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CYTOTAXONOMY OF TWO SPECIES OF PHYTOLACACEAE

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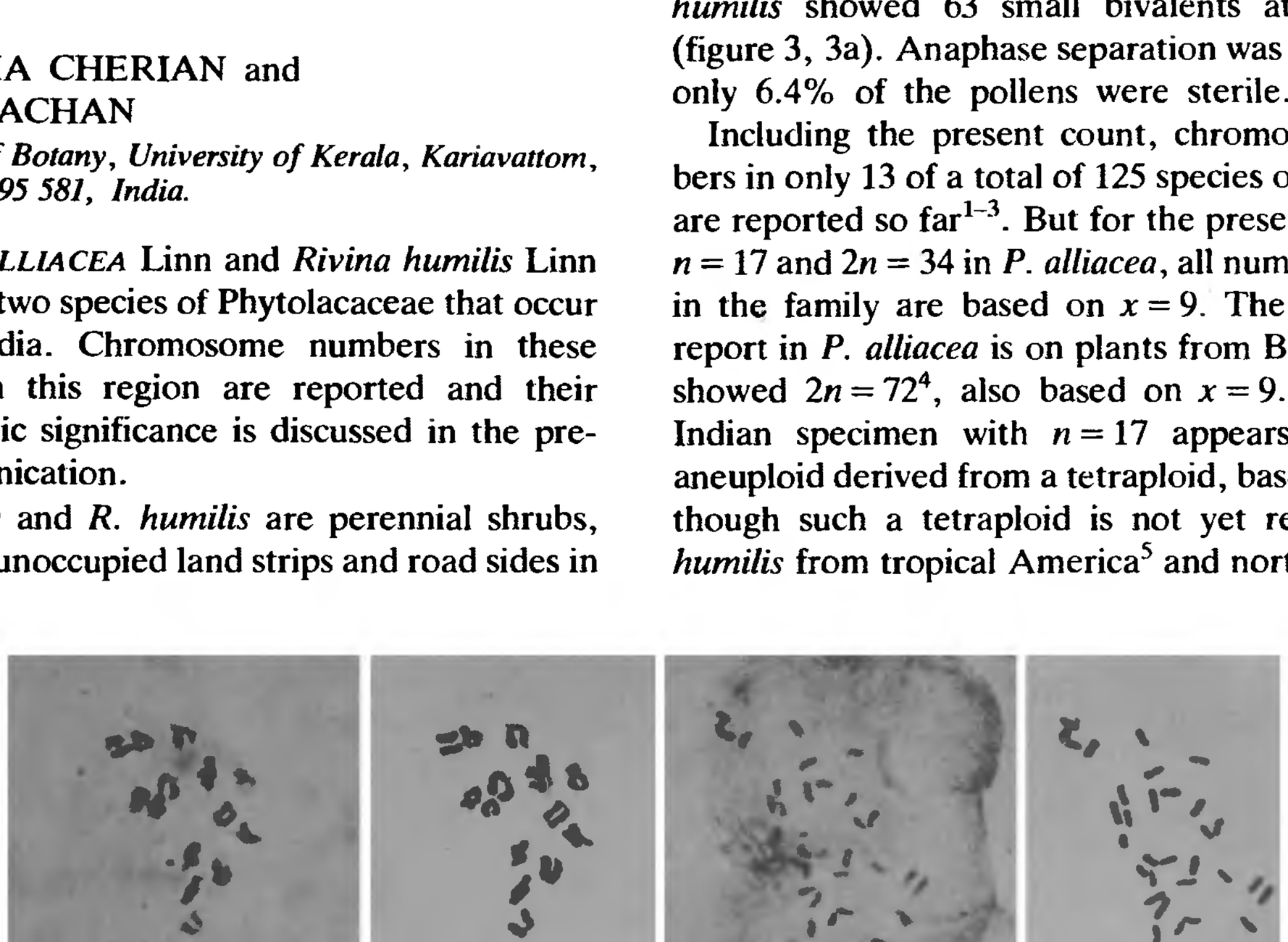
PETIVERIA ALLIACEA Linn and *Rivina humilis* Linn are the only two species of Phytolacaceae that occur in south India. Chromosome numbers in these species from this region are reported and their cytotaxonomic significance is discussed in the present communication.

