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Morphological Variation of Fruits and Seeds of *Garcinia indica* (Thouars) Choisy in Western Ghats Region of Karnataka, India

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Abstract**:** *Garcinia indica* is an important indigenous tree, which grows in tropical and evergreen forests of India and has a tremendous potential to be utilised in various value-added applications. The present study aims at understanding the variation in fruit and seed characters among 16 populations in Western ghats region of Karnataka. The large variation exists in economically important characters such as seed weight, fruit weight and rind weight and a significant variation was observed in morphological characters of different populations. The samples collected from Gundya region shown highest fruit and seed traits except in rind weight and rind thickness which was high in the fruit samples collected from Belthangady. A strong dependence of morphological characters on geoclimatic factors was revealed in correlation studies. The cluster analysis carried indicated that these16 population divided into two major groups based on the different morphological characters studied. The results concluded that *G. indica* showed a high level of phenotypic plasticity and these results can be used as a foundation for selective breeding.

Keywords: *Garcinia indica*, Fruit and seed characters, Morphological variation, Principal component analysis (PCA), Cluster analysis

Garcinia indica (Thouars) Choisy (Kokum) is a polygamodioceous tree, belongs to the family Clusiaceae. It has multifarious uses both traditionally and commercially. Kokum has been wildly used by the local communities for therapeutic, culinary and beverage purposes. Commercially, Kokum butter and rinds has been used by the cosmetic, pharmaceutical, nutraceutical and beverage industries (Swami et al 2014). The red coloured antioxidant dye present in rinds of kokum is being used as a natural food colourant. The bioactive compounds present in *G. indica* such as Hydroxy citric acid and Garcinol are known for its anti-obesity and anti-oxidant properties respectively (Ravi et al 2022). It is an under exploited tree, which grows in a dissipate manner in forest land, riverside, roadside, waste land and generally does not require much rainfall for their growth (Swami et al. 2014). This tree can flourish at coastal belts and thrive well upto an elevation of 800 metres above mean sea level (Nayak et al 2010, Braganza et al 2012). The wide variation in morphological characters of a tree species is primarily govern by geo-specific environmental conditions (Ji et al 2016). Studying of variation in phenotypic characters are the markers for the genotypic selection of a tree, which point towards the adaptive evolution (Pigliucci et al 2006). The variation in morphological traits of a same species is controlled by the genetical composition of the tree species and environmental factors or through their interaction. Morphological variation in an ecological zone is controlled or induced by micro climatic factors viz., rainfall, humidity, temperature etc. (Gouwakinnou et al 2011, Atefe et al 2015). It is vital to assess the intra specific variation among populations in a robust manner to determine the effect of geoclimatic and genetic parameters on morphological variation (Franks et al 2013). Recently few studies have been carried out to understand the morphological variation in *Garcinia* species. In *Garcinia* species the morphological variation exists within the species in same ecosystem (Parthasarathy 2014). *Garcinia gummi-gutta* grows highlands, coastal belts, and river banks however, the tree grown near river bank had high productivity when compared to other habitats (Aswathi et al 2018). The existence of tree in wide range of altitude, the dioecious and cross - pollinated nature of *G. indica* harbour a wide genetic diversity in morphological and biochemical characters among different populations (Joseph et al 2015). However, there was no studies on morphological variation on fruit and seed characters in *G. indica* in Western ghat region of Karnataka, Southern India. The present study aims to elucidate the magnitude of morphological variation in economically important fruit and seed characters of *G. indica* Choisy among 16 populations in Karnataka based on the climate and geographical parameters.

MATERIAL AND METHODS

Study area: The study was carried out in Western ghats

region of Karnataka, Southern India, based on occurrence and abundance of tree and area consists of 16 different populations of Western ghats namely, Puttur, Udupi, Mangalore, Bhatkal, Honnavar, Karwar, Belthangady, Madikeri, Dandeli, Dharwad, Yellapur, Londa, Sirsi, Charmady, Kukke and Gundya. The selected site was separated from each other by a distance of at least 50 km to avoid sampling error.

