

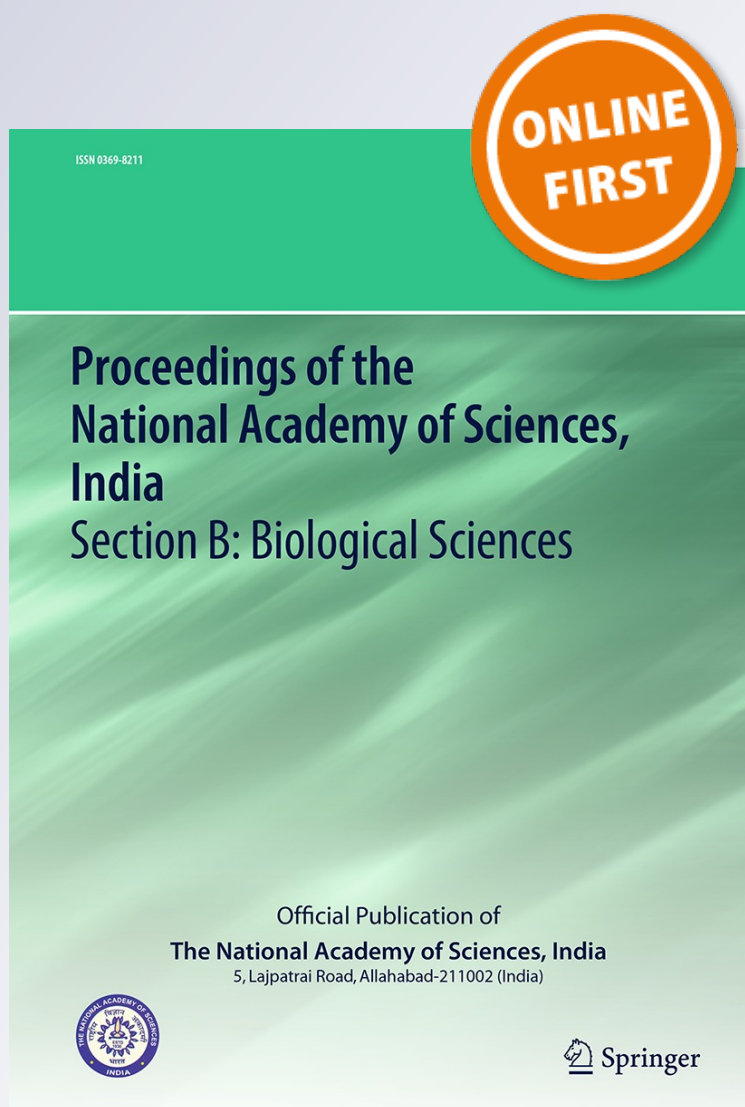
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Qualitative and Quantitative Seed Characteristics Diversity from Girnar Reserve Forest, Gujarat, India

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Abstract Seeds of 97 plant species were studied for morphological characteristics after collection during botanical field visits from Girnar Reserve Forest, Gujarat, India. There was a significant variation in quantitative characters of seeds, while qualitative traits also exhibited great diversity. Seeds of trees showed highest values for all quantitative features measured as volume, length, width, thickness and weight among all other botanical group such as climbers, herbs, undershrubs and shrubs during the study. The range of seed volume varied from 0.01 to 21.48 cm³ for 63 trees, 0.23 to 1.42 cm³ for 12 herbs, 0.31 to 0.72 cm³ for 10 undershrubs, 0.35 to 0.93 cm³ for 9 shrubs, and 0.37 to 1.77 cm³ for 3 climbers. Fruiting time varied for species within groups, but mostly for all plants seed collection time was during dry season after fruit ripening. In the principal component multivariate analysis, only one principal component had an Eigen value above one and hence found significant, which explained 73 % morphological variation in the whole data set. The separation of 97 species using PCA and cluster analysis was

mostly in agreement with their group specific taxonomical identification of each plant. This study not only provided information related to seed morphology and taxonomy, but can be used effectively in diversity study as well as social forestry programmes.

Keywords Seed morphology · Qualitative and quantitative characters · Principal component analysis · Cluster analysis · Girnar reserve forest · Seed collection time

Introduction

Plants produce seeds with enormous variation. Understanding this variation can contribute to improvement in nursery management, forest restoration and genetic structure of crops. The most critical and crucial aspect of nursery management is seed collection which is followed by identification. Local forest seeds are of interest since they corroborate with local climate, hence selected material could be of high use in suggesting guidelines for germination in that particular area as these seeds are better adapted to local climatic conditions leading to better vegetation establishment [1]. Understanding the seed within the local forest is of massive importance as it contributes to study areas of seed storage, longevity and viability in addition to forest regeneration [2]. Without understanding variability for characters of interest, genetic improvement for different species will be failed. But most important work is to identify proportion of variations present in set and it is of immense use how one can exploit this variation to meet needs of plant growers. However, it is known that phenotypic and genotypic variation exist in all species which offer a great chance to go for selection or screening

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of seed source with desirable characters which furthermore deals with seed improvement in social forestry experiments [1, 3]. Diversity study of seeds in forest is necessary because it helps in seed dispersal process studies within the forest, which also includes conservation aspects if seed types are documented [4].

Use of seed morphological parameters in taxonomical delimitation of plant species is an old approach among scientists [5]. Seeds vary in shape from filiform to fusiform, clavate to ellipsoidal and sometimes prominently winged [6]. In addition, in some cases the seed is covered by a hard coat but in most cases the seed coat is papery in texture and loosely surrounds the embryo [7]. Morphological characteristics of the seeds not only serve as taxonomical markings but also serve in deducing phylogenetic relationships [8, 9]. Seed morphology serves as a source of systematic character to describe sub-generic groups or hypothetical relationships among species within a genus [10–12]. Documenting variation in morphology of seeds is related to ecological aspects of viable seeds, germination, pollination and phenology of the species [13]. Furthermore, seed morphological variations play vital role in understanding and identifying heteromorphism of the species [14]. Strategies for the identification of functional genetic variations underlying phenotypic traits have received considerable importance [15]. The polymorphism is also associated with seed weight, germination, seedling growth and seed dry weight [16]. Nevertheless, those seedlings which were produced from large seeds had higher biomass as well as extreme water tolerance capacity [17]. Studying vegetation distribution pattern is a basic aspect of design and management; However using techniques, quantitative separation was studied by previous workers to investigate contribution of environmental factors to plant community distribution pattern [18]. Without identification it would not be easy to develop methods for growing useful plants hence, those plants which are not considered much useful or undesirable or poisonous are also of much use in documentation of further research strategies [6]. In seed identification using morphological characters, available information is given only in bits, here and there where as in most of the cases the works are mainly concerned with other aspects of plant sciences.

