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Population status and floral biology of *Trichopus zeylanicus* ssp. *travancoricus* Burkill ex K. Narayanan (Dioscoreaceae), an important ethnomedicinal plant of the southern Western Ghats, India

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Abstract: *Trichopus zeylanicus* ssp. *travancoricus* is locally known in Tamil Nadu as Arogyapachai, meaning a plant that provides good health and vitality. The unripe fruits of the plant are highly rejuvenating and are used by the Kani Tribe to ameliorate fatigue. This subspecies is also known for its pharmacological and medicinal utility and possesses choleretic, aphrodisiac, hepatoprotective, mast cell stabilizing, adaptogenic, and cardioprotective properties. A study was attempted to understand the population status and floral biology of *T. zeylanicus* ssp. *travancoricus* in its natural habitat. The population of this herb was found to be severely fragmented and settled in shaded banks of streams and rivulets in Agasthyamalai Hills. Being a rhizomatous herb, its flowers are concealed by its broad leaves, thus preventing exposure to pollinators. The number of pollinating agents in the flowers was reported as meager. The fruits are capsules with two to six seeds, each having a hard seed coat with a projected endosperm. Therefore, the conventional propagation of the subspecies is not so successful due to its poor seed set and prolonged maturation time. In addition, the fruits are damaged by rodents, livestock, and collection for medicinal usage, which severely affect the population status of the plant. Sustainable use of this wonder herb is important for its conservation in its natural habitat.

Keywords: Kani Tribe, *Trichopus sempervirens*, Indian Ginseng, Agasthyamalai Hills, pollination, conservation.

A number of scientists worked on the genus *Trichopus* Gaertn. and transferred its position from one family to another. The genus was treated under Aristolochiaceae by Lindley (1832) and Thwaites (1861), under Dioscoreales by Bentham & Hooker (1883), and under the monogeneric Trichopodaceae by Hutchinson (1934) and Dahlgren et al. (1985). Takhtajan (1980) conceded the status of a subfamily to Trichopodaceae under Dioscoreaceae but later accepted the view of Hutchinson (1934). In the recent past, Rao (1955), Ayensu (1966), Ramachandran (1968), and Kale & Pai (1979) studied the different botanical aspects of *Trichopus* with respect to its systematic position and supported its separation from Dioscoreaceae. The genus, however, is believed to be a connecting link between Dioscoreaceae and Taccaceae, combining the characters of both as well as those of Stemonaceae and Trilliaceae of the order Dioscoreales (Dahlgren

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et al. 1985). The members of Dioscoreaceae are usually rhizomatous climbing lianas or vines with small vascular bundles arranged in two circles with larger cauline bundles alternating with seeds. *Trichopus* are climbing or prostrate herbs with the vascular bundles arranged in an arc. The detailed systematic account provided by Caddick et al. (2002) on the relationship between *Stenomeris* and *Trichopus* remained unclear. After several confirmative studies conducted by the Angiosperm Phylogenetic Group (APG), *Trichopus* was placed under the individual family Trichopodaceae under the order Dioscoreals (Angiosperm Phylogeny Group 1998). In APG II, however, Trichopodaceae was merged with Dioscoreaceae (Angiosperm Phylogeny Group 2003), which was followed in APG III (Angiosperm Phylogeny Group 2009) and APG IV (Angiosperm Phylogeny Group 2016).

The genus *Trichopus* was reported with two herbaceous species, namely *T. sempervirens* (H.Perrier) Caddick & Wilkin and *T. zeylanicus* Gaertn. Both the species show restricted distribution — *T. sempervirens* is reported as endemic to Madagascar whereas *T. zeylanicus* is distributed in peninsular India, Sri Lanka, the Malay Peninsula, Singapore, and Thailand. *Trichopus zeylanicus* usually grows in lowland sandy areas of streams but in low-lying forests in the Malay Peninsula. In these habitats, it is vulnerable to flooding but similar conditions (sandy areas of streams) rule where it grows in the higher altitudes of the southern Western Ghats. *Trichopus zeylanicus* was reported with three subspecies, namely *T. zeylanicus* ssp. *angustifolius*, *T. zeylanicus* ssp. *Travancoricus*, and *T.* ssp. *zeylanicus*.

