Presence of air cavities in pods of an endemic legume *Crotalaria pusilla* DC. (Leguminosae, Papilionoideae)

The legume lineage evolved approximately 60 Mya in the region to the north of the Tethys Sea, a shallow stretch of salt water that separated the two main groups of land masses[1]. Since then legumes have adopted various mechanisms for dispersal and colonization, among which one of the probable methods is the diaspore (a dispersal unit consisting of a seed with additional tissues that assist dispersal). The papilionoid crown node is dated 58.6 Mya with further divergence into genistoid and dalbergoid, the former dated at 56.4 Mya (ref. 1). Since their divergence, both the lineages have adopted radically different approaches to life from their common ancestors. One of the approaches is seed dispersal, which plays a central role in ecology dynamics, thereby understanding and predicting the population and community change[2]. Understanding the convergent evolution of seed dispersal is a central question in evolutionary ecology, because dispersal traits are linked to plant survival strategies and are a reflection of the reproductive assurance theory[3].

Evolutionary success of a species predominantly depends on effective seed dispersal mechanisms. Most of the fruits of Leguminosae dehisce by the explosive mechanism. However, dispersal by diaspore is restricted to certain species. It can be a seed (with elaiosome), or together with fruit, or the whole plant (known as tumbleweed)[4]. In flowering plants, diaspores have been categorized into different types, viz. dust diasporas[5] (e.g. seeds of *Sedum*, *Campanula* and *Rhododendron* whose individual seed weight ranges between 0.003 and 0.004 mg), balloons[6] (e.g. pods of *Crotalaria*, *Cicer*, and inflated calyx of *Trifolium*, *Physalis*, and epidermal cells in the seeds of *Cuscuta*), plumed diasporas[7] (e.g. in *Aeschynanthus* and *Agalmyla*) and winged diaspores[8] (e.g. seeds of *Ailanthus altissima* and *Zanonia macrocarpa*). Here we report the presence of air cavities in the pod pericarp of *Crotalaria pusilla* (Crotalarieae, Papilionoideae, Leguminosae), a species endemic to the Western Ghats region of Maharashtra, India.

Crotalaria is the largest genus in the tribe Crotalariaeae in Genistoid clade of Papilionaceous legumes and a recently diverged clade (23–30 Mya) from its sister lineage *Bolusia*[9]. India hosts the maximum number of simple-leaved *Crotalaria* species (79 spp.) with a disjunct distribution[10–13] (predominantly restricted to the Western Ghats and North East India).

Fruits of almost all the species studied in *Crotalaria* dehisce by the explosive mechanism to disperse seeds. Dispersal of seeds is assisted by the sclerenchymatous zone. The angular arrangement of the sclereids in opposite directions in the two lobes of the narrow fruit (e.g. *C. pallida*) induces torsion force and enables spiralization soon after explosion of the pods. This enables effective dispersal of seeds in all the directions. During desiccation of the fruit, this fibre topography induces torsion forces that lead to the opening of the pods at the ventral and dorsal sutures where the dehiscence tissues are situated. When ripe, the pods twist and explosively rupture along both sutures ejecting the seeds. This dehiscence is said to be caused by differential stress within the pericarp, arising from the drying out and shrinkage of layers of thick-walled cells and fibres, which are arranged in opposing orientation[14,15]. The narrower and longer the pod, more the helical pitch and more effective is the dispersal.

Spiralling, twisting and subsequently releasing of the seeds in an explosive manner help them to occupy new sites which are relatively closer to the parent plant. This mechanism, however, restricts colonizing new habitats for the genus. Hence, the process of evolving new traits like diaspore would help some species occupy new habitats. Further, survival and mortality will be determined by the plant fate in space and time.

We studied the pod pericarp anatomy of 81 species of Indian *Crotalaria* (95.29% sampling, unpublished data). Earlier work on pod anatomy of South African *Crotalaria* was carried out by Le Roux *et al*.[16] showing that there is a uniformity in the pericarp anatomy of all the species under study with the three basic layers, viz. the exocarp (parenchymatous, single-layered), mesocarp (collenchymatous, multi-layered) and endocarp (sclerenchymatous, multi-layered), the cells of which are arranged in different orientations, thus enabling spiralization. *Crotalaria* corresponds to the type-I category where one, two or three zones of various numbers of cell layers of normal fibres and trichomes are found, which are occasionally associated with the endocarp. Anatomical as well as physiological functions of the pod pericarp on the one hand resemble those of leaves and on the other hand, similarities exist to storage organs during the initial developmental stages. This double function is closely related to the maintenance of a continuous ontogenetic development of the embryo and contributes to the survival of the species[17].

