Karyomorphology of a Critically Endangered Species Vateria indica L. (Dipterocarpaceae) from India

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Summary The genus *Vateria* L. (Dipterocarpaceae) with its three species namely, *V. indica*, *V. copallifera* and *V. macrocarpa* are distributed from India to Sri Lanka. The genus so far has not been screened for its cytological characters. In the present investigation, the mitotic chromosome number of 2n=22 and basic karyomorphology of *V. indica* were reported for the first time.

Keywords Chromosome number, Endemic, India, Karyotype, Vateria indica.

The family Dipterocarpaceae Blume comprises 16 genera and about 535 species worldwide (Mabberley 2017) of which five genera and 30 species are found in India (Sharma and Sanjappa 1993). The genus *Vateria* comprises three species namely, *V. indica* L., *V. copallifera* (Retz.) Alston and *V. macrocarpa* K.M.Gupta (plants of the world online.org, accessed on 2 April 2019) and all the three species occur in India (Sharma and Sanjappa 1993). *V. indica* and *V. macrocarpa* are endemic to India whereas *V. copallifera* is mainly found in Sri Lanka. *V. indica* and *V. macrocarpa* fall under critically endangered (CR) category and *V. copallifera* under endangered (EN) category of the IUCN.

Vateria indica is an economically important evergreen tree found growing in Sothern Western Ghats (Fig. 1a, b). The wood is used in plywood and veneer industry, for making tea chests, trunks, ammunition boxes, ceilings, partitions, flooring and other interior fittings; the bark and leaf juice are used in medicine; gum resin is known in the trade as Piney resin, White damar, or Dhupa in varnish industry and for making incense; the semisolid fat, known as Piney tallow, Malabar tallow or Dhupa fat obtained from the dried kernels of the seeds is used in the manufacture of candles and soap Sharma and Sanjappa (1993).

The phylogenetic assignment of Dipterocarpaceae among angiosperms has previously been problematic; however, Ashton (1982) supported their placement in the order Malvales has also done by APG IV (Chase 2016, Heckenhauer *et al.* 2017). The genus *Vateria* so far has not been studied from cytogenetic point of view. Here, we report for the first time a somatic chromosome num-

ber and karyotype of V. indica.

Materials and methods

Seeds of V. indica were collected from Agumbe-Sringeri road of Shimoga district in Karnataka state (13°51'24.9"N and 75°11'85.4"E). The voucher specimens of the species are deposited in Herbarium of Shivaji University (SUK). Mitosis was studied from the primary root tips of germinated seeds (Fig. 1c). Seeds were germinated between two moist blotting papers in glass Petri dish. Root tips of 6-10mm length were pretreated with 0.002 M 8-hydroxyquinoline for 10±2°C for 3-4h. The root tips were hydrolyzed with 1M hydrochloric acid at about 70-80°C for 1-2 min. Then the root tips were squashed in 2% propionic orcein. The wellspread chromosome plates were photographed with a LEICA DM 2000 fluorescence microscope attached with a camera. Five well spread somatic chromosomes plates were used for karyotype analysis. The nomenclature used for describing karyotype composition followed by Levan et al. (1964). The degree of karyotype asymmetry was determined using the categories of Stebbins (1971). Karyotype morphometric characters were evaluated by calculating haploid complement length together with intrachromosomal asymmetry index (A1) and interchromosomal asymmetry index (A2) as Zarco (1986).

Results and discussion

Vateria indica showed somatic chromosome number 2n=22 (Fig. 1d). The mean chromosome length (MCL) was $6.69\pm1.17\,\mu\text{m}$ and the total haploid genome length (TCL) was $73.59\,\mu\text{m}$. The shortest chromosome measured $2.08\pm1.06\,\mu\text{m}$ and the longest $5.38\pm2.41\,\mu\text{m}$ (Tables 1, 2). Figure 1e illustrates the karyogram. All

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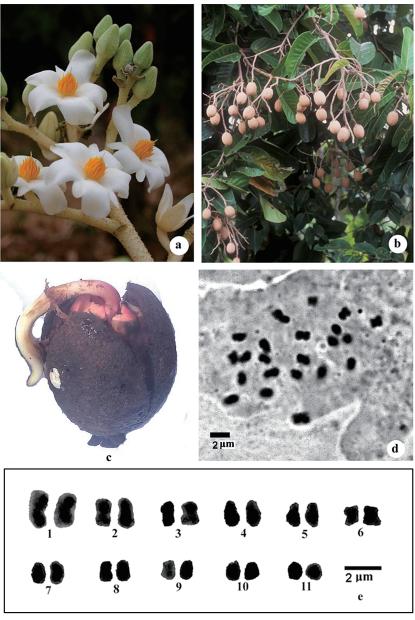


Fig. 1. *V. indica* a: flower, b: fruits, c: germinating seed, d: metaphase, e: karyogram.