Girnar Reserve Forest is a unique place of biodiversity in the Saurashtra region of Gujarat. It has at least 700 reported plant species [19] but details on seed morphology are only documented for nine tree species [20]. To remedy this situation, current work is requirement of time for describing overall seed morphological characters of trees, herbs, shrubs, undershrubs and climber like groups and compare them using direct morphological observations. Hence, the aim of this study was to describe and compare external seed morphological characters for five different

groups' viz. trees, herbs, shrubs, undershrubs and climbers of Girnar Reserve Forest, and to evaluate their possible use for taxonomic delimitation.

Material and Methods

Study Area with Climate

Girnar Reserve Forest (21°30'–21°26'N longitude and 70°28'–70°27'E latitude) is in the south Saurashtra region of Gujarat state in western India. With spread over 196 km, it is one of the major biodiversity centers of the area. It is 7 km away from main city of Junagadh. Different subareas of the Girnar Reserve Forest are Jatashankar, Bordevi, Datar, Bharatvan-Sheshavan, Jhinababani Madhi, Sharakadiya Hanuman area, Kashmiri Bapu Ashram area, Area near Gayatri Mandir, Area near Prerana Dham Ashram etc. Girnar Reserve Forest is a sub-tropical deciduous forest, dominated by teak. The year is divisible into three seasons: summer (March to mid June), monsoon (late June to October) and winter (November to February). Out of the total rainfall, 95 % occurs in the monsoon season. During the study period March, April and May temperature reached above 40 °C during all the 3 years. January and February showed the lowest temperature for all respective years although average varied with values of 19 and 21 °C in 2008–2009, 22 and 24 °C in 2009–2010 and 2010–2011. Wind speed averaged 6–8 km h⁻¹ during all the study years.

Plant Materials and Seed Sampling

Seeds of 97 plants species of trees, herbs, shrubs, undershrubs and climbers, were collected from different areas within the Girnar Reserve Forest, Junagadh from August 2008 to 2011. All plant species were identified using regional Floras, Flora of Gujarat [21] and Flora of Saurashtra [22]. The approximate time for seed collection was determined from an earlier study on phenology [23]. Approximately 800 ± 100 seeds were collected from the ground or from ripened fruits on the plant and brought to the laboratory in the paper bags for 2–3 days in every week during whole study period of 3 years. Seeds were oven dried to control moisture content according to the method given by Chalm et al. [24].

Morphological Analysis

The qualitative and quantitative observations on seed characters were noted on a data sheet devised by Gavitt [25]. Using Vernier scale, measurements of seed dimension viz. length, width, thickness and volume were carried out.

Length was measured parallel to hilum while width of the seed was measured from the center of the hilum to opposite side of seed and right angle to the plane of hilum. Thickness was measured at right angle to hilum and right angle to plane of hilum. For the weight of individual seed or average of 10–100 seeds, digital balance of 0.001 g was used. The qualitative characters as shape, colour, hilum shape and surface were studied using method given by Chalm et al. [24]. Hilum shapes were described by the terminology given by Gunn [26] and Gavit and Parabia [27].

Statistical Analysis

One way ANOVA was carried out for studying variation among five different botanical groups of seeds whereas linear correlation was also done to assess different morphological data for the study. The statistical analysis was done using methods given by Gomez and Gomez [28]. Principal component analysis was invented by Carl Pearson and now it is widely-used tool in the data analysis in research which is used in predicting models whereas, cluster analysis seeks to divide different parameters into the groups of high internal similarity with respect to species or characters used. It is also called association analysis where communities are defined by the presence or absence of single species. Cluster analysis and other pair wise analysis methods are older methods but now more accurate trees of clusters are created by using newer methods such as neighbour joining [29]. Principal component multivariate type of analysis along with the cluster analysis were carried out by the software SPSS to identify significant PCs and to find diversity among 97 species using seed morphological data. PCA uses a rigid rotation to derive orthogonal axis, which maximizes variance in the data set. Bi-plot explains various associations between different traits where negatively correlated traits or characters are arranged at cosine of 180° as its value is minus one while those traits where cosine angle is 90° have no correlation as its value is zero.

Results and Discussion

Results

The study on 97 plant species, exhibited large variation in qualitative and quantitative seed characteristics. Interestingly, two Meliaceae members, *Azadirachta indica* A. Juss and *Melia azadirachta* L. showed difference in seed colour, shape, hilum shape as well as surface structure. Similarities as well as dissimilarities among same genus but different species were found in the study also. There was similar

qualitative traits for all *Ficus* sp., viz. *Ficus benghalensis* L., *Ficus glomerata* Roxb. and *Ficus religiosa* L. but showed little variation in quantitative traits. Combretaceae species, *Terminalia arjuna* (Roxb.) W. & A., *Terminalia bellerica* (Gaertn.) Roxb. and *Terminalia chebula* Retz. showed high variation for qualitative and quantitative parameters measured (Tables 1, 2; Suppl. Fig. 1).

In general, trees had larger seeds. Order of the average lengths from highest to lowest was: trees (1.29 ± 0.65) > climbers (1.04 ± 0.70) > herbs (0.64 ± 0.32) > shrubs (0.60 ± 0.22) > undershrubs (0.44 ± 0.13). Descending order of the average width for different groups of plants was: trees (0.78 ± 0.45) > climbers (0.60 ± 0.37) > shrubs (0.41 ± 0.12) > herbs (0.38 ± 0.19) > undershrubs (0.29 ± 0.09) cm. The same order was repeated for the average thickness also which was, trees (0.47 ± 0.44) > climbers (0.45 ± 0.26) > shrubs (0.30 ± 0.19) > herbs (0.27 ± 0.14) > undershrubs (0.10 ± 0.05). For the seed volume descending order was, trees (1.08 ± 2.91) > climbers (0.49 ± 0.20) > herbs (0.10 ± 0.09) > shrubs (0.09 ± 0.07) > undershrubs (0.01 ± 0.01) cm^3 . Seeds of *Terminalia arjuna* (Roxb.) W. & A were largest among all species by showing highest length (3.19 cm), highest width (2.56 cm), highest thickness (2.64 cm) and average highest seed volume (21.48 cm^3). However, smallest seeds varied with lowest length for *F. benghalensis* L. (0.21 cm), lowest width *Bombax ceiba* L. (0.17 cm), lowest thickness for *Nyctanthus arbortristis* L. (0.06 cm) and lowest seed volume is shown by two species of *Ficus* viz. *Ficus glomerata* Roxb. and *Ficus religiosa* L. (0.01 cm^3). Maximum values for weights were recorded for *Mangifera indica* L. (2.62 g) for trees, *Corchorus fascicularis* Lam. (0.41 g) for herbs, *Datura innoxia* Mill. (0.39 g) for undershrubs, *Zizyphus rotundifolia* Lam. (0.07) for shrubs and for, *Diplocyclos palmatus* (L.). C. Jeffrey. (0.41 g) for climbers (Table 2).