There is yet another category of pods which do not dehisce at all; instead the entire fruit is dispersed as diaspore assisted by wind and gravity (e.g. *Crotalaria nana*, *C. pusilla* and *C. filipes*). Among all the species dispersed by diaspores in the genus *Crotalaria*, *C. pusilla* showed unique anatomical features in the pod pericarp, which is speculated to enhance its dispersal capacity and thereby increasing the chances of successful colonization in a distant place and habitat. Similar work done by Dayanandan *et al*.[18] on air passages in the stem of *Gloriosa superba* and *Iphigenia indica* points out to their function in gaseous exchange. We negate this probable function in *C. pusilla* as the air cavities develop only when the pod is mature and during the developmental course, the entire cells of the pericarp are parenchymatous and collenchymatous which allows for gaseous exchange. Anatomically the pericarp of *C. pusilla* is similar to the other species dispersing as diaspores in having 3–4 layers of mesocarp and 4–5 layers of endocarp. The endocarp is further distinguished into two layers: (a) adjacent to the mesocarp being arranged in a perpendicular orientation, and (b) the rest of the layers inclined at 20°–25° from the mesocarp cells (Figure 1). These differential orientations help in negating the torsion pressure and thereby resisting dehiscence. This opposing arrangement of cells of the endocarp region acts as a counter pressure layer, which negates the explosion mechanism. *C. pusilla* pericarp dif-

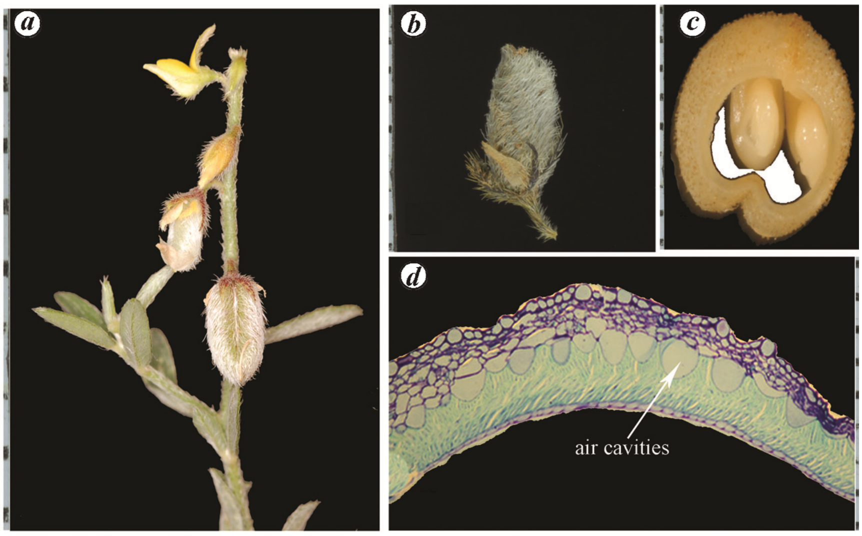

Figure 1. *Crotalaria pusilla*.: ***a***, Twig showing leaf, flower and pods; ***b***, Enlarged view of a pod; ***c***, T.S. of pod showing locule and a seed attached on the marginal placenta. ***d***, Pericarp anatomy showing three layers, viz. exocarp, mesocarp and endocarp. Arrow indicates enlarged air cavities at the juncture of the mesocarp and endocarp layers.

fers from other disaspores as they have enlarged air cavities at the junction of the mesocarp and endocarp layers. These cavities are swollen and ovoid, and are formed by retreating of walls of the mesocarp and endocarp cells, and develop only when the pod reaches maturity (Figure 1).

These cavities diminish the specific weight of the diaspore and increase the surface area. The average single pod weight (dry) of *C. pusilla* ranges between 4 and 4.5 mg, and average single seed weight ranges between 0.3 and 0.4 mg, the lowest in the genus. These cavities in *C. pusilla* are restricted to the intersection zone between the mesocarp and the endocarp. The present discovery of air cavities in the pod pericarp of *C. pusilla* (species endemic to India) suggests their probable evolutionary strategy in pod dispersal as a diaspore. The development of cavities in *C. pusilla* has thrown light on the dispersal strategies and attempts of the plant to colonize and create a niche farther from its parents, which eventually may prove to be one of its successful survival strategies.

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