

Biological study with morphological characterization of flower phenology in *Garcinia indica* (Choisy)

Abstract:

Characterization flowering pattern in *Garcinia indica* was studied using different age groups planted in a field gene bank. Flowering commenced in five-year-old trees (5YP) in all age groups targeted to study, followed by four-year-old trees. The first flowering was recorded during 3-4th week of November and continued for ten weeks among the plants. *G. indica* accessions with mixed sexuality were recorded where, the majority of bisexual flowers with female and male flower were there in the same plant. However, flower appearance started in 3rd week of November and extended up to December 3rd week and the maximum number of trees flowered in 1st week of December. Most of the accessions recorded 50 per cent flowering on 2nd and 3rd week of January. The duration of flowering varied from 8 to 10 weeks. Majority of the flowers were observed to be auxiliary and some of them were terminal. Traits such as petal length, sepal length, stamen length and style length did not exhibit significant differences among the accessions studied. However, pollen viability varied among the accessions. Maximum per cent (88.33%) of viable pollen were recorded in 5YP-1 and minimum (82%) in 5YP-3 & 15 with the mean value of 45.68%.

Keywords: Characterization, Flower, *Garcinia indica*, Anthers, phenology

1. Introduction

Garcinia species are generally evergreen trees and shrubs which thrive well in high-rainfall areas of the tropics. Sexually the trees are dioecious and hence, largely are cross-pollinated. Morphological variability exhibited by the trees in forest ecosystems is very high and diversity is obvious for all observable traits. The fruits of *Garcinia* species are known to have anti-obesity properties which is largely owing to the presence of the compound called (-)-Hydroxycitric acid (HCA). This property made *Garcinia* species popular in the international and respective local markets. Other phytochemicals present in the species are isoprenylated xanthenes, a class of secondary metabolites with multiple reports of biological effects, such as antioxidant, pro-apoptotic, anti-proliferative, antinociceptive, anti-inflammatory, neuroprotective, hypoglycemic and anti-obesity (Han *et al.*, 2004, Mathew *et al.*, 2011 and Shivakumar *et al.*, 2013). The most popular species of the genus *Garcinia* is *Garciniamangostana*, which is commonly known as mangosteen and has been named as 'queen of tropical fruits' for its unique pleasant taste and visual appearance of a crown-like structure (Chinavat and Subadrabuddhe, 2004). The seeds and pericarps of the fruit have a long history of use in the traditional medicinal practices of the region, and beverages containing mangosteen pulp and pericarps are sold worldwide as nutritional supplements (Chinavat and Subadrabuddhe, 2004).

Garcinia indica (Choisy.), commonly known as *kokum*, *murugalu* and *punarpuli* is an evergreen tree with a conical crown shape. This species is cultivated on a small scale in coastal regions and hilly areas of the *Western Ghats* of India, which covers the *Konkan* region of Maharashtra, Goa, coastal and hilly areas of Karnataka and similar ecosystems of Kerala state. The *kokum* fruit is spherical in shape, purplish orange to pinkish-red in colour, fleshy and has an acidic flavour. The *kokum* has mainly two different types - red and yellow, particularly spread over the *Western Ghat* central region in Uttara Kannada district. Red *kokum* is the common type whereas, yellow *kokum* is a unique type of *kokum*, it is normally called '*bilimurughi*' (white *kokum* by native people) (Hegde, 2019). The *kokum* fruit has been used for culinary and medicinal purposes since the age-old days. The rind of the *kokum* fruit is used to make fresh juice which is a natural coolant (Nayak *et al.*, 2010).

The dried rind is used for cooking, as a substitute for tamarind and as a food preservative. The fatty substance present around the seed of the *kokum* is called *kokum* butter, and is extensively used in the cosmetic and pharmaceutical industries. The *kokum* seed contains a fair amount of oil which has its use in pharmaceutical and cosmetic industries. Some of the pharmaceutical activities of *kokum* fruit are anti-bacterial, anti-fungal, anti-ageing, neuroprotective, anti-ulcer, hyperglycemic, anti-obesity, anti-inflammatory and anti-oxidant activities (Milind *et al.*, 2013). This species is being cultivated in non-forest regions owing to its importance in the food industry. However, not much information is available with regards to its flowering behavior including years after planting to flowering, variability in flower related traits etc. The present study aimed at unraveling some of these aspects of *Garcinia indica*.

2. Material and Method

The present study was carried out at the Department of Biotechnology and Crop Improvement, College of Horticulture, Bengaluru, UHS Campus G.K.V.K. Post, Karnataka state. Early growth dynamics of plants play a crucial role in their survival and establishment under field conditions, as part of field gene bank.