All the correlations among quantitative seed characteristics in tree species were positively significant, while most correlations were not significant for other plant groups (Table 3). There was highest positively significant correlations between length and width for all groups but value of correlation coefficient was highest (0.99**) in 3 climbers as compared to other values for 63 trees (0.83**), 12 herbs (0.76**), 9 shrubs (0.75*), and 10 undershrubs (0.67*). The analysis of variance indicated significant variation in different species among each of five different botanical groups in the Girnar Reserve Forest (Table 4).

The multivariate type of analysis, showed 5 total PCs exhibiting total cumulative variation of 100 % in whole data set. PC 1 was the only significant principal component which showed Eigen value more than one. In the PC 1, width was the dominating trait by showing highest value (0.94) followed by thickness (0.92), length (0.89), volume

Table 1 Qualitative characters of seed of four different botanical groups from Girnar Reserve Forest, Gujarat

No.	Species	Habit	Colour	Surface	Shape	Hilum shape
1.	<i>Abelmoschus esculentus</i> (L.) Moench.	H	Blackish brown	Rugose, glabrous	Wedge shaped, oblong, subreniform	Ovate, sub ovate
2.	<i>Acacia nilotica</i> (L.) Del.	T	Blackish yellow ring	Rough, glabrous	Oblong to ovate	Ovate
3.	<i>Acacia senegal</i> Willd.	T	Dark brown to blackish brown	Smooth, shining	Ovate to obovate	Circular
4.	<i>Acalypha indica</i> L.	US	Brownish colour to reddish brown	Glabrous and granular	Ovate, subovate	Circular
5.	<i>Adansonia digitata</i> L.	T	Reddish brown with black tinge, yellow spots	Rough, granular	Reniform, kidney shaped	Circular, oblong
6.	<i>Aegle marmelos</i> (L.) Corr.	T	Yellow, whitish light yellow	Rugose, glabrous	Sub ovate, Ovate, Broad elliptic	Circular
7.	<i>Ailanthus excelsa</i> Roxb.	T	Yellowish brown, light brown, light yellow	Granular, glabrous	Ovate, subovate, Kidney shape	Elliptic
8.	<i>Alangium salvifolium</i> (L.f) Bth.	T	Yellowish brown	Rough, granular	Ovate to broad elliptic	Elliptic
9.	<i>Albizia lebbek</i> (L.) Bth.	T	Yellowish brown, U shaped black line	Rough, glabrous	Ellipsoidal, subellipsoidal	Elliptic
10.	<i>Alstonia scholaris</i> R.Br.	T	Brownish black, Light brown	Rough, glabrous	Broad elliptic, ellipsoidal	Circular
11.	<i>Anisomeles indica</i> (L.) O. Ktze.	US	Brownish black	Smooth, shining	Oblong, subobong like coffee seed	Circular
12.	<i>Argemone mexicana</i> L.	H	Black to light black	Smooth, pitted, glabrous	Ovate, subovate	Circular
13.	<i>Argyreia nervosa</i> (Burm.f.) Boj.	C	Greenish brown	Rugose, glabrous	Broad elliptic, trigonal shaped	Circular, ovate type
14.	<i>Azadirachta indica</i> A. Juss	T	Yellow, whitish yellow	Rough, granular, horizontally septed	Ovate to sub elliptic	Circular
15.	<i>Balanites aegyptiaca</i> (L.) Del.	T	Yellowish, light yellow	Rough, transversely ribbed, shrinked	Spindle shaped, elliptic, broad elliptic	Circular
16.	<i>Bauhinia purpurea</i> L.	T	Reddish brown, blackish brown	Rough, Glabrous	Ellipsoidal, subellipsoidal	Circular, ovae
17.	<i>Bauhinia racemosa</i> Lam.	T	Blackish brown	Smooth, shining	Ellipsoidal, sub ovate	Circular
18.	<i>Butea monosperma</i> (Lam.) Taub.	T	Brownish	Rough, glabrous	Oblong, subreniform	Elliptic, ovate, linear
19.	<i>Boerhavia chinensis</i> (L.) Druce	H	Brown yellow	Smooth	Wedge shaped to oval	Circular
20.	<i>Bombax ceiba</i> L.	T	Reddish to blackish brown	Glabrous, granular, rough	Ovate, sub ovate, ellipsoidal	Circular, narrow elliptic
21.	<i>Caesalpinia crista</i> L.	T	Lead colour, light blue	Smooth, polished, glabrous	Broad elliptic, oblong, circular	Circular
22.	<i>Caesalpinia pulcherrima</i> (L.) Sw.	S	Brown to blackish brown	Rough, glabrous	Trigonal, ovate, broad elliptic	Circular to elliptic
23.	<i>Cassia auriculata</i> L.	S	Brownish yellow	Rough, granular	Ellipsoidal, subellipsoidal	Circular
24.	<i>Cassia biflora</i> L.	S	Brownish black	Smooth, shining	Tuber shaped, Linear	Circular
25.	<i>Cassia fistula</i> L.	H	Reddish to yellowish brown	Smooth, shining	ovate, sub ovate	Circular, ovate
26.	<i>Cassia occidentalis</i> L.	US	Greenish or Yellowish brown	Rough, glabrous	Ellipsoidal	Circular
27.	<i>Cassia siamea</i> Lam.	T	Brownish black	Rough, shining	Ovate, sub ovate, ellipsoidal	Circular
28.	<i>Cassia tora</i> L.	H	Brownish black	Smooth, shining	Tuber type, subtuberous	Circular
29.	<i>Casuarina equisetifolia</i> L.	T	Brownish black	Rough	Reniform, ovate, obovate	Circular
30.	<i>Ceiba pentandra</i> L.	T	Black, light black	Rough, granular	Sub reniform, sub ovate, oblong	Circular

