

MOLECULAR DATA AND PHYLOGENY OF FAMILY SMILACACEAE

ZABTA K. SHINWARI¹ AND SHEHLA SHINWARI²

¹*Dept. of Biotechnology, Quaid-i-Azam University-Islamabad*

²*Dept. of Botany, PMAS Arid Agriculture University, Rawalpindi*

Abstract

Family Smilacaceae's higher order taxonomy remained disputed for many years. It was treated as an order "Smilacales" and was also placed under Liliales by several taxonomists. Even some considered as part of family Liliaceae. In present paper, we investigated the family's higher order phylogeny and also compared its *rbcL* gene sequence data with related taxa to elucidate its phylogeny. The data suggests that its family stature is beyond dispute because of its advanced karyotype, woody climbing habit and DNA sequence data. The data suggest that Smilacaceae may be a sister group of order Liliales and it forms a clear clade with the order.

Introduction

The Family Smilacaceae was first recognized by Vent. However, its taxonomic status remained controversial. Smilacaceae is a small family with only 3 genera and about 320 species. (Mabberley 2008, Conran, 1998) In Pakistan it is represented by one genus and 4 species including a cultivated species (Ghazanfar, 1977). This family is mainly distributed in tropics but extending to temperate regions of both the hemispheres.

Banham and Hooker, (1883); Krause (1930) considered the genera of Smilacaceae under tribe Polygonatae of the family Liliaceae But majority of the present day taxonomists treat them under an independent family Smilacaceae because of its petiolar sheath tendrils, 1-chambered anther cells, sessile stigmas and mostly climbing habit (Fig. 1).

Hutchinson (1959) was of the view that *Smilacaceae* was highly evolved than other taxa of Liliales. Dahlgren *et al.*, (1985) treated this as a sub family under the order Dioscoreales on the basis of its leaf morphology which is very distinct from the rest of liliaceous genera but it can be distinguished by its superior ovary and baccate fruit.

Molecular systematics has replaced the orthodox taxonomy and added value to the discipline when used with morphological data and had been instrumental in identifying new taxa as well as clearing taxonomic status of complicated taxa (Shinwari, 1995 and Yousuf *et al.*, 2006). Many taxonomic issues were reported in Super Order Liliiflorae based on its morphological data (Shinwari *et al.*, 1994). To understand the intraspecific variations, RFLP of the chloroplast DNA were studied in some of the taxa like *Disporum* (Shinwari *et al.*, 1994a). However, to understand inter-specific variations and recognition of generic status of complicated taxa like *Prosartes* and *Disporum*, *rbcL* gene sequence data was analyzed and proved successful (Shinwari *et al.*, 1994b).

Taxonomic rank and placement of genus *Smilax* has also been difficult and it had a rank from independent order (Takhtajan, 1987) to a family of different orders, e.g., Liliales (Cronquist, 1981) or Dioscoreales (Dahlgren *et al.*, 1985). But it was treated at a lower level of hierarchy like member of tribe Polygonatae (Krause, 1930).



Fig.1. Smilax china-habit

To study and support morphological data rbcL gene sequence data of Smilax and other related genera were analyzed to confirm its rank in taxonomic hierarchy following Shinwari, (1998 and 2000). Our sole objective was to observe whether rbcL gene sequence data solve the relationship of Smilacaceae with different orders.

Material and Method

We studied rbcL gene sequence data of four orders namely, Liliales; Melanthiales, Asparagales and Dioscoreales sensu Dahlgren *et al.*, (1985). The genera studied are looped in Figs. 2 & 3. The voucher specimens are deposited in Herbarium Kyoto University (KYO) and the sequences have already been deposited in the DNA data bank of Japan DDBJ. The sequence data of Smilax was deposited under accession number D 28333. For DNA extraction, PCR amplification; primers used; DNA sequencing and Tree reconstruction, we followed Shinwari, (2002).