2.1. Experimental procedures

Flower phenology, reproductive traits and yield-related observations were studied in *Garcinia indica* as age groups were available in the field gene bank. The relative plasticity for these traits was attempted to gain an insight into the kind of genetic diversity prevailing among the plants present in *ex situ* field gene bank. A set of 10 traits pertaining to flower and fruit yield were recorded during the years 2019-20 and 2020-21. The observations were recorded according to the Descriptor of *Garcinia* species by Bioversity International, Roam, Italy. The following observations were recorded.

2.2 Flower phenology traits

First flower bud appearance: The time required for flower bud appearance since initiation was observed and recorded as the particular week of the month.

Sexuality of the flower: In each selected plant 20 flowers were randomly selected on different branches of a tree to know the sexuality of the flower and expressed as male, female or bisexual.

Time of 50% flowering: The time from flower initiation up to the end of flowering was recorded weekly and expressed as 50 per cent flowering.

The span of flowering: The observation of flowering was recorded from the time of initiation of the flower up to the last flowering time the observation was recorded at the particular week of the month

Flowering habit: In each selected plant 20 flowers were randomly selected on different branches to know the flowering habit and expressed as a flowering habit.

2.3 Scanning Electron Microscope analysis

Male, female and bisexual flowers of *Garcinia indica* were studied for the ultra-fine structure of anther and ovary. The facilities at the Centre for Nanotechnology, University of Agricultural Science, Raichur were utilized. The flowers were cross-cut into two-half for anther and ovary size and anther surface and the sample was dried in the vacuum chamber of SEM for 15 min to remove the moisture from the flower. Later, the SEM images were recorded at 3,300x to 20,000x. The pollen image was scanned from the top of the flower and the stigma and ovary were scanned from the top.

3. Result

In India, the genus *Garcinia* has a rich diversity with over 35 species spread across *Western* and *Eastern Ghats*. *Garcinia indica* locally known as *kokum*, are economically important to the local farmers as non-timber forest products (NTFP) both in the *eastern* and *western ghats* of India. In some regions of the *Western Ghats* of Maharashtra, Karnataka, Goa and Kerala farmers do take up systematic cultivation of these species as a source of additional income (Swami *et al.*, 2014). Characterization of *Garcinia indica* accession for growth and related traits revealed high level of variability.

Time of initiation of flower

In general, most of the trees started flowering during 3rd week of December (Table1). Flower appearance started in 2nd week of November and extended up to December 3rd week and a maximum number of trees flowered in 1st week of December in the initial year of the study. Three plants started flowering during 3rd and 4th week of November, two plants started flowering

during the 2nd week of December and one tree started in December during 2019-20. During the second year (2020-21) most of the trees started flowering in 3rd and 4th week of November. Flower appearance started in 1st week of November and extended up to 4th week of November; one plant started flowering during 2nd week of November and 4 plants started flowering during 2nd week of December. Similarly, one plant started flowering on 1st and another plant started flowering on 3rd week of December (Table1).

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Table1: Behavior of *G. indica* individual plant for flower-related traits

Sl. No.	Plant Number	Age (Year)	Sexuality of the flower	Time of initiation of flower	Time of 50 % flowering	Span of flowering (Week)	Flowering habit
1	5YP-1	5	B	4 th Week Nov.	1 st Week Jan.	10	Auxiliary
2	5YP-2	5	F	3 rd Week Nov.	4 th Week Dec.	9	Auxiliary
3	5YP-3	5	F	3 rd Week Nov.	4 th Week Dec.	10	Auxiliary
4	5YP-4	5	F	1 st Week Dec.	2 nd Week Jan.	9	Auxiliary
5	5YP-5	5	B	3 rd Week Dec.	3 rd Week Jan.	10	Terminal
6	5YP-6	5	B	3 rd Week Dec.	3 rd Week Jan.	10	Auxiliary
7	5YP-7	5	F	4 th Week Nov.	4 th Week Dec.	9	Auxiliary
8	5YP-8	5	F	2 nd Week Dec.	2 nd Week Jan.	8	Auxiliary
9	5YP-9	5	B	4 th Week Nov.	2 nd Week Jan.	9	Auxiliary
10	5YP-10	5	F	2 nd Week Dec.	2 nd Week Jan.	10	Auxiliary
11	5YP-11	5	B	2 nd Week Nov.	2 nd Week Jan.	9	Terminal
12	5YP-12	5	F	4 th Week Dec.	4 th Week Jan.	10	Terminal
13	5YP-13	5	B	3 rd Week Dec.	3 rd Week Jan.	10	Auxiliary
14	5YP-14	5	F	3 rd Week Dec.	3 rd Week Jan.	9	Auxiliary
15	5YP-15	5	B	3 rd Week Nov.	1 st Week Jan.	9	Terminal