Data collection: The latitude, longitude and altitude of study site were measured using Garmin GPS device. The mean annual rainfall from March 2021-May 2022 data were obtained from https://mausam.imd.gov.in/ (Table 1). The sample selection sites were marked in digital elevation map (DEM) by using QGIS (3.4) software (Fig. 1). Fruits were collected from the selected matured trees having total height and breast diameter of 11-50 m and 15.55-70.7cm respectively were selected. The former was measured using range finder (Forestry proII) and later by a measuring tape. In each site the fruits were collected from 10-15 selected trees and bulked for further study. Physiologically matured, disease and pest free fruits were selected. Fruits of 100 numbers from the bulk were selected randomly for the morphological variation analysis. The morphological data such as fruit weight (FW), total seed weight (SW), rind thickness (RT), rind weight (RW), fruit length (FL), fruit diameter (FD), seed length (SL) and seed diameter (SD) was recorded. The RT, FL, FD, SL, SD were measured using vernier caliper with an accuracy of 0.01mm and FW, SW and RW were using weighing balance with an accuracy of 0.001 g.

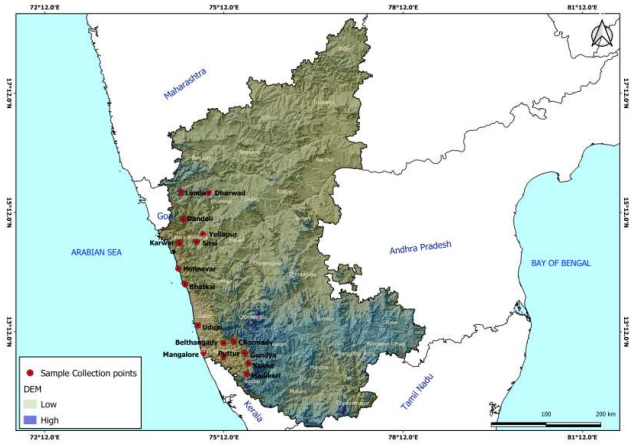

Fig. 1. Location map of population selected in Western ghats, Karnataka

Statistical analysis: The data was analysed using SPSS 20 software (IBM company, New York, USA). Significant difference among means were analysed using Tukey's post hoc test. Karl Pearson's correlation were calculated and analysed among the fruit and seed characters with geoclimatic parameters. The effect of fruit trait with seed traits were also deliberated using Karl Pearson's correlation. Principal component analysis (PCA), a multivariate statistical tool was used for elucidating the relative variation of different fruit and seed traits for estimating the total variability using Originpro 9.0 (Origin Lab company, Northampton, MA, USA) software. In principal component analysis, morphological data sets of different units were normalised with a mean of 0

Table 1. Geographical coordinates (latitude, longitude), altitude and rainfall of study area

Sample site	Latitude	Longitude	Altitude (msl)	Mean annual rainfall (MAR) (mm)
Honnavar	14º15'33.8"	74º26'12.1"	4	4613
Karwar	14º41'0.65"	74º27'21.6"	19	4293
Udupi	13º18'18.8"	74º45'47.0"	13	3609
Bhatkal	13º59'43.3"	74º32'55.2"	26	4569
Mangalore	12º51'11.7"	74º51'24.3"	32	3382
Belthangady	13°00'34.6"	75º11'37.1"	77	4383
Puttur	12º46'53.9"	75º11'34.7"	79	3609
Kukke	12º40'10.29"	75º36'36.2"	133	1122
Charmady	13º01'55.1"	75º21'55.4"	125	4383
Gundya	12º50'40.9"	75º32'52.4"	143	3609
Madikeri	12º29'25.7"	75º34'49.4"	229	3556
Dandeli	15º05'20.2"	74º31'22.1"	448	2064
Yellapur	14º50'0.38"	74º50'56.6"	522	2205
Sirsi	14º42'39.2"	74º44'35.5"	514	3194
Londa	15º32'40.9"	74º28'58.5"	687	2394
Dharwad	15º31'48.3"	74º56'40.0"	702	1060

and variance of 1. Hierarchical cluster analysis was used for constructing dendrogram between the population and was carried out using group linkage method according to squared Euclidean distance in Originpro 9.0 Software.