After several periodical field visits conducted in different forest areas of Agasthyamalai and observations of herbarium specimens in different repositories, it was confirmed that *T. zeylanicus* ssp. *travancoricus* can be considered as a valid subspecies. All the three subspecies were distinguished based on the number of nerves in their leaves. *Trichopus zeylanicus* ssp. *travancoricus*, with five to seven nerves in its leaves, is distributed in southern India and the Malay Peninsula. Both the triple-nerved subspecies, *T. zeylanicus* ssp. *angustifolius* and *T. zeylanicus* ssp. *zeylanicus*, are reported as endemic to Sri Lanka. The specimens collected in Sri Lanka were undoubtedly distinguished from those in peninsular India by their deltoid or linear-lanceolate erect leaves with 3mm long tepals. According to Burkill (1951), consent has to be given with distinguishing name to the Indian species as *T. zeylanicus* ssp. *travancoricus* and Indo-Malaysian specimens. The specimen from Sri Lanka possesses great variation, which is absent in the Indo-Malaysian subspecies.

Trichopus zeylanicus Gaertn ssp. *travancoricus* Burkill ex K. Narayanan is locally known in Tamil Nadu and Kerala as Arogyapacha or Arogyapachai, meaning leaves that give health and vitality. It is reported as one of the important ethnomedicinal plants that grows near wet banks of streams and rivulets in dense forests. The Kani Tribe contributed in bringing the multifarious uses of this wild plant to today's medicinal world. The tribe also claims that to remain healthy, agile, young, and resistant to various infectious diseases, one should consume the fresh fruits of the plant regularly (Pushpangandan et al. 1988). Its seeds are reported to be rich in saponins and a preparation from its leaves, stems, and seeds is used as health tonic (Sharma et al. 1989). The plant is treated as Indian Ginseng because of its similarity to *Panax ginseng* in pharmacological properties (Anilkumar et al. 2002). *Trichopus zeylanicus* also possesses choleretic, aphrodisiac, hepatoprotective, mast cell stabilizing (Subramoniam et al. 1997, 1998, 1999), adaptogenic (Singh et al. 2005), and cardioprotective (Velavan et al. 2009) properties. The whole plant has the potential for anxiolytic and antidepressant, hepatoprotective, immunomodulatory, and anti-ulcer activities (; Rishikesh & Sambathkumar 2016).

In India, this subspecies was reported as endemic to the southern Western Ghats with a restricted distribution in Agasthyamalai Biosphere Reserve, one of the five important centres of plant diversity in India. The area is also one of the 24 microcenters of endemism and a super hotspot of biodiversity (Ramasubbu et al. 2016). Eleven forest types were recognized from this area with 448 endemic species of angiosperms (TNFD 2016). The reserve harbours about 2,000 species of flowering plants, including about 150 strict endemics. It is also unique in having a genetic reservoir of cultivated plants.

Materials and Methods

The distribution status of *T. zeylanicus* ssp. *travancoricus* was analysed during extensive field trips conducted to forest areas of Kalakkad-Mundanthurai Tiger Reserve (KMTR) of Tamil Nadu and Rosemala and Sornagiri of Kerala. The extent of occurrence, the area of occupancy, and the number of mature individuals were recorded based on IUCN guidelines (IUCN 2012). The information gathered from the field visits was compared with that in published literature. To assess the population status of the subspecies, about 2x2 m^2 plots were laid down in the distribution areas. The

number of individuals per square metre was calculated randomly in different parts of the three study sites. The periodical assessment of the number of individuals per square kilometre was calculated for about three years (2014–2016).

Phenology is the study of the functional rhythm of plants in relation to seasonal and climatic factors. Phenological studies are important for understanding the responses of plants to various biotic and abiotic factors. In the present study, the selected individuals of *T. zeylanicus* ssp. *travancoricus* was marked in the natural habitat of the different study sites and observed during regular field visits. The flowering phenology was observed on a day-to-day basis, which included flower initiation, development and maturation, anthesis, anther dehiscence, and flowering and fruiting period. The phenophase events were recorded as per the method suggested by Dafni et al. (2005). The percentage of natural fruit set and flower-fruit ratio, if any, was also calculated for all the study sites.