5 YP = five year old plan

Table2: Behavior of *G. indica* Choisy individual plant for flower related traits

Sl. No.	Plant Number	Age (Year)	Sexuality of the flower	Time of initiation of flower	Time of 50 % flowering	Span of flowering (Week)	Flowering habit
1	5YP-1	6	B	3 rd Week Nov.	4 th Week Dec.	9	Auxiliary
2	5YP-2	6	F	4 th Week Nov.	4 th Week Dec.	10	Auxiliary
3	5YP-3	6	F	4 th Week Nov.	2 nd Week Jan.	11	Auxiliary
4	5YP-4	6	F	2 nd Week Dec.	4 th Week Jan.	10	Auxiliary
5	5YP-5	6	B	3 rd Week Nov.	1 st Week Jan.	10	Terminal
6	5YP-6	6	B	3 rd Week Nov.	4 th Week Dec.	9	Auxiliary
7	5YP-7	6	F	2 nd Week Dec.	3 rd Week Jan.	9	Auxiliary
8	5YP-8	6	F	4 th Week Nov.	4 th Week Dec.	8	Auxiliary
9	5YP-9	6	B	2 nd Week Dec.	3 rd Week Jan.	9	Auxiliary
10	5YP-10	6	F	1 st Week Nov.	4 th Week Dec.	9	Auxiliary
11	5YP-11	6	B	2 nd Week Nov.	4 th Week Dec.	10	Terminal
12	5YP-12	6	F	3 rd Week Dec.	3 rd Week Jan.	10	Terminal
13	5YP-13	6	B	3 rd Week Nov.	1 st Week Jan.	11	Auxiliary
14	5YP-14	6	F	4 th Week Nov.	4 th Week Dec.	9	Auxiliary
15	5YP-15	6	B	2 nd Week Dec.	3 rd Week Jan.	9	Terminal
16	4YP-1	5	M	4 th Week Nov.	1 st Week Jan.	9	Terminal
17	4YP-2	5	F	3 rd Week Nov.	4 th Week Dec.	8	Auxiliary
18	4YP-3	5	B	1 st Week Dec.	2 nd Week Jan.	8	Auxiliary
19	4YP-4	5	M	3 rd Week Nov.	4 th Week Dec.	8	Auxiliary
20	4YP-5	5	F	4 th Week Nov.	4 th Week Dec.	9	Auxiliary

B=Bisexual Flower; **M**= Male flower; **F**= Female Flower

Time of 50 per cent of flowering

Most of the trees recorded 50 per cent flowering on 2nd and 3rd week of January (Table1) and they took 4-5 weeks from initiation of 1st flowering in the initial year of the study. Three plants recorded 50 per cent flowering during 3rd week of December and remaining two plants were observed to be 50 per cent flowering during 1st week of January of 2019-20. During second year of study, 2020-21, most of the trees recorded 50 per cent flowering on 4th week of December and they took 4-5 weeks from initiation of 1st flowering. Four plants recorded 50 per cent flowering on 3rd week of January and 3 plants observed 50 per cent flowering during 1st week of January and only one plant given 50 per cent flowering during 1st week of January (Table2). The duration of flowering was recorded from 8 to 10 weeks during initial year of the study the maximum flowering habit was observed to be auxiliary and some of them were Terminal. And the selected plants for the study were mostly bisexual and female during the initial year of the study

Petal length (mm)

The data obtained on petal length measurement showed that maximum petal length (4.58mm) was observed in 5YP during 2019-20, while during 2020-21, the maximum petal length (5.29mm) was ded in 5YP and followed by 4YP (4.86mm) (Table3). The data five-year-old old plants was individually analyzed during 2019-20, maximum petal length (5.0mm) was recorded in 5YP-4th, 7th, 11th and 13th plants and minimum petal length (3.60 mm) was recorded in 5YP-8th plant with the mean petal length of 4.59 mm. During 2020-21, the maximum petal length (6.03mm) was recorded in 5YP-1st and 10th plants and minimum petal length (4.50mm) was recorded in 4YP-1st plant with the mean value of 5.28mm (Table5-6).

Sepal length (mm)

The length of sepal was recorded in five years old flowering plant, the maximum length of sepal was (5.07 mm) during 2019-20. While, during 2020-21, the maximum sepal length (4.77mm) was recorded by 5YP and 4.4 mm was recorded by 4YP (Table3). The data was recorded for individual plant, the maximum sepal length (5.90 mm) was recorded by 5YP-1 and minimum sepal length (4.56 mm) by 5YP-14 with the mean value of 5.08 mm during 2019-20.