Table 1 continued

No.	Species	Habit	Colour	Surface	Shape	Hilum shape
31.	<i>Clitoria ternatea</i> L.	C	Light black to black, with spots	Smooth, glabrous	Reniform, kidney shaped	Circular, ovate, oblong
32.	<i>Corchorus fascicularis</i> Lam.	H	Black to brownish black	Rough with granules	Ovate, obovate, linear	Circular
33.	<i>Cordia gharaf</i> (Forsk.) E & A.	T	Yellowish or brownish yellow	Glabrous, rough, granular	Ovate, subovate, oblong	Circular to oblong
34.	<i>Crateva nurvala</i> Buch-Ham	T	Blackish brown	Rough, granular	Curved, comma shaped	Circular, elliptical
35.	<i>Crotalaria medicaginea</i> Lam.	H	Dark brown	Smooth, glabrous, shining	Circular	Circular, ovate
36.	<i>Dalbergia paniculata</i> Roxb.	T	Dark brown to yellowish brown	Smooth, glabrous	Subreniform, kidney shaped	Circular, round shape
37.	<i>Dalbergia sissoo</i> Roxb.	T	Brownish to reddish black	Glabrous, rough	Reniform, sub reniform	Circular to linear
38.	<i>Datura innoxia</i> Mill.	US	Yellow brownish black	Rough, glabrous	Kidney shaped, reniform	Circular
39.	<i>Delonix regia</i> (Boj.) Raf.	T	Yellow with brown spots	Smooth, transversely septed	Elliptic, oblong, suboblong	Circular
40.	<i>Derris indica</i> (Lam.) Bennet	T	Reddish to blackish brown	Rough, shrinked, transversely ribbed	Reniform, kidney shaped, subreniform	Circular, ovate
41.	<i>Desmodium gangeticum</i> (L.) DC	US	Brown	Smooth	Ovate, subovate	Circular
42.	<i>Dichrostachya cinerea</i> (L.)W. & A.	S	Reddish brown	Smooth, shining	Ellipsoidal, oblong	Circular to elliptical
43.	<i>Diospyros melanoxylon</i> Roxb.	T	Brownish black, brown, black	Rough, pitted, shrinked	Ovate, subovate, oblong, suboblong	Circular to elliptical
44.	<i>Diplocyclos palmatus</i> (L.) C. Jeffrey	C	Creamy white, pale yellow	Rough, granular	Ovate to obovate	Elliptic
45.	<i>Erythrina indica</i> Lam.	T	Brownish red	Smooth, glabrous	Sub reniform, sub oblong	Oblong, elliptical
46.	<i>Eucalyptus globules</i> Labill.	T	Cremy white, Brownish white	Smooth, glabrous, granular	Oval, ovate, obovate, sub ovate	Circular, oblong
47.	<i>Ficus benghalensis</i> L.	T	Golden yellow, yellow	Smooth, glabrous	Ovate to obovate	Circular
48.	<i>Ficus glomerata</i> Roxb.	T	reddish yellow	Smooth, glabrous	Ovate, circular, trigonal, sub ovate	Circular
49.	<i>Ficus religiosa</i> L.	T	Golden yellow, yellow	Smooth, glabrous	Ovate to obovate	Circular
50.	<i>Gmelina arborea</i> L.	T	White to brown	Smooth	Ovate to obovate	Circular
51.	<i>Helicteres isora</i> L.	S	Brownish to blackish brown	Rough, granular	Ovate, sub ovate, flat	Circular
52.	<i>Holarrhena antidysenterica</i> (Heyne.ex Roth) Wall	T	Brownish black, Light brown	Glabrous, rough	Winged	Circular
53.	<i>Holoptelea intergrifolia</i> (Roxb.) Planch.	T	Light yellow, Light brown	Rough, granular	Subreniform, ovate	Circular
54.	<i>Indigofera tinctoria</i> L.	H	Black	Rough, granular	Kidney shaped, reniform	Circular, oblong
55.	<i>Indigofera trifoliata</i> L.	H	Yellowish brown	Smooth, glabrous	Reniform, kidney shaped	Elliptic to sub elliptic
56.	<i>Lagerstroemia speciosa</i> (L.) Pers.	T	Yellowish to reddish brown	Smooth, glabrous	Ovate, sub ovate, flat	Linear, elliptic,
57.	<i>Lannea coromandelica</i> (Houtt.) Herrill	T	Yellowish to light brown	Glabrous, pitted	Kidney shaped, reniform	Circular
58.	<i>Lawsonia inermis</i> L.	S	Yellowish to blackish brown	Glabrous, smooth	Wedge shaped, trigonal	Circular
59.	<i>Leea indica</i> (Burm.f.) Merrill.	S	Greenish brown	Rugose, glabrous	Kidney shaped, reniform	Elliptic, linear

