Karyological Studies in Some African Species of the Genus Sesbania (Fabaceae)

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Summary Karyological criteria of 15 African species of Sesbania, including five new records, are presented. A diploid number of 2n=12 (x=6) has been found in 12 species, 2n=14 (x=7) in one species and a tetraploid number of 2n=4x=24 was recorded in S. formosa and S. grandiflora. The karyotype in the examined species is symmetric. However, considerable variation exists in chromosome size among the species studied.

The result of the present study confirm that x=6 may be considered the basic number in the genus. Our observations further indicate that Sesbania is a primitive genus in its tribe Robinieae from which other genera may have evolved through aneuploid and polyploid changes.

The genus Sesbania belongs to tribe Robinieae of the family Fabaceae and comprises about 60, mostly annual, species distributed throughout tropical and subtropical regions, 32 in Africa, 10 in Asia and 8 in the New World (Forni-Martins et al. 1994). Species of this genus have a considerable economic importance in agriculture. They have been used as green manure for food crops, animal fodder and as fire wood (Evans and Rotor 1987). Species of Sesbania are also used to maintain soil productivity (Arumin et al. 1988) and to improve soil structure for wheat and rice cultivation (Ladha et al. 1989, Shioya and Ito 1990).

A number of cytological studies have been done for some species of Sesbania. Baquar and Akhtar (1968) have reported three different karyotypes with base numbers of x=6, 7 and 8. Bir et al. (1975) recorded tetraploid races with 2n=24 in S. sesban var. bicolor and 2n=28 in S. sesban var. picta. Lubis et al. (1981) reported that S. sesban and S. grandiflora perform infraspecific polymorphism in the shape of the mitotic chromosomes. Parihar and Zadoo (1987) studied the somatic karyotypes in diploid (2n=12) of S. aculeata and S. sesban and, diploid and tetraploid of S. grandiflora. Joshua and Bhatia (1989) presented the karyotypic variations in S. aculeata, S. rostrata, S. sesban and S. speciosa and recorded the tetraploid number of 2n=4x=24 in both S. aculeata and S. sesban. Heering and Hanson (1993) and Salimuddin (1993) reported similarities in karyotypes among three species and five species, respectively. From these and other reports (e.g. Goldblatt 1981b, 1984, 1985, 1988, Goldblatt and Johnson 1991) x=6 has been encountered in the vast majority of the species in the genus.

Phylogenetical speculations, based on chromosomal criteria, have also been concluded for some Sesbania species. Goldblatt (1981a) suggested x=10 and 11 for the tribe Robinieae and pointed out that x=6 of Sesbania is derived through aneuploid loss. However, Gill and Husaini (1985) concluded that the base number of x=6 is the primary base number of the genus. Lubis et al. (1981) postulated that S. sesban was most likely the ancestral type in the genus, while the other species have been derived from it by either chromosomal mutation or polyploidization.

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Studies on the impact of karyotypic data on the interspecific and phylogenetic relationships in the genus *Sesbania* is still limited. The present study deals with the use of some important chromosomal criteria to address the karyotype evolution in the genus as inferred from investigations on 15 African species.

Materials and methods

In addition to the Egyptian species *S. sericea* (Willd.) Link [= *S. pubescens* DC.] and *S. sesban* (L.) Merrill [= *S. aegyptiaca* (Poir.) Press], seeds of the species studied were kindly provided by International Livestock Centre for Africa (ILCA) genebank, the origin of the examined material is given in Table 1 and voucher specimens are kept at the Herbarium of Biological Sciences and Geology Department, Faculty of Education, Ain Shams University. Young and healthy root tips were taken from seedlings that had been germinated in Petri-dishes, pretreated for 3–4 hr in 0.05% colchicine solution at room temperature, washed and fixed in 3:1 absolute ethanol: glacial acetic acid overnight. The root tips were hydrolysed for 6 min in 1 M HCl at 60°C, washed and stained in Feulgen’s solution for 1–2 hr. Stained tips were squashed in a drop of 1% acetocarmine and permanent cytological preparations were made by mounting in Euparal. Cells with a good spread of chromosomes were photographed using a Carl-Zeiss Photomicroscope III and prints were enlarged to a magnification of 2500. Karyotypes of the species were made by cutting out individual chromosomes and arranging them in homologous pairs in order of their length and arm ratio. The chromosomes were classed by the arm ratio according to Levan *et al.* (1965).