During 2020-21, the maximum sepal length (6.01 mm.) was recorded by 5YP-14 and minimum sepal length (4.00 mm) was recorded by 4YP-11 with the mean value of 4.72 mm (Table5-6).

Stamen length (mm)

The data of stamen length was recorded in five year old flowering plants. The maximum length of stamen was (4.52mm) by 5YP during 2019-20. While during 2020-21, the maximum stamen length (4.44mm) was recorded by 5YP and 4.2 mm by 4YP. (Table3). The data in five year old plant was analyzed for individual plant during 2019-20. The maximum stamen length (5.06 mm) was recorded by 5YP-1 and minimum (4.20 mm) by 5YP-3 with the mean value of 2.41 mm. During 2020-21, the maximum stamen length (5.20mm) was recorded by 5YP-10 and minimum (4.04 mm) by 5YP-1 with the mean value of 1.55 mm (Table5-6 and plate 1).

Style length (mm)

The style length was measured in five-year-old flowering plant. The maximum style length (4.17 mm) was recorded by 5YP during 2019-20. During 2020-21, the maximum style length was (4.47 mm) by 5YP and (4.06 mm) by 4YP (Table4). The data in five-year-old plants was individually analyzed during 2019-20. The maximum style length (4.72mm) were recorded in 5YP-13 and minimum stamen length (4.02mm) was recorded in 5YP-7 with the mean value of 3.10 mm. During 2020-21 the maximum style length (5.06 mm) was recorded in 5YP-5 and minimum length (4.01 mm) of style was recorded by 5YP-11 with the mean value of 4.18 mm (Table5-6).

Pollen viability (per cent)

The pollen viability analysis was done for flowering tree. The maximum number of viable pollen (84.62 %) was recorded in 5YP during 2019-20. During 2020-21 the maximum per cent of viable pollen (81.72 %) was preceded by 5YP and (83 %) in 4YP (Table4). The data in five-year-old was analyzed for individual plant during 2019-20 the maximum per cent (88.33 %) of viable pollen were recorded in 5YP-1 and minimum (82 %) in 5YP-3& 15 with the mean value of 45.68 %. During 2020-21 the maximum pollen viability (85 %.) was recorded by 5YP-1 and 4YP-3 and minimum pollen viability (75 %) was recorded in 5YP-13 with the mean value of 28.40 % (Table5-6). The stigma was receptive after the opening of the flower. The flower opens in the mornfromring 6:00 AM to 8:00 AM.

Scanning Electron Microscope analysis

The pollen grain of *Garcinia indica* was examined under Scanning electron microscope. The results revealed the surface texture with protuberances of different shapes and sizes, including vertical and horizontal protuberances, often curved, some blunt-ended, some with grooves and some without grooves (Plate.2).

4. Discussion

The flower characteristics of trees of different age groups in both accessions were studied. In *Garcinia indica*, flowering commenced in five-year-old trees (5YP) followed by four-year-old trees. The flowers began appearing in the 1st week of November and blooming extended up to 4th week of November which agrees with the findings of Tripathi (2021) in this species. It was noted that the maximum number of trees flowered during 3rd week of December followed by 3rd and 4th week of November (Table1). *G. indica* is known to have different sex forms of flowers in the population. In the present study male, female and bisexual flowers were observed in different accessions. The phenomenon of different sex forms of flowers in *G. indica* is known and well-recorded by Thatte and Deodhar (2012). described *Kokum* trees with a wide variety of flower patterns, such as separate trees for male and female flowers, as well as trees with bisexual flowers or same tree producing male and female flowers, trees with bisexual flowers along with male and female flowers. George *et al.* (1992) reported the androdieocious character of this plant.

The flowers were mostly axillary, but some were terminal across accessions studied. This well corroborates with the reports of Shameer *et al.* (2016) in *G. indica*. report by Nayar *et al.* (2014) discerned that the genus *Garcinia* has small clusters of flowers that are produced in cymes or fascicles. Further, the flowers can also be sessile, solitary or in few clusters, terminal or axillary and diversely colored. The flowers are also found with numerous stamens and four to five petals.

