

Chromosome counts in two species of *Curcuma* Linnaeus (Zingiberaceae) from North-East India

Judith Mary Lamo and Satyawada Rama Rao¹

Department of Biotechnology and Bioinformatics, North-Eastern Hill University,
Shillong-793022, Meghalaya, India

¹E-mail: srrao22@yahoo.com

[Received 12.10.2014; Revised 26.11.2014; Accepted 07.12.2014; Published 31.12.2014]

Abstract

The somatic chromosome number of two species of *Curcuma* from Northeast India has been investigated. Chromosome counts were carried out in root tip cells by squash preparation. All the root tip cells analyzed show the chromosome number of $2n = 63$ in both the species viz. *C. longa* and *C. caesia*. Probable triploid nature of both the species has been indicated.

Key words: *Curcuma caesia*, *Curcuma longa*, Chromosome number, Polyploidy

INTRODUCTION

The genus *Curcuma* Linnaeus of Zingiberaceae comprises of about 100 species (Velayudhan *et al.* 1999) including some most useful and popular ones viz. *C. longa* (Sasikumar 2005). The origin and spread of the genus favoured its Indo–Malayan origin and centre of diversity theory (Harlan 1975). The genus *Curcuma* Linnaeus contains many taxa of economic, medicinal, ornamental and cultural importance (Velayudhan *et al.* 2012).

Chromosome numbers and genome size of plant species provide authentic information which is useful in systematic, evolution and conservation of plants (Bennett & Leitch 2005; Guerra 2008), especially in taxa with smaller chromosomes like *Curcuma* (Skornickova *et al.* 2007). A quick perusal of published literature reveals that somatic chromosome numbers have been investigated in various species of *Curcuma* and polyploidy has been implicated in the diversification and evolution of the species (Suigura 1931; Raghavan & Venkat 1943; Chakravorti 1948; Sato 1948; Ramachandran 1961; Prana 1977; Apavatjirut *et al.* 1996; Das *et al.* 1999; Joseph *et al.* 1999; Ardiyani 2002; Sirisawad *et al.* 2003; Islam 2004; Skornickova *et al.* 2007; Nair & Sasikumar 2009). However, there are no detailed cytogenetical reports about *Curcuma* species growing in wild and/or cultivated in northeast India. Therefore, the present investigations have been undertaken to determine the somatic chromosome number in two species belonging to the genus *Curcuma* viz. *C. caesia* Roxburgh and *C. longa* Linnaeus which are medicinally important species of the region.

MATERIAL AND METHODS

The rhizomes of *C. caesia* (IISR 700) and *C. longa* (IISR Pratibha) used in the present investigation were obtained from Indian Institute of Spices Research, Kozhikode, Kerala. The plants were grown and maintained in poly-house conditions at the Department of

Biotechnology and Bioinformatics, North-Eastern Hill University, Shillong. Actively growing root tip of 1 – 2 cm long were excised from field grown plants and were pretreated with saturated para-dichlorobenzene (HiMedia) for 3 hours at room temperature. The root tips after pretreatment were fixed in freshly prepared 1 : 3 acetic alcohol for a minimum period of 24 hrs and thereafter stored in 70 % ethanol till those are squashed. Root tips were hydrolysed in 1N HCl for 8 minutes at 60° C and stained in leucobasic-fuschin (HiMedia) for 45 minutes at room temperature. The stained root tips were then thoroughly washed with distilled water and finally squashed in 1 % acetocarmine (Shamurailatpam *et al.* 2012).

Photomicrographs of the metaphase plates were taken from temporary preparations using Leica DFC 310FX microscope. At least 5 well spread metaphase plates were used for the study. The voucher slides for future verification have been stored at Plant Biotechnology Laboratory, Department of Biotechnology and Bioinformatics, North-Eastern Hill University, Shillong.

