



Leaf morphology, floral biology, pollination behaviour of elite bael accessions under rainfed semi-arid conditions

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(Received: 27.01.2020; Accepted: 05.02.2020)

Abstract

The knowledge of floral biology, pollination behaviour and reproduction biology is important for improvement and also to enhance productivity. A study was conducted on nine elite bael germplasm which were collected from Rajasthan, Uttar Pradesh and Gujarat, established through in-situ patch budding at Experimental farm of CHES, Godhra. There was variability in leaf characters i.e., leaflet shape, leaf margin, leaf apex, leaf base and leaf surface among the accessions. Variation in bark colour and bark splitting pattern was also observed. Phyllotaxy in different genotypes was observed tristichous and pentastichous. Inflorescence consists of 2-31 lower buds and also single flowers. Flowers possess 4-5 petals within same accession, but 4 followed by 5 petals were found to be common in almost all studied accessions; however 3 and 6 sepals may be seen rarely in few accession. Considerable variations in size of bud, flower, petal, pedicel, stamen, filament, anther, pistil, ovary style, stigma and number of stamens/flower and pollen viability were recorded among the studied accessions. The maximum anthesis was observed between 6.31-8.30 am in all the varieties and it was continued up to 12.30 pm.

Key words: Floral biology, leaf morphology, pollination behaviour, bael

Introduction

Bael is an important sub tropical fruit tree indigenous to India. It belongs to family Rutaceae (Stone, 1985) which is widely distributed in India. It can also be cultivated in arid and semi-arid region successfully (Saroj *et al.*, 2006; Singh *et al.*, 2019). It has great mythological and religious significance in Indian history and culture, and is one of the underutilized fruits known from pre-historic times finding its mention in Hindu mythology. All the parts of tree, whether stem, root, bark, leaves or fruits at different maturity stages have been traditionally identified to contain medicinal properties which have been described in ancient treatise such as Charak Samhita, Sushrut Samhita, Upvan vinod, and Jain and Budhisht literature (Singh *et al.*, 2019). The ripe fruits are laxative and unripe fruits are prescribed for diarrhea and dysentery, and have a great demand in Ayurvedic system of medicine. In India, it is found in wild form in sub-Himalayan tract and in dry deciduous forest of central and southern Indian region (Singh *et al.*, 2018a and 2019). It is very hardy and has capacity to adapt successfully to a wide range of habitat (Singh *et al.*, 2016a). Bael, owing to its environmental friendly nature, is being placed among plant species group called 'climate purifiers' as it emits a greater percentage of oxygen as compared to other plants (Anurag *et al.*, 2014). A large number of land races are available in the different diversity regions in India (Saroj *et al.*, 2006). A wide range of diversity of bael has been noticed in dry sub-tropical, tropical, arid and semi-arid regions of the country (Singh *et al.*, 2019 a). The plains of U.P., and Gujarat and Rajasthan also have wide distribution of bael genotypes with great diversity particularly in dry climate,

undulating terrains, forest and wastelands (Singh *et al.*, 2019). The genotypes of bael found in different parts of the country have enormous variability with regard to their morphological characters (Singh *et al.*, 1989). Some superior genotypes identified by local people are on the verge of extinction owing to extensive urbanization, and hence, there is an urgent need to conserve them for the use of posterity (Singh *et al.*, 2018). Keeping these points in view, large number of germplasm were established *ex-situ* (199) out of which, 9 genotypes were utilized for present study.