P. alliacea and *R. humilis* are perennial shrubs, common on unoccupied land strips and road sides in Trivandrum city. Materials were collected from the Fine Arts College Campus in Trivandrum. Flower buds and root tips were fixed in acetic acid–ethanol (1:3) and stained in 1% acetocarmine. The PMCs of *P. alliacea* showed 17 bivalents (figure 1, 1a). Meiosis was regular and only 11.6% of the pollens were sterile as judged by the stainability in aceto-carmine. Root tip cells showed 34 chromosomes (figure 2, 2a). The somatic chromosomes vary in length from 1.3 μm to 2.6 μm and are either metacentric or submetacentric. The PMCs of *R. humilis* showed 63 small bivalents at diakinesis (figure 3, 3a). Anaphase separation was normal and only 6.4% of the pollens were sterile.

Including the present count, chromosome numbers in only 13 of a total of 125 species of the family are reported so far[1–3]. But for the present report of $n = 17$ and $2n = 34$ in *P. alliacea*, all numbers known in the family are based on $x = 9$. The only other report in *P. alliacea* is on plants from Brazil, which showed $2n = 72$[4], also based on $x = 9$. The south Indian specimen with $n = 17$ appears to be an aneuploid derived from a tetraploid, based on $x = 9$, though such a tetraploid is not yet recorded. *R. humilis* from tropical America[5] and northern India[6]



Figures 1–3. (×750) **1, 1a.** PMC of *P. alliacea* with 17 bivalents; **2, 2a.** Mitotic metaphase of *P. alliacea* with 34 chromosomes; **3, 3a.** PMC of *R. humilis* with 63 bivalents.

are known to be 12-ploids with $2n = 108$ ($x = 9$). The plants from south India with $2n = 126$ are 14-ploids based on $x = 9$. *P. alliacea* and *R. humilis* are native to tropical America and are naturalized in India[7]. The present study has brought to light the differences in chromosome numbers between plants of the species from their native and naturalized geographic regions. Investigation of these species throughout their range of distribution may reveal more cytotypes leading to interesting cytotaxonomic information associated with inter-continental dissemination of plants.

20 June 1986; Revised 14 July 1986

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IN VITRO LEAF SEGMENT CULTURE OF *VANDA TESTACEA* (LINDL) REICHB F (= *V. PARVIFLORA* LINDL) (ORCHIDACEAE)

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MERICULTURE, an important technique for commercial multiplication of orchids, involves sacrifice of the mother plant in the monopodial taxa. The utility of leaf tissue culture as an effective and alternate means of propagating elite and/or endangered orchid genotypes is being increasingly realized, ever since Wimber[1] reported the formation of protocorm-like bodies (plbs) from *Cymbidium* leaf tissues *in vitro*. It is possible to raise a large number of identical plants from a single leaf. Success has so far been achieved in 9 species using leaf segments as explants[2–4].

Vanda testacea ranks among the beautiful orchid species and has been extensively used as a progenitor of many merited hybrids. Its extensive collection for commercial purposes has endangered its existence in its natural habitats.

The present investigation was aimed to assess the regeneration potential of its leaves. Leaf segments (0.5–1.0 cm) procured from axenic cultures, were cultured on a nutrient medium recommended by Mitra *et al*[5] (BM). Besides peptone (2 g/l), 1 mg/l each of 3-indoleacetic acid (IAA), naphthaleneacetic acid (NAA), indole-3-butyric acid (IBA), gibberellic acid (GA_3), kinetin (KN) and 6-benzylamino purine (BAP) were used singly or in various combinations in the medium. All the experimental manipulations were carried out under aseptic conditions and the cultures were incubated at 25±2°C under a 12 hr photoperiod of 3,500 lx.

The explants perished in most of the media combinations after about 8 weeks without showing any organogenetic changes. However, their proliferation in the medium containing peptone +(GA_3)/(BAP+NAA) suggests a benign effect of selective growth adjuncts. Proliferation of leaf explants has been noted in media containing KN, NAA/IAA (*Phalaenopsis*[6], *Rhynchostylis*[4]) and coconut water/BAP/2, 4-D (*Renanthera*[7]). The meristematic activity was initiated on the entire surface or was restricted to the basal (figure 1) and/or apical (figure 2) ends of the explants depending upon the age of the source leaf. While it was restricted to the basal ends in relatively older explants, the younger explants exhibited increased meristematic activity suggesting that the physiological age of explant is an important factor for regeneration. Similar behaviour of explants was shown in *Phalaenopsis* and *Vanda* where leaf segments from juvenile plants produced plbs (protocorm-like bodies) with ease in contrast to those derived from adult plants which did not respond[8]. Irrespective of the age of the explant and the location of meristematic loculae, the proliferating cells developed into plbs in 2–3 weeks old cultures (figure 3). Leaf primordia were differentiated after 2 weeks of plb regeneration (figure 4). Further organogenesis was promoted upon subculturing the plbs on BM (figure 5) and plantlets complete with roots and shoots were obtained within 12–16 weeks (figure 6).