RESULTS AND DISCUSSION

Fruit and seed characteristics: The minimum, maximum, mean, median and data distribution of the morphological parameters among the population is represented in Figure 2 (a-h). There was significant variation in fruit and seed traits between the different population. The samples collected from population of Gundya shown highest fruit and seed characters except in RT and RW which was highest in population of Belthangady. The mean FW varied from 15.71

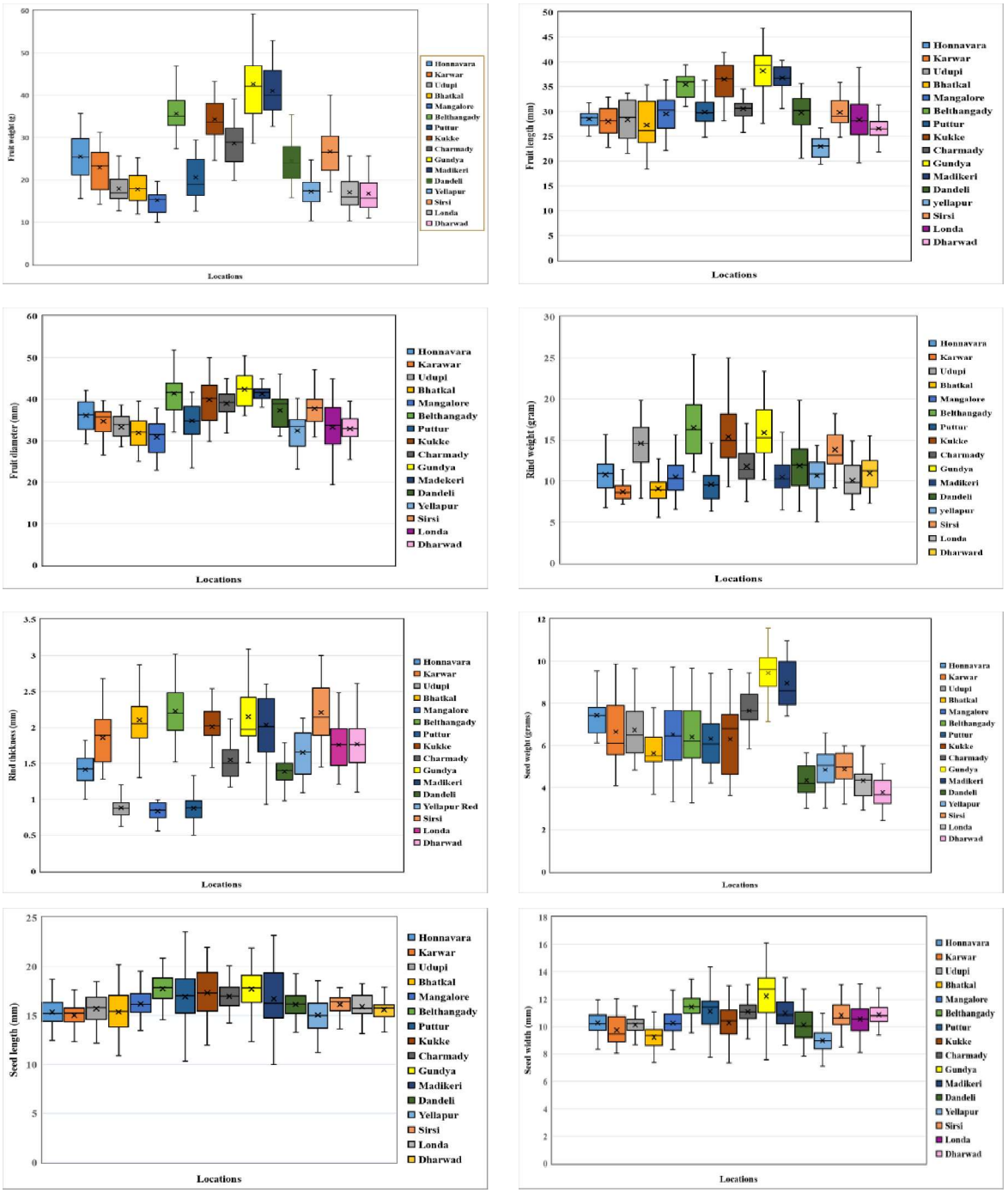

Fig. 2. The variation in morphological characters namely FW (a), FL (b), FD (c), RW (d), RT(e), SW (f), SL (g), SD (h) in *Garcinia indica* among 16 populations

to 41.50 g, with highest fruit weight in samples collected from Gundya region. The fruit length and fruit diameter of *Garcinia indica* was maximum in populations of Gundya followed by Madikeri. The highest fruit length to diameter was also observed in populations of Gundya (0.90) followed by Madikeri. The maximum RW and RT was observed in population of Belthangady with an average of 16.69 g and 2.38 mm respectively. Followed by the population Belthangady, the RW and RT were observed to be the most for the samples collected from Gundya and Kukke region. The highest total seed weight was observed in population of Gundaya (9.30 g) whereas, the least total seed weight was observed in population of Dandeli (3.50 g). The population of Gundya was also observed highest seed length (17.70 mm) and seed diameter (12.24 mm) (Table 2). *G. indica* have vulnerable status according to IUCN (Gowthami et al. 2021) however, the seed and fruits have different industrial uses and are one of the important characters for selection (Baliga et al 2011). The existence of variation in *Garcinia indica* was