Materials and Methods

Nine accessions established through *in-situ* patch budding at Experimental Farm of CHES, Godhra were utilized for study. All trees were established in Randomized Block Design which was replicated thrice. Accessions used in the experiment were CHESB-1 (IC.0629381), CHESB-5 (IC.0629382), CHESB-8 (IC.0629383), CHESB-11 (IC.0629384), CHESB-16 (IC.0629385), CHESB-21 (IC.0629386), CHESB-27 (IC.0629387), CHESB-29 (IC.0629388) and CHESB-31 (IC.0629389). The different morphological and qualitative growth characters in leaf (base, apex and margin), bark (colour, splitting and thickness) and spine were observed visually in the field, as illustrated by Wilde *et al.* (1972) and Simpson (2006). Studies were conducted during flowering seasons; April-May in the year 2016 and 2017. Basic floral biological data were collected from nine accessions. Flowering period, flower opening time days taken to flower opening, stigma receptivity anther dehiscence and opening pattern were also studied. Sixty

flowers from each accession were subjected to study and data collection was continued in consecutive seasons of each year. Data were recorded in two hour interval starting from 4.30 am to 12 noon to study anthesis and anthesis dehiscence. Pollen viability percentage was studied using 15 of acetocarmin stain test. Anthesis and dehiscence was studied in Goma Yashi, Thar Divya and Thel Nelkanth varieties of bael only. Statistical analysis was followed as per methods outlined by Gomez and Gomez (1984).

Results and Discussion

Flowering period in bael was observed from March to August. It depends upon the variety, soil moisture regime and climatic conditions of particular locality. Under dryland conditions, some of the germplasm flower at their routine period but few the flowers may appear after rain, however peak period of flowering was observed in almost all germplasm after rain, if rainfall is less than 400 mm in previous season resulting into late fruit setting with reduced fruit size.

Leaf morphology

There was variability in the morphological vegetative growth characteristics among the different bael

accessions. The data pertaining to morphological attributes of bael and its leaves showed significant differences and high degree of variability for all the characters were observed (Table 1). The growth habit included tree with upright, spreading, semi spreading and drooping type and foliage with compact, dense and sparse type among all the characterized accessions (Singh *et al.*, 2011). Tree shapes of different genotypes are dome, irregular, semi circular, broad vase and elliptical types among all the genotypes (Singh *et al.*, 2014, 2018a). There were variability in the several leaf characters *i.e.* leaflet shape (ovate, broadly lanceolate to ovate, broadly ovate, elliptical and elliptical to lanceolate), leaf margin (superficially, prominent, crenate and crenulate), leaf apex (acute, acuminate, slightly aristate and sub acute), leaf base (broadly cuneate, round, narrowly cuneate and truncate) and leaf surface (dull rough and shiny smooth) in rainfed semi-arid ecosystem of western India (Singh *et al.*, 2012, 2015) (Table 2). The bark colour was yellowish grey, grayish yellow, blackish grey, dark grey and light whereas splitting pattern was irregular intersecting striations having small rectangular blocks (Singh *et al.*, 2011b). Phyllotaxy in different genotypes was observed Tristichous and Pentastichous (Singh *et al.*, 2019)(Table 1).

Table 1. Morphological variability in leaf characters of bael accessions

Genotypes	Central leaf shape	Lateral leaf let shape	Leaf apex	Leaf base	Leaf margin	Phyllotaxy
CHESB -1	Ovate	Ovate	Aristate	Narrowly cuneate	Superficially crenulate	Tristichous
CHESB -5	Ovate	Ovate	Acute	Broadly cuneate in central oblique cuneate in laterals	Big dentate crenate but depressed at one side	Pentastichous
CHESB -8	Ovate	Ovate	Acute	Rounded	Big dentate crenate superficially	Pentastichous
CHESB -11	Elliptical to lanceolate	Ovate	Central acuminate, laterals acute	Narrowly cuneate, laterals had rounded	Crenate superficially	Tristichous
CHESB -16	Broadly lanceolate	Ovate	Acuminate	Broadly Cuneate but oblique at one side	Small crenulate, superficially	Pentastichous
CHESB -21	Ovate	Ovate	Acute	Rounded	Big dentate crenate superficially	Tristichous
CHESB -27	Elliptical to lanceolate	Elliptical lanceolate	Slightly aristate to acute	Central narrowly cuneate, lateral leaflet rounded	Small crenulate, superficially	Pentastichous
CHESB -29	Ovate	Ovate	Acute	Broadly cuneate in central oblique cuneate in laterals	Crenate but slightly depressed at one side	Tristichous
CHESB -31	Ovate	Ovate	Acute	Rounded	Small crenulate superficially	Trstichous