Table 1 continued

No.	Species	Habit	Colour	Surface	Shape	Hilum shape
60.	<i>Leucaena leucocphala</i> (Lam) de Wit	T	Blackish brown	Smooth, shining	Ellipsoidal, subellipsoidal	Circular
61.	<i>Madhuca indica</i> J.F. Gmel.	T	Yellowish brown	Shining with some spots	Ovate to sub ovate	Elliptic to linear
62.	<i>Mangifera indica</i> L.	T	Whitish yellow	Rugose, Glabrous, granular	Kidney shaped, reniform	Circular, elliptic
63.	<i>Manilkara hexandra</i> (Roxb.) Dub.	T	Yellowish brown	Rough, granular, glabrous	Broad or narrow ovate	Circular
64.	<i>Melia azadirachta</i> L.	T	Light brown to dark brown	Smooth, polished,	Ovate, subovate, obovate	Linear, oblong
65.	<i>Merremia terpehthum</i> Shah & Bhatt	H	Pale brownish, brown	Glabrous, smooth	Oblong, suboblong	Ovate, sub ovate
66.	<i>Merremia tridentata</i> (L.) Hall.f.	H	Brownish	Rough, shining, granular, little septed	Oblong, ovate, subovate, sub reniform	Circular, semi circular
67.	<i>Mimusops elengi</i> L.	T	Brownish	Shining, glabrous	Ovate, narrow ovate, oblong	Circular, elliptic
68.	<i>Nyctanthus arbortristis</i> L.	T	Yellowish brown	Smooth, glabrous	Oblong, ellipsoidal	Circular
69.	<i>Peltophorum pterocarpum</i> (DC)Backer ex Heyne	T	Blackish brown	Smooth, shining	Oblong, ovate, subovate, sub reniform	Linear, narrow ovate
70.	<i>Pithocellobium dulce</i> (Roxb.) Bth.	T	Reddish brown	Smooth	Sub ovate, elliptic, transversely septed	Circular
71.	<i>Plumbago zeylanica</i> L.	US	Yellowish brown	Smooth, horizontally septed	Oblong, suboblong	Circular
72.	<i>Polyalthia longifolia</i> (Sonn.) Thw.	T	Brownish white	Smooth, horizontally septed, granular	Oblong, suboblong	Circular, elliptic
73.	<i>Prosopis cineraria</i> (L.) Druce	T	Blackish brown to yellowish brown	Smooth shining	Sub elliptic, ovate, subovate	Circular
74.	<i>Prosopis juliflora</i> (Sw.) DC.	T	Blackish brown	Glabrous, rough, pitted	Broad elliptic, elliptic	Circular, elliptic
75.	<i>Randia spinosa</i> (Thunb.) Bl.	S	Blackish brown with yellow u shaped ring	Smooth, shining	Sub ellipsoidal, suboblong	Circular
76.	<i>Samanea saman</i> (Jacq.) Merr.	T	Light yellow, light brown	Rough, transversely granular	Subreniform, ovate	Circular
77.	<i>Santalum album</i> L.	T	Black	Smooth, glabrous	Oblong, suboblong	Circular, elliptical
78.	<i>Sapindus laurifolius</i> Vahl.	T	Dark brown, Reddish brown	Rough, glabrous	Elliptic, oblong and ovate	Linear, elliptic
79.	<i>Sida acuta</i> Burm. f.	US	Greenish brown	Smooth, glabrous	Trigonal, pyramidal, ovate	Circular, elliptical
80.	<i>Sida cordifolia</i> L.	US	Light black to whitish black	Rugose, glabrous	Wedge shaped, trigonal with two spines at the end	Circular
81.	<i>Soymida febrifuga</i> (Roxb.) A. Juss.	T	Yellowish brown	Rough, glabrous	Oblong, sub reniform	Circular
82.	<i>Sterculia foetida</i>	T	Blackish to light black	Smooth, glabrous, horizontally septed	Oblong, subovate	Circular
83.	<i>Sterculia urens</i> Roxb.	T	Blackish to light brown	Smooth, shining	Broad narrow elliptic	Circular
84.	<i>Syzygium cumini</i> (L.) Skeels	T	Whitish blue	Smooth, glabrous	Ovate, obovate, sub ovate	Linear, elliptic
85.	<i>Tamarindus indica</i> L.	T	Brownish black	Shining, glabrous	Oblong, sub oblong	Circular, suboblong
86.	<i>Tecoma stans</i> (L.) H.B. & K.	T	Light to whitish brown	Rough, glabrous	Winged, oblong, suboblong	Oblong
87.	<i>Tectona grandis</i> L.f.	T	White, yellowish white	Smooth, glabrous	Sub ellipsoidal, trigonal,	Circular

Table 1 continued

No.	Species	Habit	Colour	Surface	Shape	Hilum shape
88.	<i>Tephrosia purpurea</i> (L.) Pers.	US	Brownish black, Light brown	Smooth, glabrous	Oblong, sub oblong	Circular
89.	<i>Terminalia arjuna</i> (Roxb.) W. & A.	T	Blackish brown to yellowish brown	Granular, glabrous	Elliptic with four wings	Circular
90.	<i>Terminalia bellerica</i> (Gaertn.) Roxb.	T	Golden yellow with brown head	Rough, glabrous	Ovate to sub ovate, oblong	Circular
91.	<i>Terminalia chebula</i> Retz.	T	Brownish black, Light brown	Rough, glabrous	Elliptic with wings	Circular
92.	<i>Thespesia populnea</i> (L.) Sol.ex Cor.	T	Brownish black	Rough, rugose, glabrous	wedge shaped, trigonal pyramidal	Circular, ovate
93.	<i>Vernonia anthelmintica</i> (L.) Willd.	H	Brownish white	Glabrous	Winged seed	Circular
94.	<i>Withania somniferum</i> (L.) Dunal	US	Yellowish brown	Glabrous, rough	Trigonal, subreniform, sub ellipsoidal	Circular
95.	<i>Wrightia tinctoria</i> R.Br.	T	Brownish black, Light brown	Glabrous, rough	Winged	Circular
96.	<i>Xanthium strumarium</i> L.	H	Brownish black	Smooth, glabrous	Ovate, sub elliptic	Circular
97.	<i>Zizyphus rotundifolia</i> Lam.	S	Whitish brown to reddish brown	Rough, granular	Ovate, subovate, circular or round shaped	Circular

T: trees, H: herbs, S: shrubs, C: climbers, US: undershrubs shows different plant groups of seeds from Girnar Reserve Forest, Gujarat

Table 2 Mean, range and standard deviation of quantitative seed characteristics among species in four different plant groups: trees, herbs, shrubs, undershrubs and climbers

No.	Traits	Analysis	Different botanical groups				
			Trees (63)	Herbs (12)	Shrubs (9)	Undershrubs (10)	Climbers (3)
1.	Length	Mean	1.29	0.64	0.60	0.44	1.04
		Min	0.21	0.23	0.35	0.31	0.37
		Max	3.19	1.42	0.93	0.72	1.77
		SD	0.65	0.32	0.22	0.13	0.70
2.	Width	Mean	0.78	0.38	0.41	0.29	0.60
		Min	0.17	0.18	0.24	0.14	0.24
		Max	2.56	0.72	0.55	0.46	0.97
		SD	0.45	0.19	0.12	0.09	0.37
3.	Thickness	Mean	0.47	0.27	0.30	0.10	0.45
		Min	0.06	0.08	0.05	0.06	0.17
		Max	2.64	0.52	0.58	0.18	0.68
		SD	0.44	0.14	0.19	0.05	0.26
4.	Weight	Mean	0.33	0.07	0.03	0.05	0.18
		Min	0.00	0.00	0.00	0.00	0.02
		Max	2.62	0.41	0.07	0.39	0.41
		SD	0.50	0.03	0.03	0.03	0.11
5.	Volume	Mean	1.08	0.10	0.09	0.01	0.49
		Min	0.01	0.01	0.01	0.01	0.02
		Max	21.48	0.30	0.26	0.03	1.17
		SD	2.91	0.09	0.07	0.01	0.20