Table3: Petal length, Sepal length and Stamen length in *G. indica* accessions

Treatment	Petal length (mm)		Sepal length (mm)		Stamen Length (mm)	
	2019-20	2020-21	2019-20	2020-21	2019-20	2020-21
2-YP	0 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)
3-YP	0 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)
4-YP	0 (1.00)	4.86 (2.42)	0 (1.00)	4.4 (2,32)	0 (1.00)	4.2 (1.51)
5-YP	4.58 (2.36)	5.29 (2.50)	5.07 (2.46)	4.77 (2.40)	4.52 (2.27)	4.44 (2.33)
S.Em.±	0.01	0.01	0.009	0.02	0.03	0.15
C.D. (5%)	0.04	0.05	0.2	0.06	0.10	0.49

2YP= Two-year plant

3YP= Three-year plant

4YP= Four-year plant

5YP= Five-year plant

Table4: Style length, pollen viability and stigma receptivity in *G. indica* accessions

Treatment	Style length (mm)		Pollen viability (%)		Stigma receptivity	
	2019-20	2020-21	2019-20	2020-21	2019-20	2020-21
2-YP	0 (1.00)	1.00 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)
3-YP	0 (1.00)	1.00 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)	0 (1.00)
4-YP	0 (1.00)	4.06 (2.24)	0 (1.00)	83 (9.16)	0 (1.00)	2 (1.73)
5-YP	4.17 (2.25)	4.47 (2.26)	84.62 (9.25)	81.72 (9.09)	2.19 (1.78)	2.26 (1.80)
S.Em.±	0.004	0.03	0.02	0.05	0.01	0.01
C.D. (5%)	0.01	0.12	0.06	0.16	0.03	0.05

2YP= Two-year plant

3 YP= Three-year plant

4YP= Four-year plant

5YP= Five-year plant

Table5: Behavior of *G. indica* individual for flower-related traits

Sl. No.	Plant Number	Age (Years)	PL (mm)	SL (mm)	Sta L (mm)	Sty L (mm)	PV (%)	SR (DAY)	AT
1	5YP-1	5	4.11	5.90	5.06	-	88.33	-	6: 30 AM
2	5YP-2	5	4.50	5.00	-	4.24	-	2	7:15 AM
3	5YP-3	5	4.06	4.80	4.20	4.08	82.00	2	6:00 AM
4	5YP-4	5	5.00	4.70	4.46	4.04	87.00	2	6:10 AM
5	5YP-5	5	4.90	5.00	-	4.44	-	2	7:30 AM
6	5YP-6	5	4.60	4.90	-	4.27	-	2	5:50 AM
7	5YP-7	5	5.00	5.23	4.63	4.02	86.00	2	5:30 AM
8	5YP-8	5	3.60	5.60	4.50	-	88.00	-	5:45 AM
9	5YP-9	5	4.30	5.23	4.04	-	84.00	-	6:00 AM
10	5YP-10	5	4.70	5.06	-	4.20	-	2	6:45 AM
11	5YP-11	5	5.00	5.21	-	4.39	-	2	5:45 AM
12	5YP-12	5	4.90	4.80	4.43	4.13	88.00	2	6:45 AM
13	5YP-13	5	5.00	5.10	-	4.72	-	2	6:30 AM
14	5YP-14	5	4.70	4.56	-	4.06	-	2	5:50 AM
15	5YP-15	5	4.60	5.23	4.83	-	82.00	-	7:30 AM
Mean			4.59	5.08	2.41	3.10	45.68		
S.Em.±			0.18	0.23	0.13	0.19	2.36		
C.D. (5%)			0.52	0.68	0.38	0.57	6.83		

PL: Petal length; **SL:** Sepal length; **Sta. L:** Stamen length; **Sty.L:** Style length; **PV:** Pollen viability

Table6: Behaviour of *G. indica* individual for flower related traits

Sl. No.	Plant Number	Age (Years)	PL (mm)	SL (mm)	Sta L (mm)	Sty L (mm)	PV (%)	SR (DAY)	AT
1	5YP-1	6	6.03	5.01	4.04	4.04	85	2	7: 30 AM
2	5YP-2	6	5.00	5.00	-	4.93	-	2	6:45 AM
3	5YP-3	6	6.00	4.93	-	4.73	-	2	8:00 AM
4	5YP-4	6	6.00	5.01	-	4.06	-	3	7:10 AM
5	5YP-5	6	5.03	5.06	-	5.06	-	2	7:30 AM
6	5YP-6	6	5.06	5.06	4.34	4.70	80	2	5:50 AM
7	5YP-7	6	5.05	4.06	-	3.83	-	3	5:30 AM
8	5YP-8	6	5.29	5.01	4.53	4.13	81	3	5:45 AM
9	5YP-9	6	6.03	4.10	-	4.03	-	2	7:30 AM
10	5YP-10	6	5.13	5.05	5.20	-	79	-	6:45 AM
11	5YP-11	6	5.04	4.00	-	4.01	-	2	7:45 AM
12	5YP-12	6	5.52	4.96	-	5.00	-	2	7:15 AM
13	5YP-13	6	5.06	4.90	4.53	4.83	75	2	7:30 AM
14	5YP-14	6	5.05	6.01	-	4.90	-	2	6:50 AM
15	5YP-15	6	5.03	4.06	-	4.80	-	3	7:50 AM
16	4YP-1	5	4.50	4.03	-	3.94	-	2	5:50 AM
17	4YP-2	5	5.00	4.63	-	4.16	-	2	6:30 AM
18	4YP-3	5	4.93	4.53	4.20	4.10	85	2	6:15 AM
19	4YP-4	5	5.00	4.63	-	4.20	-	2	7:30 AM
20	4YP-5	5	4.90	4.30	4.23	4.13	83	2	6:30 AM
Mean			5.28	4.72	1.55	4.18	28.40		
S.Em.±			0.37	0.29	0.16	0.19	1.82		
C.D. (5%)			-	0.85	0.47	0.56	5.23		