Table 2. Morphological variability in vegetative characters of bael accessions

Genotypes	Leaf surface	Leaf colour	Growth habit	Foliage	Bark colour	Bark splitting pattern	Thorniness
CHESB-1	Shiny smooth	Dorsal side dark green and ventral side light green	Semi-spreading	Dense	Yellowish grey	Irregular intersecting striations having small rectangular blocks	Thornless
CHESB-5	Shiny, smooth	Dorsal side dark green and ventral side light green	Upright spreading	Compact	Dark grey	Irregular intersecting striations having small rectangular blocks	Very less thorn
CHESB-8	Shiny smooth	Light green at both side	Semi spreading,	Dense	Light grey	Irregular intersecting striations having small rectangular blocks	Less thorn
CHESB-11	Shiny Smooth	Dark green at both side	Semi spreading drooping	Sparse	Light grey	Irregular intersecting striations having small rectangular blocks	Thorny (Thin and small)
CHESB-16	Dull rough	Dark green at both side	Semi spreading, drooping	Sparse	Greyish Yellow	Irregular intersecting striations having small rectangular blocks	Thorny (small)
CHESB-21	Dull rough	Light green at both side	Semi spreading	Dense	Light grey	Irregular intersecting striations having small rectangular blocks	Thorny (very bold)
CHESB-27	Dull smooth	Dorsal side dark Green and light green ventral side	Spreading	Compact	Dark grey	Irregular intersecting striations having small rectangular blocks	Thornless
CHESB-29	Shiny, smooth	Dorsal dark green, ventral side light green	Semi spreading	Dense	Light grey	Irregular intersecting striations having small rectangular blocks	Thorny
CHESB-31	Shiny smooth	Light green at both side	Semi-spreading	Dense	Yellowish	Irregular intersecting striations having small rectangular blocks	Thorny

Flower events

Though flower showed a slight change in colour, majority were dull greenish white with appealing fragrance. Even in the same accessions number of petals may be varied from 4 to 6 (Fig. 1). Flower initiation could be observed 9-15 days after emergence of new flush and flower bud appeared in axillary cymose (Fig. 2). Flower bud appeared as minute swollen structure on inflorescence peduncles and developed into flower between 21-30 days after initiation. All the flowers in a penicle opened with in 7-10 days. Flower open from top to bottom of the penicle. Anthers and petals turn brownish in colour after 8-12 hours of anthesis (Fig. 3). Stigma also turned slightly brownish in colour after 8-10 hours of flower opening though wet surface observed. Anthers attached longitudinally

to the style and after dehiscence of anthers (Fig.3), diverted to outside of the stigma this may be the one of the avoidance mechanism of self pollination. Anthers change to dull brownish colour after 2-3 hours of direct sun light exposure became more side tracked outside of the stigma. Purple colour stigma in CHESB-29 (Fig.5) and curved petals in CHESB-16 (Fig.4) were found as distinct characters of these accessions. Flowers were protogynous and dichogamy (Fig. 6).

Floral biology

The data on the morpho-metrics of flower of bael accessions depicted in Table 4, 5 and 6 showed considerable differences for all the floral characters studied. Flower characters with respect to bud size in terms of bud length