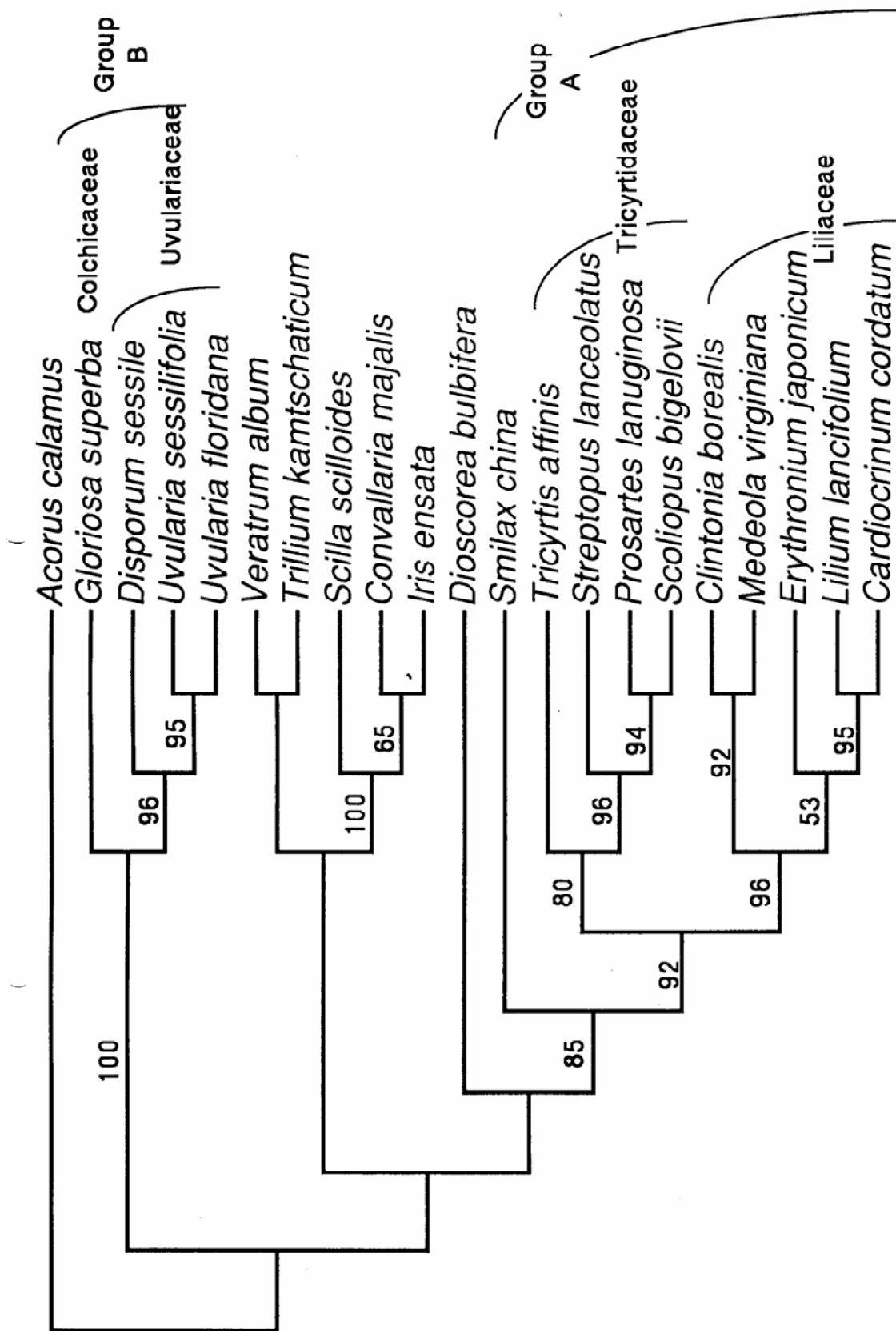


Fig. 2. The most parsimonious tree (Length=573, CI=0.640, RI=0.667). The value on branches indicate bootstrap probabilities (%) out of 100 Bootstrap replications (only more than 50 % are shown).