PL: Petal length; **SL:** Sepal length; **Sta. L:** Stamen length; **Sty.L:** Style length; **PV:** Pollen viability

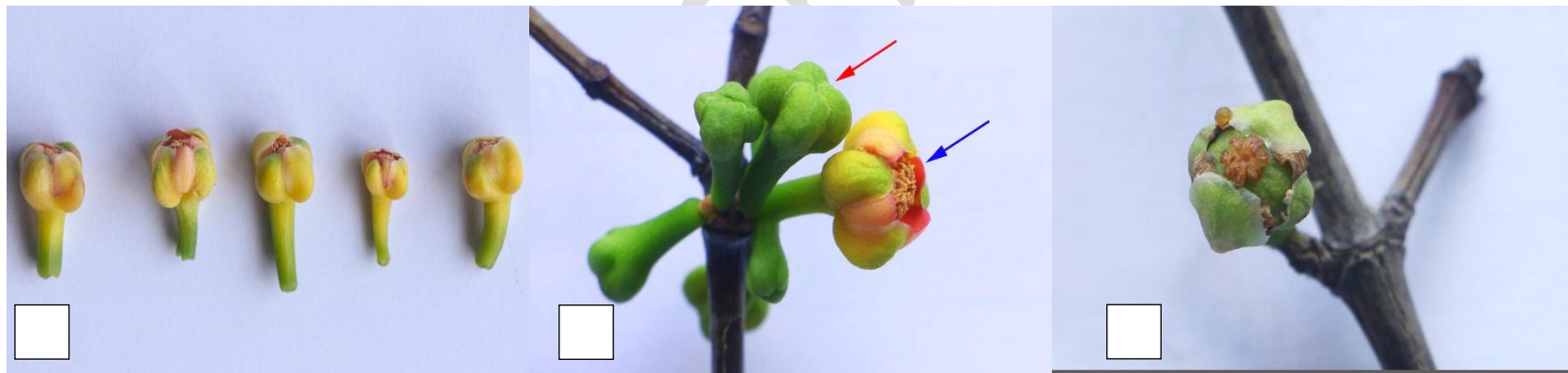


Plate 1: Morphology of different flower types and parts in *G. indica*.

A: Petal & Sepal; **B:** Petal; **C:** Sepal; **D:** Male flower; **E:** Bisexual flower; **E:** Male flower; **F:** Female flower