***Trichopus zeylanicus* ssp. *travancoricus* Burkill ex K. Narayanan**

Nair, Kew Bull. 48:127–128, 1993; Mohanan & Sivadasan, Fl. Agasthyamala 2002; Nayar et al. Fl. Pl. Kerala – A Handbook 2006; Nair et al. Fl. Pl. Western Ghats India 2014. *Trichopus zeylanicus* ssp. *travancoricus* (Bedd.) Burkill Sivarajan et al. Kew Bull. 45: 353–359; Nair, Kew Bull. 48:127–128, 1993; Mohanan & Henry, Fl Thiruvananthapuram 1994. *Trichopus zeylanicus* Gaertn. Rama Rao, Fl. Pl. Travancore 1914; Gamble, Fl. Pres. Madras 1928–1936; Sivarajan et al. Kew Bull. 45: 353–359; *Trichopodium travancoricum* Bedd. Sivarajan et al. Kew Bull. 45: 353–359.

Small, rhizomatous, glabrous, perennial herbs; rhizome 1–4 cm long, with slightly ascending tips, covered with scales, older parts loose scales and dying off progressively; scales closely set, chaffy, lanceolate, acute or acuminate, 5–8 mm long; roots many, fibrous, stem wiry, 3–10 from the axils of scales, petiole-like, each terminating in a solitary, petioled leaf, terete, longitudinally striate or faintly ribbed, 7–8 cm long, purplish; dorsally grooved, 4–20 cm long; lamina highly variable in size, shape and venation, broadly ovate, deeply cordate, truncate or cuneate at base, wavy at margin, acute at apex, 5–7 nerved from base with another pair of nerves running along the whole leaf margin; fertile branch terminal on the stem, but pushed towards one side by the petiole which usurps the line of the stem, short, sessile, densely clothed with scaly, lanceolate bracts similar to the scales on the rhizome; leaves and flowers of this plant shine like grey-black stone; flowers 1–11 on each plant, hermaphrodite; pedicel filiform, 2–8 cm long; perianth of six subequal lobes in two series, campanulate, 3–10 mm long, constricted above the gynostemium, lobes lanceolate, apiculate, broader and saccate at base, keeled on the back; stamens six, filaments short, flat, united with the top of the ovary and base of style, anthers two-celled, connective broad, projecting and meeting by their edges making a roof over a chamber; ovary inferior, three-loculed with two superposed ovules in each chamber; style short, stout; stigma three, each two-lobed, lobes fleshy, erect at first and reflexed later, always included in the staminal chamber; fruit capsular, three-lobed, three-winged, ellipsoid or trapezoid, 1–5cm length x 1 cm width, opening by irregular rupture of the fruit wall; seeds 2–6, oblong or ellipsoid, covered with dense brownish tomentum, ventrally flat and ruminate, 4x2 mm.

Vernacular name: Tamil: Arochyappachai, Nilakottai; Malayalam: Arogyappacha.

Materials examined

***Trichopus zeylanicus* ssp. *travancoricus*:** MH 166487, 10.x.1992, Ingikuzhi, coll. R. Gopalan; MH 166488, 10.x.1992, Ingikuzhi, coll. R. Gopalan; MH 166089, 28.iii.1991, Kannikatti Rain Forest, coll. R. Gopalan; MH 166090, 28.iii.1991, Kannikatti Rain Forest, coll. R. Gopalan; MH 107169, 23.iii.1979, Vallachithodu, Lower Kodayar, coll. A.H. Henry; MH 107170, 23.iii.1979, Vallachithodu, Lower Kodayar, coll. A.H. Henry; MH 51676, 1901, Kannikatti; MH 51677, 1901 , Kannikatti; MH 51678, 1901 , Kannikatti; 1873, Tirunelvelli hills, coll. R.A. Beddome; GUH 152, 22.viii.2015, Rosemala, coll. G. Manikandan & R. Ramasubbu; GUH 298, 19.ii.2016, Sornagiri, coll. F. Irudhyaraj & R. Ramasubbu; GUH 322, 16.ix.2016, Kannikatti, coll. N. Sasi Kala & R. Ramasubbu.