reported in Western ghat region of Maharashtra, the fruit weight ranges from 25.4 g to 58.38 g, the maximum fruit length and width was 4.28cm to 4.75cm and the fresh fruit rind ranges from m 13.93 g to 35.2 g (Gawankar et al 2001). Similarly, in Goa the variation within the 264 accessions was reported in tree canopy shape, leaf shape, fruit, and seed characters. The fruit characters between 11 taluks shown that the maximum fruit weight was 47.60 g in natural conditions, the fruit rind was 0.48 cm, the fruit length and diameter ranges from 1.19 to 4.36 cm and 1.80 to 5.51 cm in natural conditions (Priya devi et al 2013). Similar findings were also reported from study conducted in *Garcinia gummi-gutta* at Andaman Nicobar Islands by Bohra and Waman (2019), confirming the existence of morphological variation between the population.

Influence of locations and fruit traits: The FW, FL, FD, SL and RW had a positive correlation with longitude, but these are negatively correlated with latitude (Table 3). The significant r^2 were observed for FW, FL, FD, SL and RW with longitude i.e., 0.71, 0.75, 0.70, 0.83 and 0.50 respectively. Mean annual rainfall had a positive relation with fruit weight, seed weight, fruit length and fruit diameter, but a significant correlation was seen only with seed weight (r^2 =0.60). The altitude or elevation does not have any effect on the morphological variation in fruit and seed characters. It is negatively correlated with all the fruit and seed characters expect rind thickness, which was not significant. The correlation among fruit and seed characters were represented in Table 4. The FW is significantly correlated with FL, FD, SL, SD, SW, RW and RT. A significant positive correlation was observed for SW with FW, SL and SD.