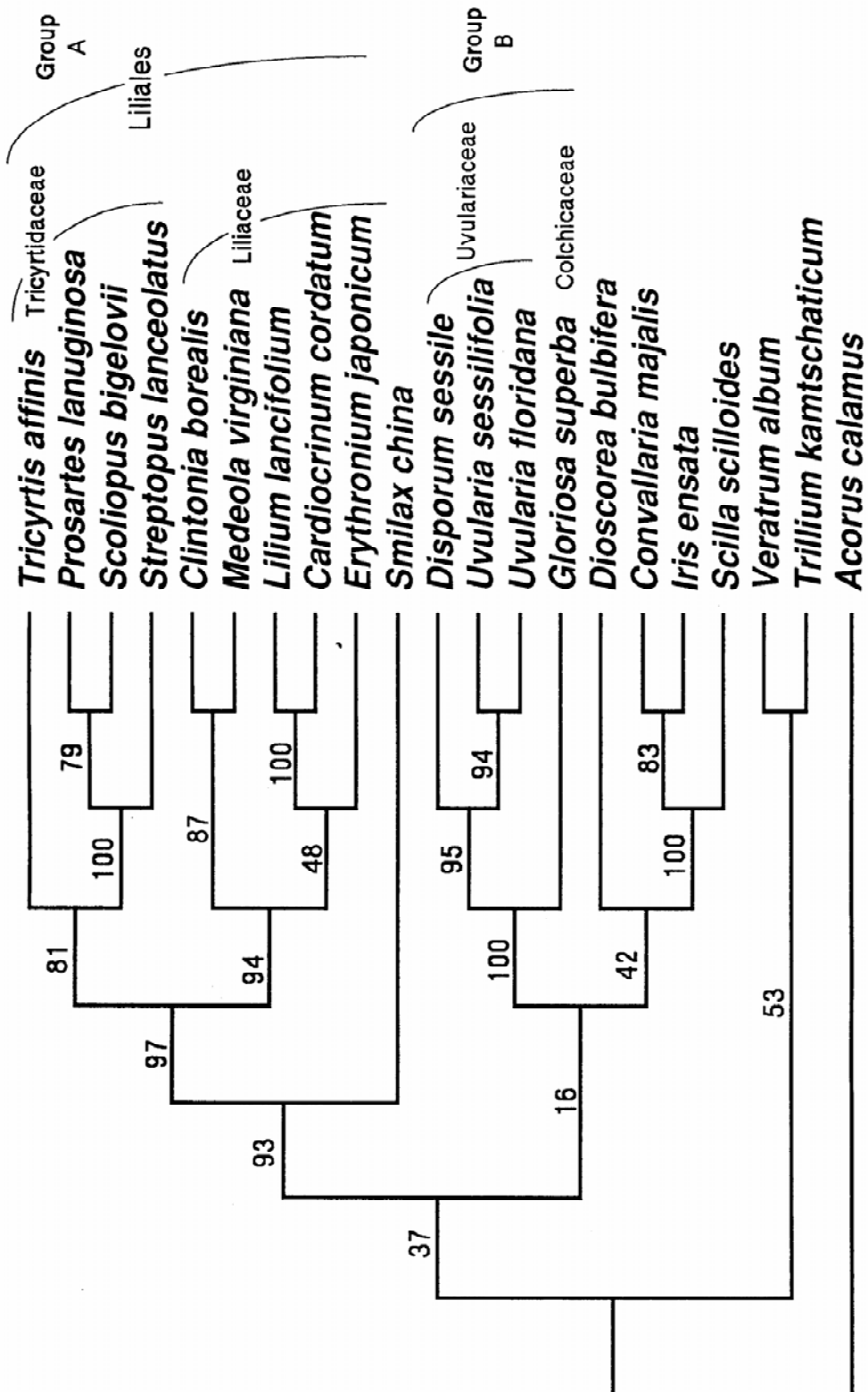


Fig. 3. Consensus tree Obtained by the neighbor joining method. Value on branches indicate the boots trapping probabilities

Results and Discussions

Dahlgren *et al.*, (1985) had studied Liliales sensu lato in detail, however, his study has raised certain questions for groups including *Smilax*. Earlier too in the last two

decades of the last century, there was debate regarding the placement of genera like *Smilax* by many (Takhtajan, 1980; Dahlgren and Clifford, 1982). There was no consensus on the placement of the family of Smilacaceae either under the order Liliales or Dioscoreales or Asparagales (Conran, 1987; Goldberg, 1989). However, phenetic and other cladistic approaches recommended splitting of Liliales sensu lato into small and more homogenous orders (Shinwari, 2002).

The number of substitution per site and other details has already been reported by Shinwari, (2002). Our study supported that *Smilax* is better placed within Liliales as it is clustered with the other genera of Liliales with a boot strap probability of 85%. Though Dioscoreales in turns comes closer to it. This study confirms that Liliales sensu Dahlgren *et al.*, (1985) is not a monophyletic group.