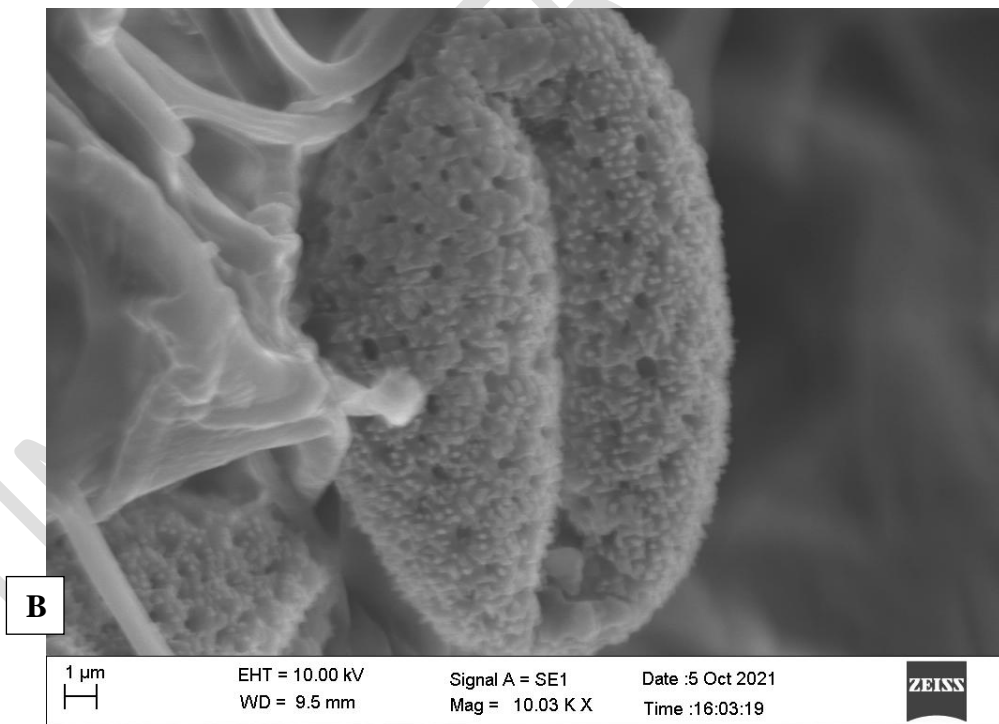
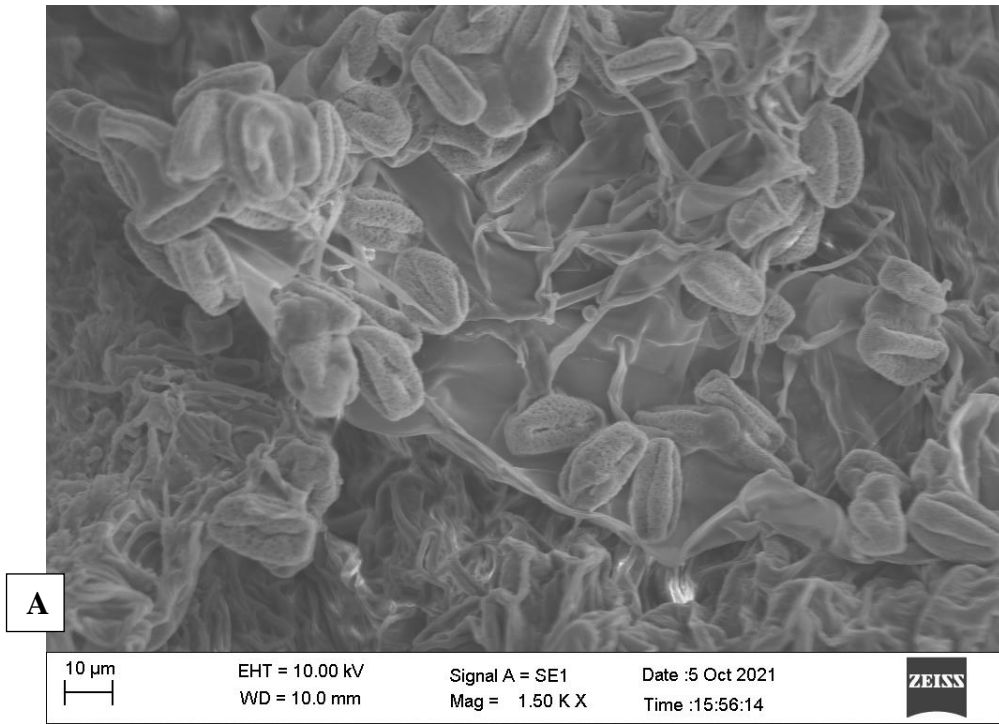


Plate 2 SEM image of *G. indica* anther

A: Anther surface **B:** Pollen shape

The flowering is highly influenced by the climatic and edaphic factors of a location, as it appears from the data. Factors such as temperature, rainfall, nutrient and water availability in the soil besides multiple other factors seem to have influence in flowering behaviour of *Garcinia indica* in western ghats of India (Palkar *et al.*, 2020). During the period of flower initiation, the temperature in the location was 28.40 to 26.00 °C. An elevated temperature shortens the growth period of flower clusters, flowering durations, and lifespans of individual flowers, further, it also decreased the number of hermaphrodite and male flowers (Pramanik *et al.*, 2018). A warm temperature, however, increased the rates and percentage of anther dehiscence and fertilization (Shu, 1999). During the present study, temperatures were low during the flowering period, which led to increased flowering duration, lifespan, and growth rate of flower clusters. In addition to those lower temperatures also increased the number of hermaphrodite and male flowers. In contrast to the present study, Sawant *et al.*, (1997) reported the flower bud appearance from September 29 to 8th October in different genotypes of *kokum*. This variation in the flowering season might be due to the different sets of genotypes studied by each worker and also as a result of the experimental location of the study conducted. Similar variations were reported by Abraham *et al.* (2002); Patil (2005); Himabindu *et al.*, (2010) and Devi *et al.*, (2012) in *kokum*. Gogoi *et al.* (2015) documented the total span of flowering to be ranged from 42-52 days in case of *Garcinia pedunculata* and in *Garcinia cowa* duration of flowering was from 44-50 days. Whereas, in *Garcinia lanceaefolia* and *Garcinia xanthochymus* the total span of flowering ranged from 38-45 days and 45-56 days respectively. Mansyah (2002) also reported a similar result in *Garcinia mangostana*.

