

MATERIALS AND METHODS

South India which lies between North latitude 8° and 18° and East longitude 73° and 85° lies in the monsoon belt, and the region enjoys a tropical climate. The Western Ghats which extends from the Cape Comorin to the Tapti Valley, running parallel to the West Coast is exposed to the South West monsoon, and it receives over 250 cm of annual rainfall. The family Compositae is fairly well represented in this part of India. Most of the materials investigated during the present study were collected from different low and high altitude regions of the Western Ghats which lies in the Kerala and Tamil Nadu regions of South India such as Agasthia malai (950 m), Ponmudi (450-900 m), Bonacaud (450-850 m), Kulathupuzha (200 m), Aryankavu (200 m), Courtallum (450 m), Peermade (900-1200 m), Devicolam (1200-1600 m), Munnar (1050-1500 m), Santhenpara (1000 m), Kodaikanal (1800-2100 m), Sultans Battery (1200 m), Yercaud (1600 m) and places of the Nilgiris (1800-2500 m) such as Pykara, Glenmorgan, Coonoor and Ootacamund. Collection trips to the wild forest ranges of these places are beset with many difficulties including lack of means of conveyance to reach the interior of the forests and danger from wild animals. In spite of this, several collection trips were conducted to these regions, and a number of species collected. Materials from a few coastal regions and plains such as Trivandrum, Quilon, Ranni, Kottayam, Ernakulam, Alwaye, Calicut (Kerala State); Cape Comerin, Coimbatore (Tamil Nadu),

Bangalore (Karnataka State) and Hyderabad (Andhra Pradesh) were also included in the study. Wherever possible, attempt was made to collect materials of the same taxon from different localities of its incidence in this region. Most of the species covered by the study are indigenous to South India, while a few were exotic species which have been known under cultivation here as popular ornamental garden plants.

Chromosome studies were made from PMCs at meiosis and/or root tips. For meiotic studies materials were mostly taken from plants growing in the wild state, and this was supplemented with materials procured from plants maintained under green house conditions in the University Botanical Garden. Flower buds were fixed in Carnoy's fluid (3:1 absolute alcohol and glacial acetic acid). Somatic chromosome studies from root tip cells were possible only in cases of plants which thrived under green house conditions. The potted plants kept in the green house were watered well and exposed to bright sunlight for two hours before collecting root tips from them. Root tips were collected between 10 and 11.30 AM and pretreated with 0.002 M solution of 8-hydroxyquinoline (Tjio and Levan, 1950) for 2 hr at 4°C. The pretreated roots were washed well in tap water and fixed in Carnoy's fluid.

For staining meiotic chromosomes, acetocarmine (1%) and Haematoxylin (2%) were tried. Since the latter was found to yield better results the haematoxylin staining procedure (Henderson and Lu, 1968) was followed throughout. For studying

meiosis, one or two flower buds were smeared in a drop of haematoxylin and a cover glass put on after removing the debris. The slide is then gently heated and pressed uniformly under the folds of a blotting paper. For mitotic chromosome studies, acetocarmine, fuelgen and haematoxylin staining techniques were tried of which acetocarmine and fuelgen techniques did not yield satisfactory preparations in quite a few of the species, and hence haematoxylin technique was followed throughout. Root tips, after 24 hr fixation in Carnoy's fluid were washed in distilled water and hydrolysed in 1 N HCl for 10 minutes at 60°C. The hydrolysed root tips were thoroughly washed in distilled water and one or two meristematic tips were squashed on a slide in a drop of haematoxylin. After putting on a coverglass, the slide is gently heated and pressed uniformly in the folds of a blotting paper. Observations were made from temporary slides, and the slides are then made permanent following Mc Clinlock's (1929) method.

Photomicrographs of PMCs and root tip cells were taken at magnifications ranging from x 750-850. For preparing photographic plates, the photographs of chromosome preparations were taken at convenient magnifications ranging from x 1000 - 1500, and then the plates were reduced to desired size. In the case of preparations which lacked sufficient clarity, explanatory figures are provided. For this, chromosomes were outlined in Indian ink on enlarged photographic prints, and the photographic image then bleached out with iodine solution. The details were

filled in with Indian ink, and the figures recopied photographically and reduced to convenient magnifications. Explanatory figures were given the same number as that of the photomicrographs with suffix 'a'.

Karyomorphological analysis was made based on average measurements from 8 - 10 cells at metaphase in each species. Measurements of chromosomes were taken from photographs magnified 3000 times. Values of measurements such as Long arm (L), Short arm (S) and total lengths of chromosomes are tabulated, and arm ratios (r) for each of the homologous pairs calculated (L/S). Classification of chromosomes were made following the system proposed by Levan et al (1964) in which chromosomes with absolute median position of centromere ($r=1$) are designated as M-types; those with arm ratios between 1 and 1.7 as m-type; arm ratios between 1.7 and 3.0 as sm-types; arm ratios between 3 and 7 as st-type and those with arm ratios exceeding 7 as t-type. Relative chromosome length is abbreviated as RCL; Total chromosome length as TCL and Average chromosome length as ACL. Chromosomes with satellites are referred to as Sat-chromosomes. Categorisation of karyotype assymetry has been made according to the method proposed by Stebbins (1958). Idiograms are provided for all the species whose karyomorphology was analysed.

Pollen fertility was determined by smearing mature anthers in a 1:1 mixture of 1 per cent acetocarmine and glycerine,

and the slides kept for half an hour. Pollen fertility was estimated by scoring the stained and unstained pollen grains, the stained ones being considered as fertile and the unstained as sterile. Percentage of sterility was assessed from the data on about 1000 pollen grains in each species obtained from counts of several slides at the rate of 15 random fields from each slide.

Herbarium sheets of the species studied were prepared, and verification of species determinations done at the regional office of the Botanical Survey of India at Coimbatore. Herbarium specimens are deposited in the Central Herbarium, Kerala University.

For arrangement of the tribes and genera in the results recorded here, the classification of Bentham and Hooker (1873) was followed.