Fig. 1. Variation in number of petals in same accession



Fig.2. Flower buds arrangement



Fig. 3. Dehiscent anthers



Fig. 4. Flower on trunk with curved petal while dehiscence



Fig. 5. Purple colour stigma



Fig. 6. Herkogamy

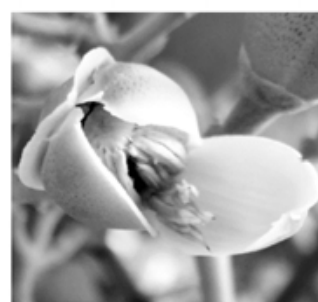


Fig. 7 a. Initiation of petals



b. Opening of petals



c. Complete opened flower and



d. Initiation of dehiscence

(10.0-13 mm), bud breadth (6.84-9.53 mm), flower length (12.05-117.00 mm), flower breadth (23.05-35.17 mm), pedicel length (7.00-9.00 mm), pedicel breadth (1.94-2.50 mm), petal length (11.95-16.54 mm), petal breadth (7.00-9.50 mm), stamen length (6.50-8.61 mm), filament length (5.00-6.90 mm), breadth (0.47-0.73 mm), anther length (4.33-8.00 mm), breadth (0.62-0.84 mm), pollen diameter (40.51-45.53 micron), ovary length (4.33-8.00 mm), diameter (3.00-5.55), style length (1.00-1.50 mm), width (2.00-2.50 mm), stigma length (2.63-3.53 mm), breadth (2.07-3.00 mm) and pistil length (7.29-10.45 mm) exhibited wide variations in their floral organs. Size and shape of flower buds varied in different accessions possibly owing to genetic variation. More or less similar findings for variability in flower organs has been reported in jamun (Singh *et al.*, 2016 b), tamarind (Singh and Singh, 2005, Singh *et al.*, 2008, 2010), chironji (Singh *et al.*, 2010), khirmi (Singh *et al.*, 2016), wood apple (Yadav *et al.*, 2017; Singh *et al.*, 2016 c), custard apple (Yadav *et al.*, 2017),

bael (Singh *et al.*, 2014b, 2018b, 2019b) and wild noni (Singh *et al.*, 2014a). Pollen viability was found to be significantly highest in CHESB-5 (95.67%), whereas it was recorded least B-21 (90.17%) among the accessions. Bud length was the minimum in CHESB-16 (13.47 mm) and same was recorded lowest in CHESB-29 (10.02 mm) whereas flower length was recorded the highest in CHESB-21 (17.00 mm) and it was CHESB-29 (12.02 mm). Similar finding for variation in flower organs in bael varieties has been reported by Singh *et al.* (2018a). Bud and flower breadth were recorded the minimum value in CHESB-29, whereas bud and flower breadth were recorded the maximum in CHESB-5 (9.53mm) and in CHESB-21 (35.17), respectively. Number of stamens per flower was recorded the highest in CHESB-8 (43.35) and lowest value of the same was recorded in CHESB-29 (37.25) among the accessions. Among the accession, pedicel size (7.00x1.94mm), stamen size (6.50mm), filament size (6.90x0.47mm), anther size (2.50x0.62 mm), pollen diameter

(40. 51 micron), pistil length (7.29 mm), ovary size (4.33x3.00mm), style size (1.00x2.00mm) and stigma size (2.63x2.07 mm) was recorded the minimum value in CHESB-29, whereas these values were recorded the highest in different accessions. The maximum breadth of petal, anther, filament, ovary, style and stigma was recorded 9.50, 0.84, 0.73, 4.55, 2.50, 1.50 and 3.53 mm in CHESB-11, CHESB-1, CHESB-11, CHESB-1, CHESB-27, CHESB-16 and CHESB-31, respectively. These finding are agreement with the results reported by Singh *et al.*, (2014a, 2014b, 2014c, 2014d) in bael varieties under rainfed semi-arid conditions. In different accessions, types of flowers were observed axillary biparous, axillary multiparous and compound cyme (Singh *et al.* 2018a, 2018b, 2019a). Commencement of bud emergence was observed from 20th April and lasted up to 05th July whereas

flowering started from 14th May and lasted up to 25th July. The maximum flowering duration was observed in CHESB-1 (Singh *et al.*, 2014e and 2014f) (Table 3).