Min: minimum, Max: maximum, SD: standard deviation, in bracket number indicates total number of species studied for particular botanical group, length, width and thickness are expressed in cm while weight and volume are expressed in g and cm³

Table 3 Results from linear correlations between pairs of quantitative seed characteristics in different plant groups

Traits	Trees	Herbs	Shrubs	Undershubs	Climbers
<i>Length</i>					
Width	0.83**	0.76**	0.75*	0.67*	1.00**
Thickness	0.71**	0.53	0.41	-0.14	0.97
Weight	0.46**	0.41	0.57	0.17	0.98
Volume	0.63**	0.85**	0.69*	0.88**	0.98
<i>Width</i>					
Thickness	0.79**	0.79**	0.46	-0.45	0.98
Weight	0.51**	0.05	0.61	0.30	0.97
Volume	0.75**	0.97**	0.70	0.70*	0.97
<i>Thickness</i>					
Weight	0.49**	0.41	0.51	-0.06	0.89
Volume	0.87**	0.77**	0.89**	0.17	0.89**
<i>Weight</i>					
Volume	0.29**	0.20	0.54	0.22	0.99**

All values are r values, and ** indicates significance at 0.01 whereas * shows significance at 0.05 (P value <0.01, <0.05)

Table 4 One way ANOVA (MS values) for variation among species within five botanical groups viz. trees, herbs, shrubs, undershrubs and climbers for quantitative traits

Traits	Analysis	Trees	Herbs	Shrubs	Undershubs	Climbers
Length	Replication	0.01	0.00	0.01	0.00	0.00
	Species	1.43**	0.26**	0.15**	0.05**	1.49**
	Error	0.05	0.00	0.01	0.00	0.00
	Total	0.50	0.08	0.05	0.02	0.37
Width	Replication	0.03	0.02	0.00	0.00	0.00
	Species	0.75**	0.12**	0.04**	0.03**	0.40**
	Error	0.03	0.00	0.00	0.00	0.00
	Total	0.26	0.04	0.01	0.01	0.10
Thickness	Replication	0.04	0.01	0.00	0.05	0.01
	Species	0.55**	0.05**	0.12**	0.01**	0.20**
	Error	0.02	0.00	0.00	0.00	0.00
	Total	0.19	0.02	0.04	0.00	0.05
Weight	Replication	0.00	0.05	0.00	0.00	0.00
	Species	0.76**	0.04*	0.00**	0.04**	0.13**
	Error	0.02	0.02	0.00	0.00	0.01
	Total	0.26	0.02	0.00	0.01	0.04
Volume	Replication	0.28	0.00	0.00	0.00	0.01
	Species	25.80**	0.03**	0.03**	0.00*	1.09**
	Error	0.26	0.00	0.00	0.00	0.00
	Total	8.45	0.01	0.01	0.00	0.28

** and * indicate significance level at 0.01 (P value <0.01) and 0.05 (P value <0.05)

(0.84) and weight (0.66) for seeds of all 97 species studied, hence width proved as dominating trait in PCA however other traits viz. thickness, length, volume and weight also played significant role. In general, PC 1 explained 73 % variation among data set however; PC 1 and PC 2 together

explained 87.44 % variation, whereas PC 1 and PC 3 together showed 80.93 % variation. The bi-plot analysis gave overall correlation among traits, those which showed positive correlation were near while those which stayed more than at 90° showed negative correlation. PCA along

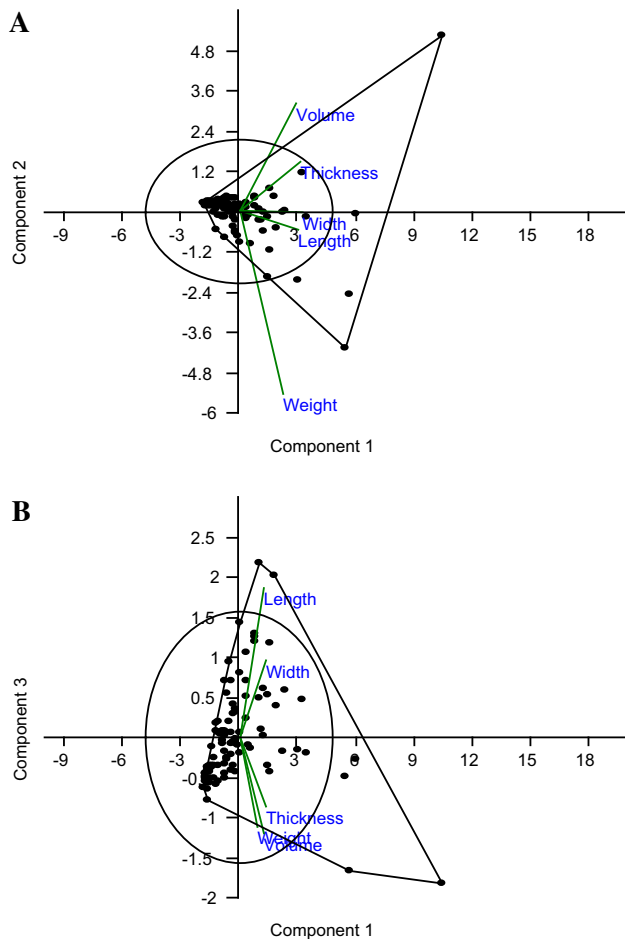


Fig. 1 Principal component analysis (Bi plot) for seed morphological study. **A** and **B** explain cumulative variance of 87.44 and 80.93 % respectively, **A** (component 1 and 2) is most significant PC among others

with bi-plot analysis showed only one significant PC which could be of high use in studying morphological variation (Fig. 1A, B; Table 5, Suppl. Fig. 2C.).

