 ^{bcd}	0.80 ^{a}	0.39 ^{a}	21.44 ^{bc}	7.16 ^{a}	3.60 ^{a}	37.90 ^{a}
S_7	5.42 ^{cd}	4.85 ^{ab}	0.58 ^{bc}	0.29 ^{bc}	22.11 ^{bc}	6.07 ^{ab}	2.84 ^{ab}	39.07 ^{a}
S_8	5.48 ^{cd}	4.31 ^{abc}	0.63 ^{bc}	0.35 ^{ab}	23.31 ^{bc}	6.55 ^{ab}	3.48 ^{a}	39.93 ^{a}
S_9	4.92 ^{d}	2.70 ^{e}	0.51 ^{c}	0.26 ^{c}	21.03 ^{c}	5.97 ^{ab}	3.28 ^{a}	40.33 ^{a}
Mean	5.87	3.78	0.64	0.33	22.03	6.63	3.23	39.32

1. Groups sharing the same letter (e.g., 'a') are not significantly different from each other.
2. Different letters (e.g., 'a' vs. 'b') indicate significant differences between the means of those respective groups.
3. The larger the differences in letters, the greater the significance of the contrast between the groups.

Table 3. Average seed oil yield percent of individual trees from the respective sites

Site	Tree number	Oil yield (%, w/w)	Average oil yield of site
S_1	T_1	39.70	38.99
	T_2	39.99	
	T_3	37.25	
	T_4	40.04	
	T_5	37.98	
S_2	T_6	39.27	41.06
	T_7	34.94	
	T_8	44.00	
	T_9	41.12	
	T_{10}	45.96	
S_3	T_{11}	39.67	39.02
	T_{12}	39.22	
	T_{13}	44.42	
	T_{14}	34.36	
	T_{15}	37.41	
S_4	T_{16}	39.90	38.84
	T_{17}	41.46	
	T_{18}	33.85	
	T_{19}	37.05	
	T_{20}	41.93	
S_5	T_{21}	37.02	38.78
	T_{22}	41.26	
	T_{23}	41.03	
	T_{24}	38.36	
	T_{25}	36.22	
S_6	T_{26}	42.75	37.90
	T_{27}	39.12	
	T_{28}	33.88	
	T_{29}	36.85	
	T_{30}	36.88	
S_7	T_{31}	45.09	39.07
	T_{32}	35.22	
	T_{33}	39.60	
	T_{34}	41.78	
	T_{35}	33.66	
S_8	T_{36}	33.67	39.93
	T_{37}	43.76	
	T_{38}	46.64	
	T_{39}	36.93	
	T_{40}	38.64	
S_9	T_{41}	40.14	40.33
	T_{42}	42.24	
	T_{43}	43.68	
	T_{44}	39.50	
	T_{45}	36.08	
Overall Mean		39.32	

There is no significant difference between the means of all sites

Table 4. Fatty acid profile of seed oil samples of *Garcinia gummi-gutta*

Fatty acids		Study sites									Mean
		S_1	S_2	S_3	S_4	S_5	S_6	S_7	S_8	S_9	
Saturated	Palmitic acid ($C_{16}H_{32}O_2$)	3.08 ^{a}	2.91 ^{ab}	2.84 ^{bc}	2.83 ^{bc}	2.68 ^{cd}	2.52 ^{d}	3.01 ^{a}	2.13 ^{e}	3.05 ^{a}	2.78
	Stearic acid ($C_{18}H_{36}O_2$)	33.57 ^{f}	34.38 ^{e}	35.69 ^{d}	37.54 ^{b}	40.33 ^{a}	40.24 ^{a}	35.69 ^{d}	36.99 ^{c}	36.79 ^{c}	36.80
	Arachidic acid ($C_{20}H_{40}O_2$)	0.26 ^{cd}	0.28 ^{abcd}	0.35 ^{a}	0.33 ^{ab}	0.25 ^{d}	-	0.26 ^{bcd}	0.33 ^{ab}	0.33 ^{abc}	0.27
	Total (%)	36.91	37.57	38.88	40.7	43.26	42.76	38.96	39.45	40.16	39.85
Unsaturated	Oleic acid ($C_{18}H_{34}O_2$)	54.56 ^{g}	59.31 ^{b}	55.88 ^{ef}	57.43 ^{d}	55.43 ^{f}	55.98 ^{e}	59.79 ^{a}	59.23 ^{b}	58.55 ^{c}	57.35
	Linoleic acid ($C_{18}H_{32}O_2$)	7.45 ^{a}	2.07 ^{b}	1.84 ^{c}	1.48 ^{d}	1.30 ^{de}	1.25 ^{ef}	1.06 ^{fg}	0.96 ^{g}	1.04 ^{g}	2.05
	Linolenic acid ($C_{18}H_{30}O_2$)	0.72 ^{b}	0.63 ^{c}	3.15 ^{a}	0.13 ^{d}	-	-	-	-	-	0.52
	Gadoleic acid ($C_{20}H_{38}O_2$)	0.32 ^{a}	-	-	0.26 ^{b}	-	-	-	-	-	0.07
	11- Eicosenoic acid ($C_{20}H_{38}O_2$)	-	0.42 ^{a}	0.24 ^{c}	-	-	-	0.19 ^{c}	0.36 ^{b}	0.24 ^{c}	0.16
	Total (%)	63.09	62.43	61.11	59.3	56.73	57.23	61.04	60.55	59.83	60.15

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unsaturated fatty acids seem to have strong positive correlation with number of seeds per fruit and positive correlation with altitude, seed kernel weight, seed breadth, seed thickness and percent oil yield. The saturated fatty acids were inversely proportional to the unsaturated fatty acids (Fig. 2). Oleic acid (mono-unsaturated, C18), is the maximum appropriate for proper biodiesel with 20 ºC melting point, very low viscosity, which makes it appropriate to be used below cold climatic conditions (Knothe 2008).

CONCLUSIONS

The observed differences in parameters across various sites did not exhibit significant variability or notable distinctions. The average oil yield from the seeds suggests that yield does not significantly differ between the trees, regardless of the location of collection. Eight fatty acids identified, five were unsaturated fatty acids and had the highest composition overall compared to the three saturated fatty acids. The fatty acid composition showed variation between sites and in some sites certain fatty acids were completely absent, to ascertain the reason for this kind of observation more research should be done on the aspect of biosynthesis of this kind fatty acids with respect to climatic and environmental conditions. One of the more significant challenges that was noted during the investigation is seed decortication, this step is very much necessary for proper use of seeds.

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