Karyotype criteria were measured from 5 to 10 chromosome complements; these are mean length in μm, mean arm ratio (r-value) and the standard error (SE) of these parameters. Karyotype asymmetry has been estimated using the equation of Huziwara (1962) \[TF\% = \frac{\text{sum of short arm length}}{\text{sum of total chromosome length}} \times 100\]. The asymmetry based on the relations between the chromosome arms \(A_1\) and length \(A_2\) have been estimated for each species using the equations of Zarco (1986) as follows:

\[
A_1 = 1 - \frac{\sum b_i}{n_i} \quad \text{and} \quad A_2 = \frac{S}{\bar{x}}
\]

where \(A_1\) is the intrachromosomal asymmetry index ranging from zero to one. The equation is formulated in order to obtain lower values when chromosomes tend to be metacentric. \(n_i\) is the number of homologous chromosome pairs or groups. \(b_i\) is the average length for short arms in every homologous chromosome pair or group and \(B_i\) is the average length for their long arms. On the other hand, \(A_2\) is the interchromosomal asymmetry index, where \(\bar{x}\) = mean chromosome length (MCL) and \(S\) is its standard deviation.

Results and discussion

A summary of the cytological data of the species studied is given in Table 1 and their karyotypes are illustrated in Figs. 1–19. Among the 15 species studied a somatic chromosome number of \(2n=12\) with \(x=6\) has been found in 12 species, \(2n=14\) \((x=7)\) is recorded only in *S. pachycarpa* and a tetraploid number of \(2n=24\) was scored in *S. formosa* and *S. grandiflora*.

The counts of \(2n=12\) as recorded in each of *S. bispinosa*, *S. macrantha*, *S. macrocarpa*, *S. rostrata*, *S. sericea* and *S. sesban*; \(2n=14\) as recorded in *S. pachycarpa* and the record of \(2n=4x=24\) in *S. grandiflora* are in agreement with earlier reports on these species (Gillett 1963, Fedorov 1969, Goldblatt and Davidse 1977, Goldblatt, 1981b, 1984, 1985, 1988, Goldblatt and Johnson 1991),
Table 1.  Origin of the species studied and summary of their karyological criteria