Anthesis and dehiscence

Under hot semi-arid climatic conditions, there was no anthesis before dehiscence in any accession, whereas anthesis in bael started from 4.30 am and continued up to 12.30 pm. Some flowers opened all petals at a time while in other petals start opening one by one which takes 45 to 60 minutes in complete opening which may vary flower to flower. In the inflorescence, centrally located buds open first as compared to lateral buds. After anthesis, the hint of the anthers dehiscence started which continued till noon (Fig. 7). Among the varieties, the maximum anthesis was observed in

Table 3. Morphological variability in phenological characters in bael accessions

Genotypes	Type of inflorescence	Flower bud emergence			Flowering period			Number of petals	
		Start	Peak period	End	Start	Peak period	End	Common	Rare
CHESB-1	Axillary multiparous cyme	30 April	24-25 May	05 July	14 May	20-30 June	25 July	4,5	6
CHESB-5	Compound cyme	20 April	23-28 May	20 June	16 May	9-19 June	23 June	4,5	6
CHESB-8	Axillary cyme	8 May	23-28 May	22 June	26 May	7-18 June	24 June	4,5	5
CHESB-11	Axillary multiparous cyme	3 May	24-25 May	20 June	24 May	9-16 June	23 June	4,5	---
CHESB-16	Axillary cyme	1 May	23-28 May	16 June	26 May	10-18 June	19 June	4	5
CHESB-21	Axillary biparous cyme	20 April	24-29 May	16 June	15 May	7-19 June	28 June	4,5	---
CHESB-27	Axillary biparous cyme	1 May	25-30 May	19 June	21 May	9-16 June	27 June	4,5	6
CHESB-29	Axillary multiparous cyme	21 April	23-28 May	24 June	15 May	9-16 June	25 June	4	5
CHESB-31	Axillary biparous cyme	20 April	24-29 May	20 June	16 May	10-20 June	24 June	4	----

Table 4. Morphological variability in flower characters in bael accessions

Genotypes	Bud size (mm)		Flower size (mm)		Petal size (mm)		Pedicel size (mm)	
	Length	Breadth	Length	Breadth	Length	Breadth	Length	Thickness
CHESB-1	12.05	8.48	13.43	27.10	14.34	9.05	7.40	2.07
CHESB-5	12.14	9.53	14.53	35.20	15.24	7.50	7.25	2.20
CHESB-8	11.25	8.57	14.47	25.07	14.17	8.00	4.78	2.00
CHESB-11	10.48	7.25	16.57	32.13	16.54	9.50	8.33	2.50
CHESB-16	13.47	9.73	14.07	24.27	15.00	8.50	6.23	2.00
CHESB-21	13.00	8.50	17.00	32.31	16.00	7.53	9.00	2.00
CHESB-27	13.00	8.51	12.30	35.17	12.08	9.59	5.70	2.40
CHESB-29	10.02	6.84	12.05	23.05	11.95	7.00	4.58	1.94
CHESB-31	12.00	9.50	18.00	26.08	18.45	8.50	7.00	2.13
C.D. at 5%	1.21	0.41	0.75	1.42	0.72	0.81	0.70	0.17

Table 5. Variability in flower characters in bael accessions

Genotypes	Number of stamens/ flower	Stamen size (mm)	Filament size (mm)		Anther size (mm)		Pollen diameter (micron)
		Length	Length	Breadth	Length	Breadth	
CHESB -1	42.19	6.50	3.50	0.50	3.00	0.62	42.53
CHESB -5	38.97	7.00	3.53	0.72	3.54	0.70	45.00
CHESB -8	43.35	8.60	5.00	0.50	3.05	0.81	43.74
CHESB -11	43.30	7.78	4.07	0.73	3.75	0.84	41.28
CHESB -16	38.25	7.50	4.76	0.54	2.75	0.70	41.27
CHESB -21	39.00	7.43	4.00	0.70	3.55	0.77	43.03
CHESB -27	38.25	8.61	3.20	0.60	5.26	0.72	43.07
CHESB -29	37.25	6.90	3.00	0.47	2.52	0.70	40.51
CHESB -31	39.68	7.53	3.45	0.70	4.50	0.75	42.47
C.D. at 5%	--	0.31	0.22	0.04	0.23	0.04	2.17