The cluster analysis on the basis of similarity-dissimilarity gave four main clusters which could be used in the group specific identification of plants. On the data set, most of the species were arranged in such a way that those in the common family mostly remained nearer to one another while those who were not related, remained away. The cluster analysis showed different clusters with seed size ranging from smallest to largest falling in four different clusters. Two species of Mimosaceae, *Acacia nilotica* (L.) Del. and *Acacia senegal* Willd., two species of Caesalpinaceae, *Bauhinia purpurea* L. and *Bauhinia racemosa* Lam., three species of Moraceae, viz. *Ficus benghalensis* L., *Ficus glomerata* Roxb. and *Ficus religiosa* L., along with two Papilionaceae members, *Dalbergia sissoo* Roxb. and *Dalbergia paniculata* Roxb. shared similar clusters each. First cluster was dominated by trees whose seed

volume was large, second was dominated by trees and shrubs where seed volume remained medium, third was by shrubs, herbs and undershrubs where seed volume remained small and fourth cluster showed association of herbs, undershrubs, shrubs and climbers with very small seeds (Fig. 2). It is clear from the analysis that, first cluster which is determined by trees, is mostly characterized by the highest seed volume with blackish brown colour, rough surface, ovate shape and circular hilum shape. Trees and shrubs are mostly occupied by cluster 2, showing pattern of brownish black seeds, with smooth surface, ovate shape and circular hilum shapes with medium seed volume. Cluster 3 was mostly in agreement with blackish brown colour, glabrous surface, ovate shape, circular hilum shape and small seeds for shrubs and herbs as well as single species of undershrub. Cluster 4, which showed smaller seeds, were mostly characterized by yellowish brown colour, glabrous surface, reniform, kidney shaped as well as ovate and circular hilum shapes, were mostly characterized by herbs followed by undershrubs, climbers and shrubs. Diversity among different clusters, helped in understanding species differentiation pattern along with proportions of seeds with their features. It also showed that size and trait based identification of species could be done using these clusters (Figs. 2, 3A–D).

In the different groups of plants, seed dispersal time varied as the seed dispersal was highest in June for trees with 41.3 % species among total 63 trees while it was December for herbs when 42.3 % species seeds were collected out of 26 species. In case of 5 shrubs studied, 4 months, December, January, March and April contributed 40 % of seed dispersal whereas in 3 climbers November was the best time to collect seeds as all species were in seeding during that time. Most of seeds were dispersed during the dry season whose main reason can be that the seeds might prepare themselves for germination in the next rainy season. Moreover, during May and June where average wind speed remained the highest in the forest, trees shaded 36.5 % seeds in May followed by 41.3 % in June however, only 7.7 and 3.3 % seeds were dispersed during those months for herbs. Shrubs and undershrubs did not contribute in the seed dispersal during this season. Interestingly, during May–June season average weight of seeds for trees was high with value of 0.51 g as compared to overall average weight value of 0.33 g however in case of herbs, average weight of herbs was little low with value of 0.05 g as compared to overall average value of 0.06 g.

Discussion

Significant variation (P value <0.05) was observed among the different plant groups in qualitative and quantitative traits studied for seeds which were manifested by width,

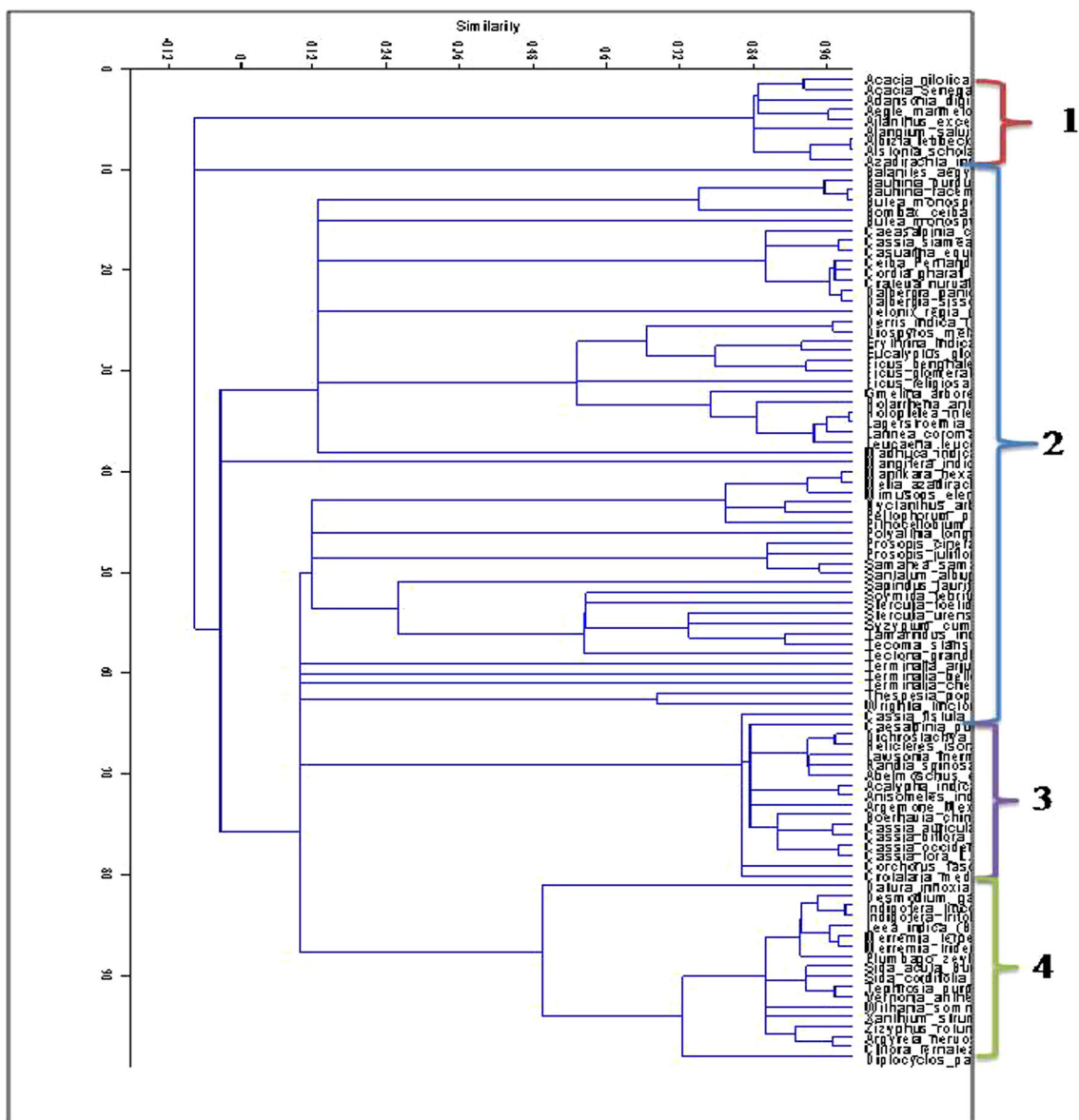


Fig. 2 Cluster analysis among 97 plants species on basis of seed morphology. Cluster 1 pertains 10 species, which is dominated by trees, cluster 2 contains 53 species which includes trees and shrubs. Cluster 3 and 4 which contain 18 and 17 species respectively, are characterized