***Trichopus zeylanicus*:** K00029181, 14.xi.1891, Flauggi, coll. H.N. Ridley; K001140902, 20.i.1928; K001140903, 00.xi.1893; K001140904, 16.i.2003, Induruwa Forest, coll. M.W. Chase & M. Fay; K001140905, 28.i.1972, Hallawakellae Forest, coll. K. Balakrishnan; K001140910, 12.i.1972, Badagama Forest Reserve, coll. M. Jayasurya & S. Balasubramanium; K001140912, 00 .vi.1901, Kannikatti; K001140913, 1974 , Sungel Kolok, Nikom Waeng, coll. K. Larsen & S.S. Larsen.

***Trichopodium cordatum*:** K001140906, 1867, Ceylon, coll. Walher; K001140907, 1867, Ceylon; K001140908, 1867, Ceylon, coll. Walher; K001140909, 1867, Mountale Falls, coll. Hooker.

***Trichopodium angustifolium*:** K001140911, 1867,

Ceylon, coll. Wehler.

Avetra sempervirens: K001140922, 19.xii.1997, 500m beyond a tomb on the path to Vatovavy, coll. P. Wilkin, G. Rafamananatsoa, C. Foster & L. Caddick; K001140935, 13.ii.2003, Madagascar, coll. J. Rabenantoandro et al.

Distribution and ecology

Trichopus zeylanicus ssp. *travancoricus* grows near the wet banks of streams and rivulets in the dense forests of Agasthyamalai Hills of southern Western Ghats, India.

Population studies

The distribution of the subspecies is known to be restricted to Agasthyamalai Biosphere Reserve, which was also confirmed by various field explorations conducted in different forest areas. According to the literature (Sivarajan et al. 1990), the herb is largely distributed in Thiruvananthapuram District of Kerala and its adjoining forest areas of KMTR in Tamil Nadu; This has also been seen in Kanyakumari District. The present study confirmed that the population of *T. zeylanicus* ssp. *travancoricus* is largely fragmented and eradicated in most of the forests of Kerala and Tamil Nadu due to illegal collection and over exploitation. Populations with small to medium number of individuals were observed near streams and shady areas and were seen to prefer a warm humid climate and heavy litter.

Since the species is known to have a wide range, it is not possible to analyse its global distribution status. During the study period, however, the extent of occurrence of *T. zeylanicus* ssp. *travancoricus* was analysed within the Agasthyamalai Biosphere Reserve. The subspecies has extended to about 46±11 km^2 of the forest area. The area of occupancy of the subspecies in the forest areas of KMTR is comparatively larger than that in the forest areas of Rosemala and Sornagiri. Other distribution areas of the subspecies, like lower Kodayar, were not explored in the current study. The area of occupancy of *T. zeylanicus* ssp. *travancoricus* is about 25–34 km^2 within the distribution area of the Agasthyamalai Biosphere Reserve. The number of individuals recorded per population was about 4–13 and the number of individuals per square kilometre was recorded as 77.4±15.67 in Kannikatti and Ingikuzhi forest areas. The populations of *T. zeylanicus* ssp. *travancoricus* at Sornagiri and Rosemala areas were not as healthy and the numbers of individuals recorded per square kilometre were 56.2±16.55 and 49.55±23.88, respectively. A major part of this forest reserve

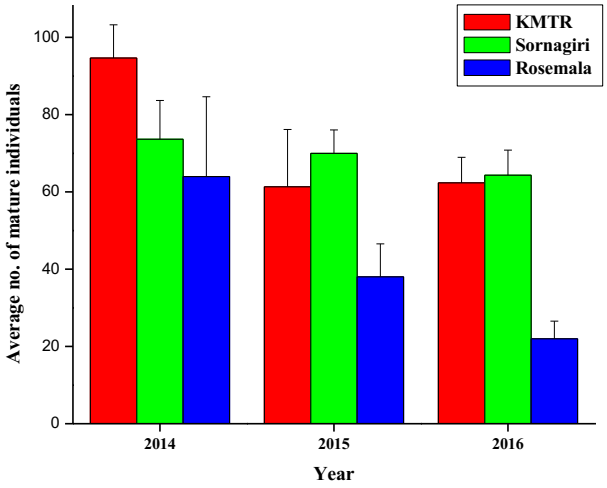

Figure 1. Average number of mature individuals of *Trichopus zeylanicus* ssp. *travancoricus* recorded from various study sites in the southern Western Ghats, India

is occupied by commercial plantations like rubber and coffee estates, vegetable cultivation, and other agriculture. Of the three study sites analysed, KMTR has comparatively healthy populations with more number of individuals (Fig. 1).