Present data supports the placement of Smilacaceae in *Liliales* (Fig. 2 & 3) which is congruent with Hutchinson (1959), Sen (1975) and Cronquist (1981). The results indicate that this can be treated as a sister group of Liliales sensu stricto.

Acknowledgements

We are grateful to all friends in Japan who supported the principal author while he was a scholar of Ph.D. degree. Presently, Higher Education Commission, Pakistan is supporting financially the lab which is highly appreciated and acknowledged.

References

- Banham G. and Hooker, J.D. 1883 *Genera Plantarum* 3: 748-837 London.
- Conran, J.G. 1987A phenetic study of the relationships of Drymophila R. Br. Within the reticulate veined Liliiflorae. *Austr. J. Bot.* 35: 283-300.
- Conran, J. G. 1998. Smilacaceae in Kubitzki, K. (Ed.) *The Families and Genera of Tascular Plant* vol.III springer- verlag, Berlin.
- Cronquist, A. 1981. *An integrated system of classification of flowering plants*. Columbia Univ. Press, New York.
- Dahlgren, R.M.T. and Clifford, H.T. 1982. *The Monocotyledons- A Comparative Study*. Academic Press, London.
- Dahlgren, R.M.T. Clifford, H.T. and Yeo, P.F. 1985 *The Families of the Monocotyledons*. Springer, Berlin.
- Ghazanfar, S. 1977 "Smilacaceae" in Nasir, E. and S.I. Ali, *Flora of Pakistan*. Fase. No. 107. Islamabad.
- Goldberg, A. 1989 *Classification, Evolution, and Phylogeny of the families of Monocotyledons*. *Smithsonian Contr. Bot.* 71: 1-74
- Hutchinson, J. 1959 *The Families of the Flowering Plants*. 2: 591-621. Oxford Clarendon Press.
- Krause, K. 1930 Liliaceae- In Krause, A. and Prantle, K. (Eds.): *Die Natürlichen Pflanzenfamilien* 15a, pp. 227-386.- Leipzig: Engelman.
- Mabberley, D.J. 2008. *A portable Dictionary of plants, their classification and uses*. Cambridge University press.
- Sen, S. 1975. Cytotaxonomy of Liliales. *Fedd. Rep.* 86(5): 255-305.
- Shinwari, Z.K., R. Terauchi and S. Kawano 1994 Phylogenetic relationships among genera in the Liliaceae-Asparagoideae-Polygonatae sensu lato inferred from rbcL gene sequence data. *PI. Systematic & Evolution* 192: 263-277.
- Shinwari, Z.K.; R. Terauchi and S. Kawano 1994a Molecular Systematics of Liliaceae-Asparagoideae-Polygonatae. 1. RFLP analysis of cpDNA in several species of Asiatic Disporum species. *Plant Species Bio.* 9: 11-18.

- Shinwari, Z.K.; R. Terauchi; F.H. Utech and S. Kawano 1994b Recognition of the New World Disporum Section Prosartes as Prosartes (Liliaceae) based on the sequence data of the *rbcL* gene. *Taxon* 43(3): 353-366.
- Shinwari, Z.K. 1995 Congruence between morphology and molecular phylogenetics in Prosartes (Liliaceae). *Pak. J. Bot.* 27(2): 361-369.
- Shinwari, Z.K 1998 Molecular Systematics of the genus *Uvularia* and related taxa based upon *rbcL* gene sequence data. *Pak. J. Bot.* 30(2): 161-172
- Shinwari, Z. K. 2000 Chloroplast DNA variation in Polygonatae (Liliaceae). *Pak. J. Bot.* 32(1): 7-14
- Shinwari, Z. K. 2002 Sequence divergence of *rbcL* gene and Phylogenetic relationships in Liliales. *Pak.J.Bot.* 34(2): 191-204
- Takhtajan, A.1980 Outline of the Classification of Flowering Plants. *The Botanical Review*, 46: 225-359.
- Yousuf Z., Masood S., Shinwari Z.K. Khan, M.A. and Rabbani, A.2006 Evaluation of Taxonomic Status of Medicinal Species of the Genus Solanum and Capsicum, Based on Poly Acrylamide Gel Electrophoresis. *Pak. J. Bot.*, 38(1): 99-106