Table 1.	Karyomorp	hological	lanalysis	of V.	indica.
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Chromosome pairs	Long arm l (µm)	Short arm s (µm)	Total length $c=l+s$ (μ m)	'd' value l–s (μm)	'r' value l/s	$i=s/c \times 100$	Centromeric position
1	5.38±2.41	3.62±1.81	9.00±4.06	1.76	1.54	40.10	m
2	4.69 ± 2.21	3.42 ± 1.18	8.11±3.33	1.27	1.34	42.93	m
3	4.22 ± 1.74	3.22 ± 1.17	7.44 ± 2.83	1.00	1.34	43.13	m
4	3.96±1.29	3.05 ± 1.35	7.01 ± 2.62	0.91	1.41	42.11	m
5	3.75 ± 1.60	2.93 ± 1.00	6.68 ± 2.60	0.82	1.24	44.75	m
6	3.59 ± 1.46	3.02 ± 1.19	6.61 ± 2.60	0.57	1.19	46.06	m
7	3.53 ± 1.46	2.66 ± 0.93	6.18±2.39	0.87	1.30	43.47	m
8	3.63 ± 1.78	2.43 ± 0.81	6.06 ± 2.47	1.20	1.48	41.03	m
9	3.51±1.34	2.55 ± 1.20	6.06 ± 2.47	0.96	1.44	41.54	m
10	3.20 ± 1.48	2.40 ± 1.00	5.60 ± 2.47	0.80	1.32	43.10	m
11	2.74±1.06	2.08 ± 1.06	4.82 ± 2.11	0.66	1.45	41.40	m

Table 2. Comparative account of chromosome number, chromosome range and TCL of six species of Dipterocarpaceae from India.

Species name	Chromosome number (2 <i>n</i>)	The range of chromosome length	TCL	References
Hopea odorata	14	2.4–3.6 µm	46.4 <i>µ</i> m	Roy and Jha 1965
Shorea robusta	14	1.4–2.8 µm	27.9 µm	Roy and Jha 1965
S. talura	14	1.75–2.5 μm	29.8 µm	Roy and Jha 1965
Dipterocarpus alatus	22	2.1–4.7 µm	66.8 µm	Roy and Jha 1965
Vatica grandiflora	22	1.6–3.4 µm	57.0µm	Roy and Jha 1965
Vateria indica	22	1.16–2.21 µm	35.92 µm	Present study

Table 3. The species of family Dipterocarpaceae occurring in India, gametic (n) and sporophytic counts (2n) and their geographical distribution.

Taxon	n, 2n	Authors	Geographical distribution
Dipterocarpus alatus Roxb. ex G.Don	20, 22	Tixier (1953), Roy and Jha (1965)	Andaman and Nicobar Islands.
D. costatus C.F.Gaertn.	20	Tixier (1960)	Andaman & Nicobar Islands and Tripura.
D. turbinatus C.F.Gaertn.	20	Tixier (1960)	Assam, Manipur, Meghalaya, Mizoram and Tripura.
Hopea odorata Roxb.	20–22, <i>n</i> =7, 14, 20	Tixier (1960), Roy and Jha (1965), Jong and Lethbridge (1967), Sarkar <i>et al.</i> (1982), Kaur <i>et al.</i> (1986)	Andaman and Nicobar Islands.
Shorea robusta Gaertn.	14	Roy and Jha (1965), Pal <i>et al.</i> (1993)	Andhra Pradesh, Assam, Bengal, Bihar, Himachal Pradesh, Madhya Pradesh, Orissa, Tripura, West Bengal.
S. roxburghii G.Don	14	Roy and Jha (1965)	Andhra Pradesh, Karnataka and Tamil Nadu.
Vateria indica L.*	n=10 22	Mehra (1976), Present communication	Karnataka, Kerala and Tamil Nadu.
Vatica chinensis L.	22	Mane et al. (2018)	Karnataka, Kerala and Tamil Nadu.

*Represents endemic taxa.

the 11 pairs of chromosomes were metacentric (m) with karyotype formula: 11m. Karyotype was symmetrical and fell into the 4A category of Stebbins (1971). The A_1 and A_2 asymmetry indices were found to be 0.25 and 0.19, respectively.

Roy and Jha (1965) reported the chromosome number for five species belonging to four genera of Dipterocarpaceae. They reported chromosome numbers 2n=14 in *Hopea odorata*, *Shorea robusta*, and *S. talura* while chromosome number 2n=22 was reported in *Dipterocarpus alatus* and *Vatica grandiflora*. Their chromosome range and TCL of all studied species were summarized in Table 2.

On the basis of chromosome number, members of family Dipterocarpaceae can be divided into two groups viz. 2n=22 (Dipterocarpus, Vatica and Vateria) and 2n=14 (Hopea and Shorea). The V. indica showed 2n=22 and it comes under Dipterocarpus and Vatica group. There are two base chromosome number in Dipterocarpaceae *i.e.*, x=7 and x=11. The group with x=11 has only diploid species but the group with x=7 base number possesses triploid and tetraploid species viz. Hopea jucunda 2n=21 and Shorea ovalis 2n=28 (Table 3). This polyploid nature of Hopea and Shorea also showed their derived nature as compared to Dipterocarpus, Vatica and Vateria.

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