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Morpho-palynological Studies on the Climbing Fern Lygodium japonicum

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Abstract: The Japanese climbing fern *Lygodium japonicum* (Thunb) Swartz was collected from Aalra Muzaffarabad, Azad Kashmir. The leaflets venation pattern was dichotomous and open. Anatomy of the rachis shows a central protostele with exarch to mesarch xylem surrounded by phloem. The protostele is surrounded by endodermis and pericyle. The phloem is protruded into the bays formed by a three angled protoxylem of protostele. The sporangia were globular shaped and have size 600-650 μm wide and 500-550 μm height. The number of spores per sporangium calculated was 256. Spores have 64-80 μm equatorial diameter, isospores, anisomorphic, contour subtriangular, trilete, exine 3-4 μm thick. Distal surface of the spores was warty at equatorial lines.

Key words: Lygodium japonicum, rachis, sporangium, Schizaeceae, protostele

INTRODUCTION

The genus Lygodium is placed in the Schizaeceae, a small primitive family that also includes Actinostachys, Schizaea, Anemia and Mohria[1,2]. Recent molecular research on the phylogeny of ferns found Lygodium, Actinostachys and Anemia to have more intergeneric distance than occurs within most fern families^[3]. These genera were nevertheless called sister genera and left in the Schizaeceae. The molecular data and the antiquity of the Schizaeaceae suggest that the group diverged at an earlier time than other groups of ferns^[4]. The genus Lygodium may contain as many as 40 species^[5]. All but a few occur in warm temperate to tropical areas [6]. In Pakistan Lygodium japonicum has been recorded from Kaghan^[7], Mansehra, Domail and Mirpur^[8]. It seems to be very odd about the distribution in Mirpur due to climatic barrier. Morphological and anatomical specializations of the climbing leaves of Lygodium japonicum have been investigated by earlier workers[9].

The fern flora of Pakistan is not completely known. Since the list published by Stewart^[8], the only reference can be made of Sheikh^[7] who described certain fern species from Kaghan Valley. Although morphological data for *Lygodium japonicum* are available^[7], the anatomy, sporangial structure and spore morphology have not been investigated. The present study is an effort to add some information about the rare fern *Lygodium japonicum* found in Pakistan and Azad Kashmir.

MATERIALS AND METHODS

The specimens were collected fresh along with rhizome. The material was preserved in acetic alcohol (1:3) till further use. The clearing of leaflets/fronds was made according to the technique used by Bhutta and Sadiq[10]. The material was kept in aqueous solution of chloral hydrate (500 g of chloral hydrate was dissolved in 100 mL of distilled water) for 5 h. The fronds were washed thoroughly with distilled water and chlorinated as suggested by Bhutta[11]. The fronds were then stained with alcoholic basic fuchsin (saturated solution of basic fuchsin in ethanol was filtered in a brown coloured glass bottle. Ammonia solution (33%) was added drop by drop to alcoholic basic fuchsin till the red colour changes to pale yellow. Filtered and was used to stain the plant fronds, sections, sporangium and spores). The stained fronds were mounted in Euperal on a glass plate (size 8x6 inches). The sporangia were detached from the fertile fronds with the help of needle under binocular microscope and then mounted in glycerin jelly on a glass slide. For spore investigations, the sori were crushed with glass rod and passed through 1.5 inch dia funnel placed in a centrifuge tube and plucked with loosely placed glass wool. The sieved spores were treated with 5% KOH solution to remove the oils and humic acid. After chlorination the spores were mounted in 2% glycerin jelly already stained with 2% saffranin. The mounted slides were sealed using synthetic enamel on a ringing disc. For anatomical studies the transverse sections of the rachis were made. After staining with 2% saffranin solution the sections were mounted in Euperal for microscopic studies. Microphotographs were taken on Photomicroscope Kyowa Medilux-12.

RESULTS AND DISCUSSION

Lygodium japonicum has slender and dark brown rhizomes. Frond climbing, twining, glabrous and of indeterminate growth, often reach up to 100 feet; main rachis wiry, stem like and adapted for twining. Leafy branches off main rachis (constituting the pinnae) compound, triangular in overall outline, 4-8 inches long and about as wide. Leaflets (pinnules) lobed, stalked, with terminal lobes often dissected (pinnatifid), basal lobes irregularly lobed or dissected; leaf-blade tissue pubescent below with short, curved hairs. Fertile leaflets contracted in shape, with two rows of sporangia along the leaf margin, which is enrolled to partially cover the sporangia (Fig. 1). The dichotomous type of venation remains basically open (Fig. 2) but sporangia are produced individually and protected by a specially produced flap of the lamina. These morphological characters are in agreement with earlier workers [7,8].

The rachis of Lygodium japonicum is three angled, protostele, solid, covered by endodermis and pericyle. The phloem is protruded into bays formed by a three angled protoxylem and protostele. The cortical region is parenchymatous (Fig. 3).

The sporangia of Lygodium japonicum are 600-650 µm wide, 500-550 µm high and are globular in cross section. Sporangia are seated near each vein and are protected by the curled margin. Stalk of the sporangium is



Fig. 1: A portion of fertile leaflet of Lygodium japonicum



Fig. 2: A leaflet of Lygodium japonicum showing open dichotomous venation

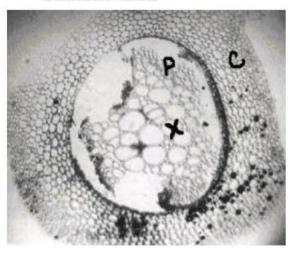


Fig. 3: Transverse section of rachis of Lygodium japonicum. P, Phloem; X, xylem and C, cortex

not detectable but the sporangium is placed horizontally (Fig. 4) where annulus is clearly shown and having 65 µm thickness in cross section. The mature sporangium has spores countable up to 256. Spores have 64-80 µm equatorial diameter, isospores, contour subtriangular, anisomorphic, trilete, arms of laesurea almost to the length of the spore radius, suture fine straight, commissure open, extremities of arms slightly arcuate, proximal surface slightly reticulate, distal surface heavily warty at equatorial line and variable in dimensions (Fig. 5). At certain points they are oriented in reticulate pattern.

Exosporium is heavily thickened, exine 3-4 µm thick. Isospores of Lygodium japonicum have the same relationship as has been recorded in Erdtman and Sorsa^[12]. The foregoing discussion suggest that

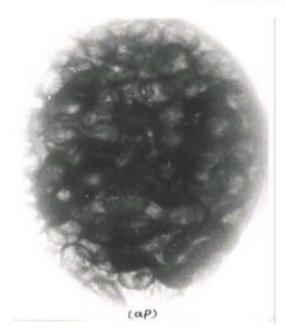


Fig. 4: Sporangium of Lygodium japonicum filled with spores. ap, aperture



Fig. 5: Trilete spore of Lygodium japonicum

Lygodium japonicum is a primitive fern having protostele stem, dichotomous branched venation of open type as these characters are quite primitive^[13]. To conclude all, the anatomy of the rachis, morphology of sporangia, placement of annulus and morphology of the spores are additional characters to differentiate

Lygodium japonicum from other species in the genus. It is recommended that steler structure in the rhizome up to pinnules be studied to help in the classification of the ferns of Pakistan.

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