RESULTS AND DISCUSSION

In the present study, the somatic chromosome numbers of *C. caesia* and *C. longa* were determined to be $2n = 63$ (Figs. 1 a & b). Karyological investigation of *Curcuma* species is rather challenging and errors may easily be introduced due to relatively high number but smaller chromosomes (Ramachandran 1961; Apavatjrut 1996; Joseph *et al.* 1999; Sirisawad *et al.* 2003). Different chromosome counts for the same species have also been reported in some cases. However, in present investigations the occurrence of $2n = 63$ has been clearly recorded with the help of neat, clean and unambiguous preparations in *C. longa* and *C. caesia* with no evidence of numerical variations, whatsoever. The occurrence of $2n = 63$ as somatic chromosome number is in agreement with the findings of Ramachandran (1961),



Fig. 1. Mitotic complements in *Curcuma* spp. **a.** *C. caesia* ($2n = 63$); **b.** *C. longa* ($2n = 63$). Scale bar = 10 μ m.

Joseph *et al.* (1999) and Islam (2004). However, the somatic chromosome number in *C. caesia* has also been reported to be $2n = 22$ (Das *et al.* 1999) which is quite unrelated chromosome count and thus our observations are at variance with the report of Das *et al.* (1999). On contrary, $2n = 22, 48, 62, 64$ and 93 have been reported by different workers for *C. longa* (Raghavan & Venkatasubban 1943; Ramachandran 1961; Sharma & Bhattachrya

1959; Sato 1960; Scornickova *et al.* 2007). The present authors opine that these unrelated chromosome numbers might be due to ambiguity in cytological preparations and/or technical difficulties in obtaining quality preparations. There has been continued disparity and controversy concerning the basic chromosome number of the genus *Curcuma* (Scornickova *et al.* 2007). Three basic numbers of $x = 7, 16, 21$ have been proposed for the genus *Curcuma* by various cytogeneticists (Raghavan & Venkatasubban 1943; Ramachandran 1961; Sharma & Bhattacharya 1959; Sato 1960; Scornickova *et al.* 2007). However, our study indicates that the basic chromosome number could be $x = 21$ in conformity with the reports of Ramachandran (1961), Prana (1978), Ardhiyani (2002) and Islam (2004) which is a widely accepted basic chromosome number. Therefore, based on our study, *C. caesia* and *C. longa* could be regarded as possible triploid ($3x$) species. Chromosome counts provide indispensable information on genetic discontinuities within and among the species and they contribute to our understanding of phylogenetic relationships at all taxonomic levels (Semple *et al.* 1989). However, it is untenable to depend only on chromosome counts to resolve the basic chromosome number in *Curcuma*. Therefore, further cytogenetical investigations related to meiotic analysis should be carried to ascertain the basic chromosome number in *Curcuma* and determine the type of polyploidy ($3x$) which is recorded here in *C. caesia* and *C. longa*.

Acknowledgements

The authors are thankful to the Head, Department of Biotechnology and Bioinformatics, North-Eastern Hill University, Shillong for providing necessary facilities. The authors are also thankful to Indian Institute of Spices Research, Kozhikode for providing the germplasm, DBT-JRF Program, Department of Biotechnology, Government of India for financial assistance. Sincere thanks are also due to the members of Plant Biotechnology, Department of Biotechnology and Bioinformatics, North-Eastern Hill University Shillong, for their constant help and encouragement.

LITERATURE CITED

- Apavatjirut, P.; Sirisawad, T.; Sirirugsa, P.; Voraurai, P. & Suwanthada, C. 1996. Studies on chromosome number of seventeen Thai *Curcuma* species. *Proceed. 2nd Nat. Conf. Flwr. Ornam. Pl.* 2: 86 – 99.
- Ardhiyani, M. 2002. *Systematic study of Curcuma L.: turmeric and its allies*. PhD thesis, University of Edinburgh, Scotland.
- Bennett, M.D. & Leitch, I.J. 2005. Nuclear DNA amounts in angiosperms: progress, problems, and prospects. *Ann. Bot.* 95: 45 – 90.
- Chakravorti, A.K. 1948. Multiplication of chromosome numbers in relation to speciation in Zingiberaceae. *Sci. Cult.* 14: 137 – 140.
- Das, A.B.; Rai, S. & Das, P. 1999. Karyotype analysis and cytophotometric estimation of nuclear DNA content in some members of the Zingiberaceae. *Cytobios* 97: 23 – 33.
- Guerra, M. 2008. Chromosome numbers in plant cytotaxonomy: Concepts and implications. *Cytogen. Genome Res.* 120: 339 – 350.
- Harlan, J.R. 1975. *Crops and Man*. American Society of Agronomy, Madison, Wisconsin, USA. 295 pp.
- Islam, M.A. 2004. *Genetic diversity of the genus Curcuma in Bangladesh and further biotechnological approaches for in vitro regeneration and long-term conservation of C. longa germplasm*. PhD thesis, University of Hannover, Germany.