The rhizomatous herb grows prostrate along with a meagre number of associated species, namely *Cinnamomum* sp., *Pellionia heyneana*, *Ochlandra travancorica*, *Dioscorea* sp., *Anaphyllum wightii*, and *Calamus* sp. The subspecies also prefers to grow in the shaded areas of river banks. The mature individuals of *T. zeylanicus* ssp. *travancoricus* showed major difference in growth performance across the three study sites. The size of the leaves of the specimens collected from Sornagiri was larger (13.74±0.87 cm) when compared to that of Rosemala (12.34±1.6 cm) and KMTR (6.12±1.4 cm). Also, it had a greater variation in the average number of caulescant leaves per plant. The individuals collected from the populations of Ingikuzhi had more number of leaves (18–22) than those of Sornagiri (12–14) and Rosemala (11–13).

This subspecies is commercially collected for several medicinal drug formulations. Therefore, illegal collection from the forest areas must be banned while encouraging cultivation for commercial purposes. Moreover, due to the failure of intermittent rainfall, a major percentage of the seeds failed to germinate. The failures of the reproductive ability of the herb may considerably reduce its number of individuals per population.

Image 1. ***Trichopus zeylanicus*** **ssp.** ***travancoricus*****: a - adult mature individual; b - leaf flushing; c - closer view of flower; d - closer view of Gynostemium; e - SEM view of Gynostemium; f - SEM view of anther; g - closer view of fertilized ovary; h - mature fruits; i - infected fruit.**

Floral biology

The phenological events of the subspecies were observed during periodical field visits conducted at the different study sites. At leaf flushing, two-folded tender leaves developed with pale yellowish green colour before opening to its normal form. During the initiation of a fresh leaf, the size of the petiole was 4–6 cm, which then extended upto 22–30 cm as the leaf attained maturity. In most of the cases, the plant was completely dried off and the leaf flushing started in July and extended upto November, even though the plant emerged after the monsoon. The leaf flushing was followed by flowering in the middle of August. The peak flowering period was observed from September to

October and sometimes extended upto December. At the same time, the fruiting also started in November and extended upto March. In Rosemala and Sornagiri, however, flowering and fruiting were observed in most parts of the year due to occasional rainfall.

The flowers emerged from the pulvinus part of the petiole-like stem and the pedicel pushed towards one side by the petiole that usurps line of the stem. The flowers are solitary and highly attractive with a pedicel of approximately 16cm and are actinomorphic. The anthesis of the flower was recorded between 09.00h and 11.00h. The tepals are highly attractive and arranged in two rows, polypetalous, lanceolate in shape, apiculate at the tip, dark brownish with maroon colour at the center, and sandal with white at the tip (Image 1). The androecium comprising of six stamens are arranged in two whorls that alternate, opposite to the trifid stigma. Stamens have short filaments with two-celled anther, monosulcate, orange in colour that attached with the gynoecium and the anther dehisced on the day of anthesis. Fleshy, wet, two-lobed trifid stigma is reported at the centre of the flower. The development stages of the flower to fruit were observed and the ratio was calculated as 1:1. The fruit has three locules, each containing two seeds and the seeds located at the superposed position. The seeds are covered with dense brownish tomentum and the surface of the seeds are deeply furrowed and cerebriform are distinctive. Since,the seeds have a strong seed coat, they take more time to imbibe water. Majority of the fruits were foraged and damaged in its natural habitat. The seedlings were not observed in the natural habitat and preliminary studies on seed propagation did not yield good results. *Trichopus zeylanicus* ssp. *travancoricus* mostly propagated through the rhizome part of the subspecies.

In general, Agasthyamalai and its environments are already affected due to the conversion of the reserve forests into plantations, reservoirs, and roads. The present population studies of *T. zeylanicus* ssp. *travancoricus* revealed that the subspecies is facing a high risk of extinction in the distribution areas. Based on the field data on the extent of occurrence, area of occupancy, and numbers of populations and mature individuals, this subspecies can be included under the Endangered category of IUCN. Little observation only made on distribution, we have plan to observe detailed distribution of the herb in future. Effective conservation protocol and propagation techniques have to be developed for the sustainable utilization of this wonder herb.

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