KARYOMORPHOLOGY AND CHROMOSOME NUMBER CONFIRMATION IN
URARIA PICTA (JACQ.) DC.
Bhattacharya Arnab and Datta K. Animesh*
Department of Botany, Cytogenetics and Plant Breeding Section, University of Kalyani, Kalyani – 741235, West Bengal, India

ABSTRACT
Karyomorphological studies performed in Uraria picta (Jacq.) DC. (Family: Leguminosae; Subfamily: Papillionidae), an important Ayurvedic herb (Ayurvedic name: Prishni parni) revealed 7 (2n = 22; 2L_m + 2L_sac + 4M_n + 2M_m + 2S_m + 8S_a + 2S_s) morphologically distinct chromosome types. The chromosomes in the species were graded (long: L - ≥ 2.0µm; medium: M - ≥ 1.0µm; small: S - < 1.0µm) on the basis of absolute chromosome length (0.59µm to 2.625µm). The karyotype showed predominance of chromosomes with metacentric (m) primary constrictions; although, it was asymmetric in nature (TF%: 37.65). Total haploid chromosome length was noted to be 12.63µm ± 3.73. Two pairs of chromosomes in the complement were with secondary constrictions and they were associated both with short as well as long arms. The chromosome number (2n = 22) analyzed in root tip mitosis was in agreement to the number noted in microsporocytes (n = 11). The confirmed number in the species is 2n = 22.

KEYWORDS: Uraria picta, Karyomorphology, meiotic analysis chromosome number confirmation.

INTRODUCTION
Uraria picta (Jacq.) DC. (Family: Leguminosae; Subfamily: Papillionidae) is an important herb (also reported to be a woody herb\(^2\), a perennial herb\(^3,4\) and a shrub\(^5\)) in Ayurvedic (Ayurvedic name: Prishni parni) medicine (used parts: roots/leaves/whole plant\(^6\)). The chromosome number is reported to be variable (\(n = 11\) \(, 12\) \( \pm 2\) in the genus Uraria\(^7\) in the species; although, chromosome number alone does not provide any cytogenetical information which may be used to design breeding strategies for crop improvement. The present communication reports on the karyomorphology of U. picta, which is previously undescribed, with an objective to collect cytological information for efficient breeding. The chromosome number recorded in root tip mitosis was also confirmed from meiotic preparations. Although, cytological studies are rather meager in Ayurvedic plant species\(^8\), they often carry a wealth of information for breeders, geneticists and other researchers.

MATERIALS AND METHODS
Root tips (generated from pre-soaked petri plate germinated seeds of Uraria picta; seed samples were collected from Medicinal Plant Garden, Narendrapur, Ramkrishna Mission, Govt. of West Bengal) of about 2.0 to 3.0mm length were pretreated in 0.1% colchicine for 3 hours 30 mins (room temperature – 33.0°C ± 1.0°C) washed in distilled water and kept in 0.01M KCl: 0.005M NaCl mixture (3:1) for overnight before fixing in 70.0% alcohol and storage under refrigeration. The root tips were stained in 4.0% aceto-orcein: 4N HCl mixture (9:2) for 2 hours and finally squashed in 45.0% acetic acid and observed under the microscope. Suitable metaphase plates (3 properly and uniformly condensed) were scored and measurements of the chromosomes were taken from camera lucida drawings at the magnification of 20×100x.

The centromeric nature in the chromosomes were classified as metacentric (m), sub-metacentric (sm), sub-acrocentric (sac) and acrocentric (ac) in accordance to Hirahara and Tatuno\(^7\). On the basis of chromosome length (long: L - ≥ 2.0µm; medium: M - ≥ 1.0µm; small: S - < 1.0µm), centromeric position and the presence of secondary constrictions (sc), the chromosomes were morphologically graded: type A: long metacentric chromosomes with secondary constrictions associated to short arm, type A1: long sub-metacentric chromosomes with satellites and it is with long arm, type B: medium sized chromosomes possessing metacentric primary constrictions, type C: medium sized chromosomes with sub-acrocentric primary constrictions, type D: small sub-metacentric chromosome, type E: small metacentric chromosomes and type F: small chromosomes with acrocentric primary constrictions. Total haploid chromatin length, TF% (proportion of short arm in the total chromatin length) and S% (relative length of shortest chromosomes compared to the longest) were also analyzed.