- Joseph, R.; Joseph, T. & Joseph, J. 1999. Karyomorphological studies in the genus *Curcuma* Linn. *Cytologia* 64: 313 – 317.
- Nair, R.R. & Sasikumar, B. 2009. Chromosome number variation among germplasm collections and seedling progenies in turmeric, *Curcuma longa* L. *Cytologia* 74(2): 153 – 157.
- Prana, M.S. 1977. *Studies on some Indonesian Curcuma species*. PhD thesis, University of Birmingham, England.
- Raghavan, T.S. & Venkatasubban, K.R. 1943. Cytological studies in the family Zingiberaceae with special reference to chromosome number and cyto-taxonomy. *Proceed. Indian Acad. Sci., Series B* 17: 118 – 132.
- Ramachandran, K. 1961. Chromosome numbers in the genus *Curcuma* Linn. *Curr. Sci.* 30: 194 – 196.
- Sasikumar, B. 2005. Genetic resources of *Curcuma*: diversity, characterization and utilization. *Pl. Genet. Resour.* 3: 230 – 251.
- Sato, D. 1948. Karyotype and systematics of Zingiberales. *Jap. J. Genet.* 23: 44 – 45.
- Sato, D. 1960. The karyotype analysis in Zingiberales with special reference to the protokaryotype and stable karyotype. *Scientific Papers of the College of General Education, University of Tokyo* 10: 225 – 243.
- Seiple, J.C.; Chmielewski, J.G. & Windham, M.D. 1989. Chromosome number determination in fam. Compositae, tribe Astereae. III. Additional counts and comments on generic limits and ancestral base number. *Rhodora* 91: 296 – 314.
- Shamurailatpam, A.; Madhavan, L.; Yadav, S.R.; Bhat, K.V. & Rao, S.R. 2012. Chromosome diversity analysis in various species of *Vigna* Savi from India. *Nucleus* 55(2):107 – 114.
- Sharma, A.K. & Bhattacharya, N.K. 1959. Cytology of several members of Zingiberaceae. *La Cellule* 59: 297 – 346.
- Sirisawad, T.; Sirirugsa, P.; Suwanthada, C. & Apavtjirut, P. 2003. Investigation of chromosome numbers in 20 taxa of *Curcuma*. In: Chantaranonthai P, Larsen K, Sirirugsa P, Simpson D, (eds.). *Proceedings of the 3rd Symposium on the family Zingiberaceae*. Khon Kaen: Applied Taxonomic Research Centre, Khon Kaen University, Pp. 54 – 62.
- Skornickova, J.; Sída, O.; Jarolimova, V.; Sabu, M. & Suda, J. 2007. Chromosome numbers and genome size variation in Indian species of *Curcuma* (Zingiberaceae). *Ann. Bot.* 100: 505 – 526.
- Velayudhan, K.C.; Dikshit, N. & Nizar, M.A. 2012. Ethnobotany of turmeric (*Curcuma longa* L.). *Indian J. Trad. Knowl.* 11(4): 607 – 614.
- Velayudhan, K.C.; Muralidharan, V.K.; Amalraj, V.A.; Gautam, P.L.; Mandal, S. & Dinesh, K. 1999. *Curcuma Genetic Resources*. Scientific Monograph No. 4. New Delhi: National Bureau of Plant Genetic Resources.