Chromosome Counts in Wild and Cultivated Species of
Curcuma Linn.

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Summary In the present study, chromosome numbers in 13 wild and cultivated Curcuma species were investigated. Chromosome number of C. sylvatica is reported for the first time. Cytological analyses on 13 Curcuma species indicated the presence of 2n=42, 63 and 105 somatic chromosome number, which indicate the plausible occurrence of polyploidy in the genus.

Key words Curcuma, Chromosome number, Polyploid, Cytological analysis.

The genus Curcuma, an important member of the family Zingiberaceae (Jatoi et al. 2007), consists of many species which are economically and ornamentally important. Many species are highly prized for their medicinal properties. The genus comprises of 120 species (Leong-Škorničková et al. 2007) which are distributed in south and south-east Asia including China, Australia and the South Pacific (Larsen et al. 1998, Wu and Larsen 2000, Ye et al. 2008, Chen and Xia 2011). Curcuma species display a great quantum of diversity in habitat, morphology and ethnobotanical uses (Syamkumar and Sasikumar 2007).

Cytological studies have been of considerable value for solving taxonomic riddles and can also provide insights into the evolutionary mechanisms that shape the plant genome (Bennett and Leitch 2005, De Storme and Mason 2014). Significant variations in the chromosome number and ploidy level in the genus Curcuma have been reported by many authors (Sugiura 1931, Sharma and Bhattacharya 1959, Ramachandran 1961, Das et al. 1999, Joseph et al. 1999, Leong-Škorničková et al. 2007). Curcuma is a complex genus and polyploidization has further complicated our understanding towards the evolution and phylogeny (Leong-Škorničková et al. 2007, Chen and Xia 2011). Therefore, the present study was undertaken to study the somatic chromosome numbers and verify the ploidy level in some cultivated and wild species of Curcuma.

Materials and methods

The rhizomes of 13 Curcuma species used in the present investigation were obtained from Indian Institute of Spices Research, Kozhikode, Kerala (Table 1). The plants were grown and maintained in poly-house conditions at the Department of Biotechnology and Bioinformatics, North-Eastern Hill University, Shillong.

For chromosome count, actively growing root tips of 1–2 cm long were excised from field grown plants and were pretreated with saturated para-dichlorobenzene (HiMedia) for 3 h at room temperature. The root tips were then fixed in Carnoy`s solution for 24 h and thereafter stored in 70% ethanol. Hydrolysis was carried out in 1 N HCl for 8 min at 60°C and stained in leucobasic-fuschin (HiMedia) for 45 min at room temperature. The stained root tips were then thoroughly washed with distilled water and finally squashed in 1% acetocarmine (Kumar and Rao 2002). Photomicrographs of the metaphase plates were taken from temporary preparations using a Leica DFC 310FX microscope. At least five well spread metaphase plates were used for the analysis.

Table 1. Specimen voucher and chromosome count in Curcuma species.

<table>
<thead>
<tr>
<th>Sl. No</th>
<th>Species</th>
<th>IISR voucher numbers</th>
<th>2n</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>C. amada Roxb.</td>
<td>521</td>
<td>42</td>
</tr>
<tr>
<td>2</td>
<td>C. aromatic Salisb.</td>
<td>711</td>
<td>42</td>
</tr>
<tr>
<td>3</td>
<td>C. comosa Roxb.</td>
<td>644</td>
<td>42</td>
</tr>
<tr>
<td>4</td>
<td>C. haritha Mangaly &amp; M. Sabu</td>
<td>1136</td>
<td>42</td>
</tr>
<tr>
<td>5</td>
<td>C. mangga Valeton &amp; Zijp</td>
<td>1049</td>
<td>42</td>
</tr>
<tr>
<td>6</td>
<td>C. montana Roxb.</td>
<td>649</td>
<td>42</td>
</tr>
<tr>
<td>7</td>
<td>C. aeruginosa Roxb.</td>
<td>1144</td>
<td>63</td>
</tr>
<tr>
<td>8</td>
<td>C. latifolia Roscoe</td>
<td>638</td>
<td>63</td>
</tr>
<tr>
<td>9</td>
<td>C. leucorrhiza Roxb.</td>
<td>1169</td>
<td>63</td>
</tr>
<tr>
<td>10</td>
<td>C. sylventica Valeton</td>
<td>526</td>
<td>63</td>
</tr>
<tr>
<td>11</td>
<td>C. zanthorrhiza Roxb.</td>
<td>1108</td>
<td>63</td>
</tr>
<tr>
<td>12</td>
<td>C. zedoaria (Christm.) Roscoe</td>
<td>760</td>
<td>63</td>
</tr>
<tr>
<td>13</td>
<td>C. raktakanta Mangaly &amp; M. Sabu</td>
<td>1137</td>
<td>105</td>
</tr>
</tbody>
</table>