Table 6. Variability in floral organs in bael accessions

Genotypes	Pistil length (mm)	Ovary size (mm)		Style size (mm)		Stigma size (mm)		Pollen viability (%)
		Length	Breadth	Length	Breadth	Length	Breadth	
CHESB -1	10.44	7.57	4.55	1.50	2.50	3.20	2.50	94.10
CHESB -5	09.27	5.54	4.00	1.00	2.50	3.40	2.50	95.67
CHESB -8	10.35	6.52	4.49	1.10	2.05	3.12	2.20	94.27
CHESB -11	09.00	4.07	4.37	1.53	2.50	3.53	2.07	95.17
CHESB -16	09.57	4.28	3.03	1.00	1.55	3.00	2.51	92.05
CHESB -21	07.00	4.50	2.50	1.05	2.31	3.08	2.32	90.17
CHESB -27	10.45	5.50	3.51	1.10	2.50	3.10	2.50	93.22
CHESB -29	07.29	4.33	3.00	1.00	2.00	2.63	2.40	91.35
CHESB -31	07.75	8.00	3.53	1.05	2.10	3.50	3.00	95.22
C.D. at 5%	0.53	0.33	0.21	0.15	0.13	0.11	0.33	4.94

Goma Yashi between 6.31 am to 8.30 am, whereas in all the varieties, maximum anthesis was observed during this period. Among the varieties, the highest anthesis was observed in Thar Neelkanth between 8.31am -10.30 am. In all the varieties, maximum anthesis between 8.31am - 12.30 pm was observed in Thar Divya (Fig 8). Early anthesis was observed in eastward branches compared to rest of the branches owing early exposure of branches to sunlight. Generally, anther dehiscence was synchronous with anthesis and all the anthers in the flower in flower dehiscence synchronously presenting the

pollen mass. More or less similar pattern of anthesis was observed by Singh *et al.*, (2009) and Singh *et al.*, (2014e, 2014f & 2019b).

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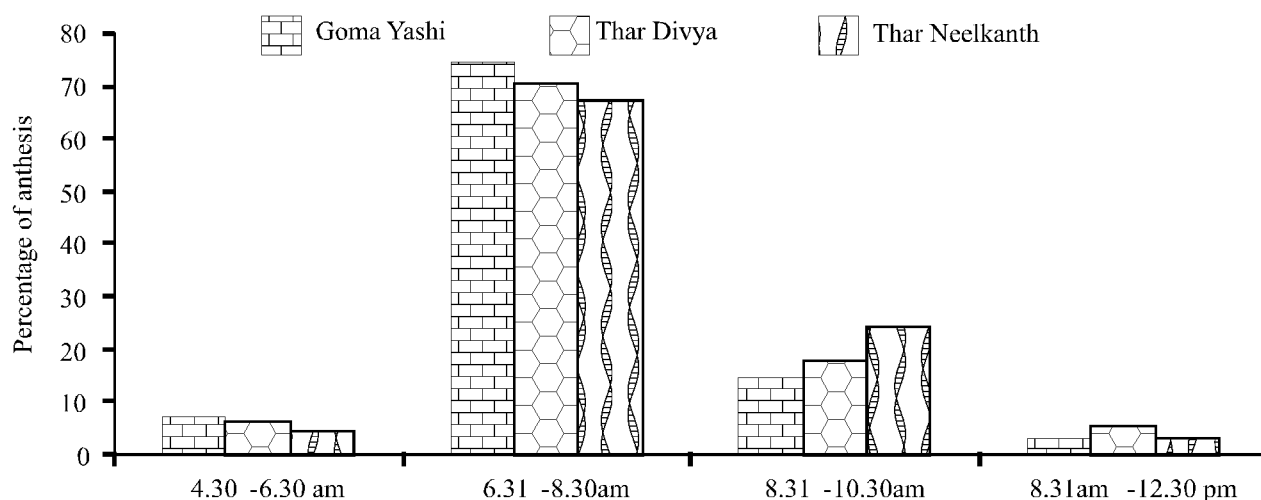


Fig 8. Percentage of dehiscence with the time in bael varieties

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