The results for pollen viability showed that the highest percentage of viable pollen (84.62 %) was recorded in the 5-year period 2019-20. Sutthinon *et al.* (2018) demonstrated the pollen viability in *G. celebica* through 2,3,5-triphenyl tetrazolium chloride (TTC) assays and found that the viability was 68 per cent. During 2020-21, the maximum per cent of viable pollen (83%) was recorded in the 4-YP (Table 6). Rajkumar *et al.* (2017) also reported maximum pollen viability (89.16 %) when treated with 0.2 per cent TTC. High pollen viability has also been reported in several *Garcinia* male flowers such as *G. cowa* (96-100 %), *G. speciosa* (93-100 %), and *G. schomburgkiana* (95-100 %) (Te-chato, 2007). Meanwhile, *G. atroviridis* had a very low rate

with approximately 3-5% viable pollen, and no viable pollen was found in *G. dulcis* (Te-chato, 2007) and *G. mangostana* (Sutthinonet *et al.* 2013). According to the findings of the harvested pollen grains of many *Garcinia* species have high viability as assessed by their stainability like 99.4% of *Garcinia corymbosa*, 92.5 per cent of *Garcinia forbesii*, and 85 per cent of *Garcinia cf. forbesii* (Ha *et al.*,1988).

Five-year-old plant data were analyzed for individual plants in 2019-20. The highest percentage of pollen viability was found in 5YP-1 (88.33%) and the lowest in 5YP-3 and 15 (82%) with a mean of 45.68 per cent. During 2020-21 the maximum pollen viability was recorded in 5YP-1 and 4YP-3 (85 %) and minimum pollen viability was recorded in 5YP-13 (75%) with the mean value of 28.40 per cent (Table6). The pollen viability was $89.19 \pm 2.11\%$ as tested by 2% TTC. The *in vitro* pollen germination was 72.73 ± 9.20 per cent with 67.13 ± 21.33 per cent long pollen tubes in 20 per cent sucrose solution. This is consistent with earlier studies demonstrating that sucrose is an important component for *in vitro* pollen germination (Honshoet *et al.*, 2007). This optimal sucrose concentration is due to the essential role sucrose plays in nutrient provision and environmental osmotic maintenance (Baloch *et al.*, 2001). It has been suggested that bicellular pollen requires a low level of sucrose (10-15%) (Shivanna, 2003). In *G. celebica*, the most effective conditions for pollen germination were observed on treatment with 20% sucrose for 10-12 h (Sutthinonet *et al.*, 2018). The stigma was receptive two days after anthesis and the flower opened between 6:00 AM and 8:00 AM (Dike *et al.*, 2020). The stigma became receptive only after the opening of flowers. Aswathi *et al.* (2018) observed that in *G. cambogia* maximum stigma receptivity was found at 7.00 PM. On the day of anthesis high percentage of pollen grains were fertile. Stigma is the wet type and broad umbrella-shaped. Karnik and Gunjate (1984) reported that the maximum stigma receptivity (80%) was observed on the day of anthesis followed by 68, 64 and 44 per cent, 1, 2 and 3 days after anthesis respectively. Rajkumar *et al.* (2017) reported that in *G. imberta*, receptivity of stigma gradually decreased with age and within one or two days as the stigmatic lobes became brown and completely dry, indicating a complete loss of receptivity.

When flower visitors contact with stigmas and anthers, the stigmatic exudates can support them to attach to the body of the biotic pollinator. This corresponds with the prevailing notion that

most *Garcinia* species are pollinated by biotic pollinators such as social bees, diverse insects and *Apis* spp. (Richards, 1997; Momose *et al.*, 1998).

4. Conclusions:

The flowering and fruiting were recorded only in five-year-old plants of *G. indica*, the first flowering was recorded during 4th week of November and continued for ten weeks among the plants. *G. indica* accessions with mixed sexuality were recorded where, the majority of bisexual flowers with female and male flower were there in the same plant. The analysis of variance revealed highly significant differences for petal length, sepal length, stamen length, style length, pollen viability and stigma receptivity during 2019-20 and 2020-21. In general, most of the trees started flowering during 3rd week of December. However, flower appearance started in 2nd week of November and extended up to December 3rd week and the maximum number of trees flowered in 1st week of December. Most of the accessions recorded 50 per cent flowering on 2nd and 3rd week of January. The duration of flowering varied from 8 to 10 weeks. Majority of the flowers were observed to be auxiliary and some of them were terminal.

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