by shrubs and herbs as well as by herbs and climbers. Diversity among different clusters in this study showed that *blackish brown colour* was in maximum proportion among all, while glabrous type was maximum in surface trait and dominant hilum shape was circular

thickness, length, weight and volume. The quantitative characters related to size of seeds contributed more in total variation however the qualitative characters also proved as one of the factors in variation. The seed morphological parameters with multivariate type analysis resulted that seed coat surface and seed weight are important in identification of variation among different sites [30]. It is in line of the results as weight proved as one of the factors governing variation. Furthermore, a study states that size and shape have larger discriminating power than colour and texture but other morphological features are also important

for complete seed identification [31]. It accounts same direction of current work as all morphological features were found useful in identification. Hence it is clear that, morphological findings are important tool in the taxonomic delimitation and the classification of number of genera and species, which can be used for making seed atlas of the particular area [31–40]. Even in monocotyledon plant study, priority was given on characters such as weight, colour and seed coat surface for taxonomic identification of 12 species and 11 genera in the West Bengal, India [41]. According to Lu et al. [42], each place has the unique

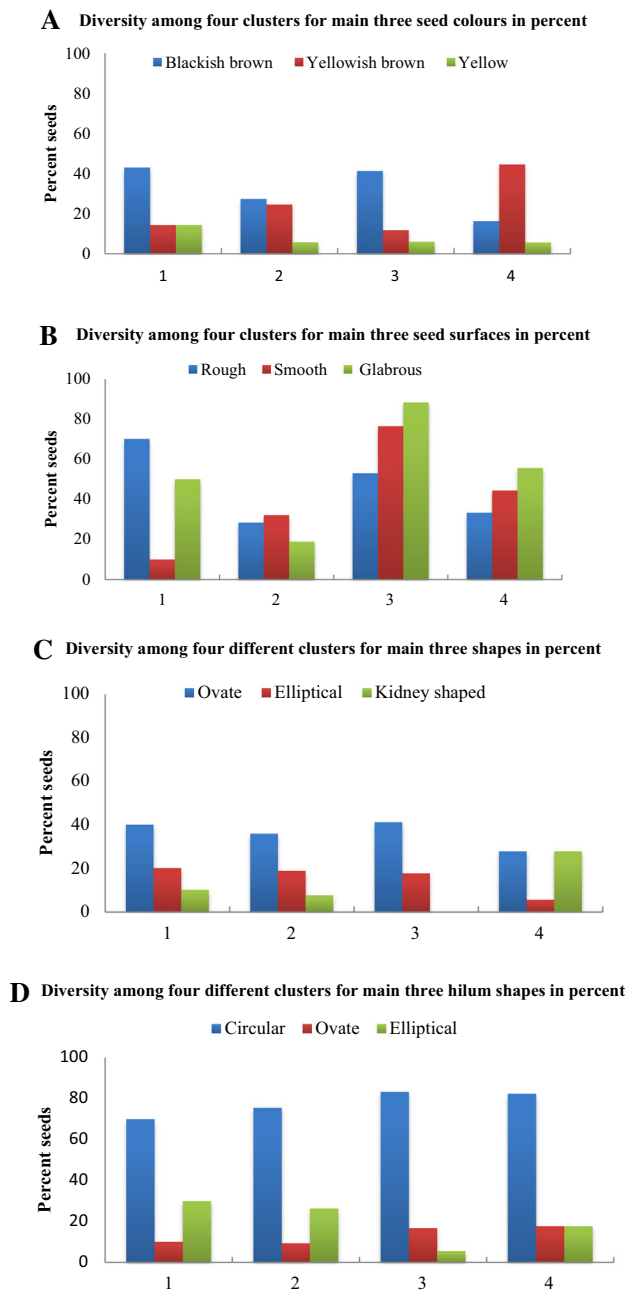


Fig. 3 Different proportions (in percent) of seeds for important qualitative traits in all clusters. **A** Diversity among four clusters for main three seed colours in percent. **B** Diversity among four clusters for main three seed surfaces in percent. **C** Diversity among four different clusters for main three shapes in percent. **D** Diversity among four different clusters for main three hilum shapes in percent

diversity of seeds although inter and intra specific variations are mostly found and the diversity found in Girnar Reserve Forest also showed variation. It can be said that large variation could be due to different adaptation mechanisms of different group of plants.

PCA and cluster analysis can be used in identification of seeds with desirable traits for morphological data of seed

set furthermore, it discriminates groups of related species [43, 44]. Characterization of herbs was done with large size and assisted dispersal [45] however in the current study, herbs showed blackish brown colour, glabrous surface, ovate shape, circular hilum shape and small seed size as a pattern. It can be due to variation in the number of samples studied, furthermore environment is also different in both the studies. Thus, classification of species by functional characteristics is important, and can be done easily if common groups are to be identified among different taxa. Moreover, the positive significant association between seed mass and seed size were observed in different studies [43, 45]. They account the same line of the work as there were significant correlations between most of the morphological traits studied.

The seed dispersal can be useful in understanding of population and community dynamics and the variation in seed weight can be an adaptive feature for any forest during the dispersal season, additionally there is a relationship between seed size, seed dispersal and other seed traits which insights that trait like seed mass provides important finding in trait based generalization [46, 47]. This is linked with this study as average weight value of the seed was little low for groups such as herbs, shrubs, undershrubs and climbers during the dispersal season but interestingly in trees, average weight was little high as compared to other months in the dispersal season. In subtropical broad leaved forest of China, peak in seed dispersal was observed during dry season [48] whereas similarly in the Girnar Reserve Forest of India, most of plant species showed the seed dispersal time during dry season of the area. Hence, same motivation inspires the results of this study. The seed dispersal occurs in the dry season in the Girnar Reserve Forest and hence, it may be recommended as the best time for the seed collection in the same area.

Conclusion

The present study demonstrated a large variation among seeds of five different botanical groups viz, trees, herbs, shrubs, undershrubs and climbers. Average value of tree seed volume was the highest among other groups followed by climbers, herbs and shrubs. Based on statistical analysis, the work insights inter and intra specific variation among different five groups of seeds with 97 plant species. Similarities and differences can be justified by the qualitative and the quantitative data obtained from the seeds collected from the Girnar Reserve Forest, Gujarat, India. This work is highly useful in trait specific identification of genus and species without going too deep, considering other plant characteristics. The seed atlas of the area can also be prepared from the available data. Although the work will

provide important details for those working on germination, viability, storage and other aspects related with seed science, physiology and different branches of plant sciences, more needs to be done in the proper direction for the concrete output.

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