Table 2. Relationship between geoclimatic factors and fruit traits

Morphological parameters		Geo-climatic parameters			
		Latitude	Longitude	Altitude	Mean Annual Rainfall (MAR)
Fruit characters	Fruit weight	-0.56	0.71*	-0.37	0.16
	Fruit length	-0.6	0.75*	-0.34	0.07
	Fruit diameter	-0.43	0.70*	-0.19	0.01
	Rind weight	-0.36	0.50*	-0.12	-0.11
	Rind thickness	0.14	0.27	0.24	-0.14
Seed characters	Seed weight	0.38	0.54*	-0.77	0.60*
	Seed length	-0.79	0.83*	-0.50	0.14
	Seed diameter	0.09	0.09	-0.09	-0.01

*Denotes values are significant at 95% confidence interval

Table 3. Relationship between fruit and seed traits

Morphological parameters	Fruit weight	Fruit length	Fruit diameter	Rind weight	Rind thickness	Seed weight	Seed length	Seed diameter
Fruit weight	1							
Fruit length	0.94*	1						
Fruit diameter	0.95*	0.59*	1					
Rind weight	0.57*	0.60*	0.59*	1				
Rind thickness	0.62*	0.54*	0.65*	0.50*	1			
Seed weight	0.54*	-0.23	-0.08	-0.36	-0.03	1		
Seed length	0.72*	0.71*	0.70*	0.32	0.18	0.59*	1	
Seed diameter	0.92*	0.04	-0.06	0.42	0.02	0.58*	0.06	1

*Denotes values are significant at 95% confidence interval

Similarly, RW was significantly correlated with RT, FW, FL and FD. Wang et al (2020) and Ji et al. (2016) reported that the variation in phenotypic character was largely influenced by local environmental characters. The variation caused by climatic variation may led to adaptive evolution and phenotypic plasticity among the species (Aitken et al 2008, Hoffmann and Sgro 2011, Alberto et al 2013, Franks et al 2014). Topographical (latitude, longitude, altitude) or abiotic factors (rainfall, temperature, precipitation) stimulate the variation in morphological traits of tree species between the population (Liu et al 2011, Soper Gorden et al 2016, Jesus et al 2017, Mojzes et al 2018, Wu et al 2018).

Principal component analysis (PCA) and cluster analysis: The PCA is a statistical tool used to elucidate total variation caused by different morphological traits such as seed and fruit characters. The results of the multivariate analysis for principal component revealed that maximum amount of variation was for PC1, PC2 and PC3 constituting total of 88.27%, which was evident in screen plot (Fig. 3). The PC1, PC2 and PC3 causing 64.56, 13.47 and 10.23% respectively accounted for major variation. The PC3 point in screen plot is known as elbow point after which the variation is slight. The component scores of variables contributing to coefficient of PC were depicted in Table. 5. The major morphological traits causing variation in PC1 are FW, FL and FD, whereas, PC2 is contributed by RT and SW contributes to PC3. The biplot represented in interpret the relationship among different fruit and characters and 16 different populations using extracted eigen value of coefficient of PC1 and PC2 (Fig. 4).

The hierarchical dendrogram between different population based on Euclidean distance from Principal component 1 and 2. The dendrogram forms two main clusters between 16 different populations of *G. indica* based on the variation in morphological characters. The cluster I consist of Honnavar, Karwar, Bhatkal, Yellapur, Dandeli, Londa, Dharwad, Udupi, Mangalore, Puttur whereas, the cluster II consist of Belthangady, Kukke, Charmady, Sirsi, Gundya and Madikeri. The cluster II populations had showed superior various morphological traits when compared with cluster I based on the study. From the dendrogram it was evident the

Table 4. Component scores of the first three factors of the total variance

Variables	Component 1	Component 2	Component 3
Fruit weight	0.41	0.03	0.31
Fruit length	0.41	-0.02	-0.06
Fruit diameter	0.40	0.13	0.17
Rind Weight	0.31	0.22	-0.48
Rind thickness	0.22	0.72	0.33
Seed weight	0.25	-0.54	0.55
Seed length	0.38	-0.15	-0.36
Seed diameter	0.34	-0.27	-0.28