Karyomorphological data was used to compute karyotype formula. Meiotic chromosome preparations were made as was suggested by Bhattacharya and Datta\(^11\, 18\). Microphotographs for both mitotic and meiotic chromosomes were taken from temporary squash preparations and subsequently magnified.

RESULTS AND DISCUSSION
Karyomorphological studies revealed 7 (2n = 22; 2L_m + 2L_sac + 4M_n + 2M_m + 2S_m + 8S_a + 2S_s) morphologically distinct chromosome types (Table 1, Fig 1.A-B). The karyotype showed prevalence of chromosomes (14 out of 22) with metacentric primary constrictions (F%: 40.27 to 48.47); although, 4 chromosomes with sub-metacentric (F%: 32.62 to 37.84), 2 with sub-acrocentric (F%: 23.08) and 2 with acrocentric (F%: 15.25) constrictions were also present. Absolute chromosome length in the complement ranged from 0.59µm to 2.625µm. Two pairs of long chromosomes (2.08µm to 2.625µm) were with satellites and the satellites were associated with both the short arm (type A) as well as long arm (type A) of the chromosomes. Total haploid length and S% in the species were 12.63µm ± 3.73 and 22.48 respectively. The karyotype was asymmetric in nature as evident from TF% (37.65).
Meiotic analysis revealed (study performed for 3 consecutive years in 3 to 5 randomly selected plants every year; data pooled over the plants) 2n = 22 chromosomes (1st year – 0.11 IV + 10.29 II + 0.98 I, 236 cells scored; 2nd year - 0.16 IV + 10.78 II + 0.98 I, 182 PMCs analyzed; 3rd year -0.14 IV + 10.63 II + 0.96 I, 164 meioctyes studied) always at metaphase I (Fig. 2A-D) and anaphase I (all cells were cytologically balanced – 11/11 separation of chromosomes). This result is in conformity to the chromosome number observed in root tip mitosis, and therefore chromosome number in the species is 2n = 22 as suggested earlier (n = 11, x = 6) by Bhattacharya and Datta.

REFERENCES


Table 1. Karyomorphological details in *U. picta*

<table>
<thead>
<tr>
<th>Chromosomes</th>
<th>No. of Chromosomes</th>
<th>Length (µm)</th>
<th>F%</th>
<th>Cetromeric nature</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>2</td>
<td>2.625</td>
<td>40.27</td>
<td>Metacentric</td>
</tr>
<tr>
<td>A1</td>
<td>2</td>
<td>2.08</td>
<td>37.84</td>
<td>Sub-metacentric</td>
</tr>
<tr>
<td>B</td>
<td>4</td>
<td>1.19 – 1.145</td>
<td>47.06-48.47</td>
<td>Metacentric</td>
</tr>
<tr>
<td>C</td>
<td>2</td>
<td>1.105</td>
<td>23.08</td>
<td>Acrocentric</td>
</tr>
<tr>
<td>D</td>
<td>2</td>
<td>0.935</td>
<td>32.62</td>
<td>Sub-acrocentric</td>
</tr>
<tr>
<td>E</td>
<td>8</td>
<td>0.65 – 0.81</td>
<td>45.21 – 48.05</td>
<td>Metacentric</td>
</tr>
<tr>
<td>F</td>
<td>2</td>
<td>0.59</td>
<td>15.25</td>
<td>Acrocentric</td>
</tr>
</tbody>
</table>

Fig 1: Metaphase chromosomes of *U. picta* (2n = 22). A- Karyomorphology (secondary constrictions marked: →). B- Idiogram. Scale bar = 2.0 µm.
Fig 2: Meiotic metaphase I. A-C- 11 II; D- 10 II + 2 I (univalents marked). Scale bar = 10.0 μm

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