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Results and discussion

In the present investigation, the somatic chromosome numbers were counted in root tip cells by squash preparations. Photomicrographs of the metaphase plates are depicted in Figs. 1–3, and data are summarized in Table 1. The somatic chromosome number in root tip cells of *C. amada*, *C. aromatica*, *C. comosa*, *C. haritha*, *C. mangga* and *C. montana* were found to be $2n=42$. That of *C. aeruginosa*, *C. latifolia*, *C. leucorrhiza*, *C. sylvatica*, *C. zanthorrhiza* and *C. zedoaria* was found to be $2n=63$, indicating their possible triploid/polyploid nature. The same somatic chromosome number was reported for *C. longa* and *C. caesia* in our earlier observations (Lamo and Rao 2014). However, *C. raktakanta* had $2n=105$, indicating the possible occurrence of pentaploidy. Among the species studied, the somatic chromosome numbers of *C. sylvatica* with $2n=63$ is reported for the first time (Fig. 2D). However, our observations in case of the other 12 remaining species are in complete agreement with other previous chromosomes reports (Raghavan and Venkatasubban 1943, Chakravorti 1948, Sharma and Bhattacharya 1959, Ramachandran 1961, Islam 2004, Leong-Škorničková et al. 2007, Joseph et al. 1999). Different chromosome numbers ranging from 20 to 84 for the same species have also been reported in some cases, which might be due to potential error in chromosome counting, small size of the chromosome, difficulty in obtaining good mitotic spread, and staining. In the present investigation, the chromosome numbers were recorded with the help of clean, neat and unambiguous preparations as depicted in the photomicrographs (Figs. 1–3).

There has been continued disparity concerning the basic chromosome number in the genus *Curcuma*. Different basic chromosome numbers, $x=9$, 12, 16, 21, were highlighted in early cytological reports (Raghavan and Venkatasubban 1943, Venkatasubban 1946, Chakravorti 1948, Sharma and Bhattacharya 1959, Ramachandran 1961, Nambiar 1972). However, Leong-Škorničková et al. (2007) regarded $x=7$ as the most likely basic chromosome number of the genus *Curcuma*. The present

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![Fig. 1. Mitotic complements of Curcuma species. A. C. amada ($2n=42$); B. C. aromatica ($2n=42$); C. C. comosa ($2n=42$); D. C. haritha ($2n=42$); E. C. mangga ($2n=42$); F. C. montana ($2n=42$). Scale bar=5μm.](image-url)
study, however, indicate that $x=21$ could be the probable basic chromosome number and is in concordance with the other reports (Ramachandran 1961, Islam 2004). Most species analysed are found to be diploids with chromosome number $2n=2x=42$ (C. amada, C. aromatic, C. comosa, C. haritha, C. mango and C. montana), some are triploids $2n=3x=63$ (C. aerugiosa, C. latifolia, C. leucorrhiza, C. sylvatica, C. zanthorrhiza and C. zedoaria), while C. raktakanta with chromosome number $2n=105$ may be a probable pentaploid with $2n=5x=105$. Polyploidy is most common among perennial plants possessing various vegetative means of reproduction (Hieter and Griffiths 1999, Ram et al. 2012) like Curcuma.

Relatively large chromosome numbers with smaller chromosome size, as observed in the genus Curcuma,

![Fig. 2. Mitotic complements of Curcuma species. A. C. aeruginosa (2n=63); B. C. latifolia (2n=63); C. C. leucorrhiza (2n=63); D. C. sylvatica (2n=63). Scale bar=5 μm.](image)

![Fig. 3. Mitotic complements of Curcuma species. A. C. zanthorrhiza (2n=63); B. C. zedoaria (2n=63); C. C. raktakanta (2n=105). Scale bar=5 μm.](image)
is apparently a consequence of polyploidization which is an adaptive nature of the chromosomes (Darlington 1956, Stebbins 1966). Chromosome counts provide indispensable information about the natural genetic variation among the species and also contribute to our understanding about the phylogenetic relationship/evolution at all taxonomic levels (Semple et al. 1989, Kumar and Singhal 2011). The very high chromosome number coupled with the small size of the chromosomes indicates a higher evolutionary status of many a plants (Sharma 1984) and polyploidy could be implicated in the diversification. It can be considered as an important factor in the evolution of the genus Curcuma too. Further, male meiotic analysis can help to authenticate the basic chromosome number in the genus Curcuma, and the efforts are in progress in this direction.

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References