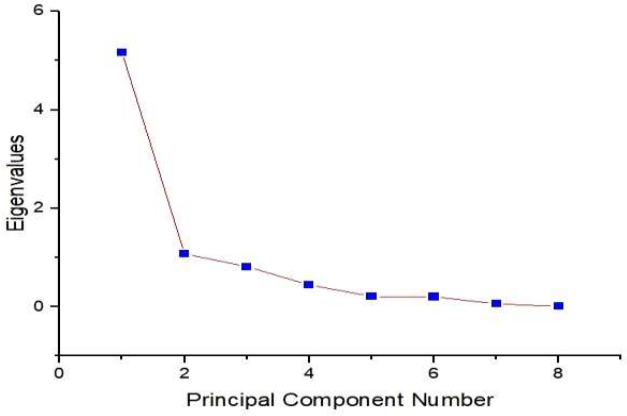

Fig. 3. Screen plot for PCA of different fruit and seed variable

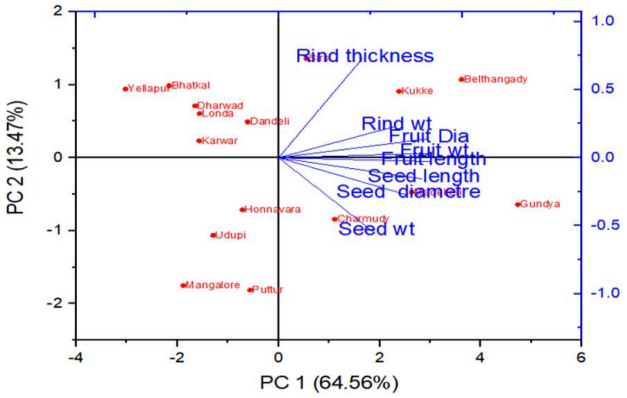

Fig. 4. Two dimensional bi plot of PCA of 16 different populations of *G. indica*

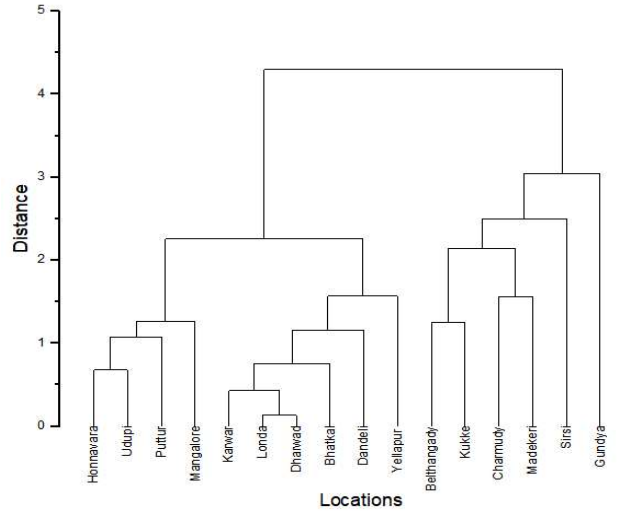

Fig. 5. Hierarchical dendrogram between different populations of *G. indica*

least representative was in population of Gundya whereas the most representative population was Dandeli. Despite, the Gundya population was distinct because FW, FL, FD, SW, SL and SD character was higher than other populations.

CONCLUSIONS

The study was conducted to understand the variation present in morphological characters of *Garcinia indica* in 16 different populations of Karnataka. Based on Karl Pearson correlation study it is evident that the morphological parameters had a strong relationship with longitudinal geoclimatic parameters. The PCA and dendrogram analysis revealed that the population in cluster II have distinguished morphological characters. The study also revealed that samples collected from population of Gundya showed a prominent character in fruit and seed character except rind thickness and rind weight which was highest in the samples collected from Belthangady. Thus, the variations between the population provide us an abundant source for breeding material based on the need of industry.

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