

**MATERIAL
AND METHODS**

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Collection:

The endemic grass species selected for present studies were collected from Western Ghats during months between June to January. This study is mostly concentrated on the monotypic endemic grass genera. The list of endemic grass genera with localities of collection have been given in following Table:

Sr.no	Name of grass genus	locality
1.	<i>Danthonidium gammiei</i> (Bhide)	Ratnagiri, Goa, Vaibhavwadi, Dagipur.
2.	<i>Hubbardia heptaneuron</i> Bor	Tillari ghat
3.	<i>Indopoa paupercula</i> (Stapf)	Ratnagiri, Satara, Panhala, Mahabal- eshwar, Kas.
4.	<i>Pseudodicanthium serrafalcoides</i> (Cooke & Stapf)	Amba ghat, Phonda ghat, Amboli ghat, Kas, Those ghar.
5.	<i>Pogonachne racemosa</i> Bor	Amba ghat, Phonda ghat, Amboli ghat.
6.	<i>Trilobachne cookei</i> (Stapf)	Amba ghat, Malshes ghat.
7.	<i>Triplopogon ramosissimus</i> (Hack)	Sinhgad, Purandhar, Khandala.

Collected grasses were properly processed and dried by using blotting papers and newspapers. After proper processing and poisoning, the specimens were mounted on herbarium sheets and deposited in the Herbarium of Botany Department, Shivaji University, Kolhapur after confirmation of their identity. Wet Materials preserved in FAA for anatomical studies were stored in glass bottles at Laboratory conditions.

MORPHOLOGICAL STUDIES:

External morphology of selected grasses was studied using dissection and stereomicroscopes. Spikelets were dissected under Labomed Zoomer Stereomicroscope and dissected parts were measured under Peak Peakometer and illustrated to the scale. Spikelet photographs have been taken under Carl Zeiss Microscope using Coolpix 4500 digital camera with MDC lens.

ANATOMICAL STUDIES:

For anatomical studies of grasses, free hand section of leaf and culms were taken using a sharp blade. The section was mounted and stained in phloroglucinol stain (1% in 95% alcohol) by allowing 25% HCl to penetrate below coverslip. Lignified cells were turn pink. All the sections were observed under Carl Zeiss Jenaval compound microscope. Photographs were taken using Nikon Coolpix 4500 digital camera with MDC lens. Measurements were taken by using micrometry technique and diagrams were drawn with the help of photographs.

Epidermal studies:

The epidermis of leaves cannot be peeled off. Therefore, for studying epidermal structure following methods were used.

I) Scraping method:

In this method the leaf piece was placed on a glass slide in such way that the epidermis which is to be examined being face down. The leaf tissue were scraped away carefully with the help of sharp razor blade. Then turn the piece of epidermis upside down was used for microscopic examination.

II) Impression Technique Using Nail Polish:

The method of studying grass epidermis with the help of nail polish is proposed by Khidir W.Hilu and John L.Randall (1984).

The technique requires the application of a film of clear nail polish directly to the leaf surface using the brush provided with nail polish bottle. The film is allowed to dry for 1-2 hours. The use of air current speeds up the drying process but the film should be allowed about fifteen minutes to dry before air current can be used without distortions. Pair of fine forceps are used to peel off the film and placed it on a slide for examination. A cover slip is sometimes used to keep the film flattened; water or other liquids are not needed for viewing. In case of villous leaves, a thicker film of the nail polish is recommended. The direct application of nail polish to grass leaves is a quick, simple and inexpensive method to study all epidermal features.

CYTOLOGY:

Smear preparations were made from young flower buds fixed in Carnoy's fixative (ethanol: Acetic acid 3:1). Anthers were squashed in Propionic orcine and meiotically analyzed. Photographs were taken under Carl Zeiss Jenaval compound microscope.