<table>
<thead>
<tr>
<th>No.</th>
<th>Species</th>
<th>Origin</th>
<th>2n</th>
<th>x</th>
<th>MCL (µm) ± SE</th>
<th>Mean r-ratio ± SE</th>
<th>Karyotype asymmetry</th>
<th>Chromosome types</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>S. hispina</em> (Jacq.) W. Wright</td>
<td>Tanzania, Dar-el-Salaam</td>
<td>12</td>
<td>6</td>
<td>3.40±0.26</td>
<td>1.22±0.02</td>
<td>45.00 0.86 0.19</td>
<td>6</td>
</tr>
<tr>
<td>2</td>
<td><em>S. formosa</em> (F. Muell) Burbidge</td>
<td>Australia</td>
<td>24</td>
<td>6</td>
<td>2.70±0.37</td>
<td>1.53±0.12</td>
<td>40.74 0.94 0.33</td>
<td>11 1</td>
</tr>
<tr>
<td>3</td>
<td><em>S. goetzei</em> Harms.</td>
<td>Tanzania, Arusha</td>
<td>12</td>
<td>6</td>
<td>3.35±0.39</td>
<td>1.39±0.07</td>
<td>42.09 0.88 0.28</td>
<td>5   1</td>
</tr>
<tr>
<td>4</td>
<td><em>S. grandiflora</em> (L.) Poiret</td>
<td>Ethiopia, Addis-Ababa</td>
<td>24</td>
<td>6</td>
<td>3.38±0.30</td>
<td>1.81±0.10</td>
<td>35.50 0.95 0.22</td>
<td>4   8</td>
</tr>
<tr>
<td>5</td>
<td><em>S. greenwayi</em> J. B. Gill.</td>
<td>Tanzania, Coastal Region</td>
<td>12</td>
<td>6</td>
<td>2.95±0.29</td>
<td>1.47±0.06</td>
<td>40.34 0.89 0.24</td>
<td>6</td>
</tr>
<tr>
<td>6</td>
<td><em>S. hirtystyla</em> J. B. Gill.</td>
<td>Tanzania, Kisarawe</td>
<td>12</td>
<td>6</td>
<td>3.36±0.41</td>
<td>1.82±0.19</td>
<td>35.71 0.91 0.31</td>
<td>2   4</td>
</tr>
<tr>
<td>7</td>
<td><em>S. keniensis</em> J. B. Gill.</td>
<td>Tanzania, Arusha</td>
<td>12</td>
<td>6</td>
<td>2.64±0.21</td>
<td>1.53±0.18</td>
<td>39.33 0.89 0.20</td>
<td>2   4</td>
</tr>
<tr>
<td>8</td>
<td><em>S. macrantha</em> E. Phill &amp; Hutch.</td>
<td>Tanzania, Mara</td>
<td>12</td>
<td>6</td>
<td>3.29±0.35</td>
<td>1.60±0.09</td>
<td>38.30 0.90 0.26</td>
<td>3   3</td>
</tr>
<tr>
<td>9</td>
<td><em>S. microphylla</em> E. Phill &amp; Hutch.</td>
<td>Tanzania, Mwanza</td>
<td>12</td>
<td>6</td>
<td>3.29±0.39</td>
<td>1.55±0.07</td>
<td>38.91 0.89 0.29</td>
<td>4   2</td>
</tr>
<tr>
<td>10</td>
<td><em>S. pachycarpa</em> DC</td>
<td>Senegal</td>
<td>14</td>
<td>7</td>
<td>2.33±0.31</td>
<td>1.69±0.11</td>
<td>36.96 0.92 0.35</td>
<td>3   4</td>
</tr>
<tr>
<td>11</td>
<td><em>S. quadrata</em> J. B. Gill.</td>
<td>Tanzania, Tamby</td>
<td>12</td>
<td>6</td>
<td>3.78±0.51</td>
<td>1.38±0.09</td>
<td>42.33 0.88 0.33</td>
<td>6</td>
</tr>
<tr>
<td>12</td>
<td><em>S. rostrata</em> Brem. &amp; Obem.</td>
<td>Tanzania, Lake Rukwa</td>
<td>12</td>
<td>6</td>
<td>2.72±0.17</td>
<td>1.52±0.12</td>
<td>40.07 0.89 0.15</td>
<td>3   3</td>
</tr>
<tr>
<td>13</td>
<td><em>S. sericea</em> (Willd.) Link</td>
<td>Tanzania, Dar-el-Salaam</td>
<td>12</td>
<td>6</td>
<td>2.85±0.29</td>
<td>1.57±0.16</td>
<td>38.60 0.82 0.25</td>
<td>5   1</td>
</tr>
<tr>
<td>14</td>
<td><em>S. sericea</em> (Willd.) Link</td>
<td>Egypt, Cairo</td>
<td>12</td>
<td>6</td>
<td>2.21±0.39</td>
<td>2.12±0.21</td>
<td>31.67 0.92 0.43</td>
<td>2   4</td>
</tr>
<tr>
<td>15</td>
<td><em>S. seshan</em> (L.) Marr.</td>
<td>Tanzania, Chamloroma</td>
<td>12</td>
<td>6</td>
<td>3.58±0.40</td>
<td>1.67±0.10</td>
<td>37.99 0.90 0.27</td>
<td>2   4</td>
</tr>
<tr>
<td>16</td>
<td><em>S. seshan</em> (L.) Marr.</td>
<td>Rwanda, Kigali</td>
<td>12</td>
<td>6</td>
<td>4.20±0.50</td>
<td>1.41±0.20</td>
<td>42.38 0.88 0.29</td>
<td>1   4 2</td>
</tr>
<tr>
<td>17</td>
<td><em>S. seshan</em> (L.) Marr.</td>
<td>Tanzania, Mara</td>
<td>12</td>
<td>6</td>
<td>3.21±0.35</td>
<td>1.80±0.14</td>
<td>36.14 0.91 0.27</td>
<td>3   3</td>
</tr>
<tr>
<td>18</td>
<td><em>S. seshan</em> (L.) Marr.</td>
<td>Egypt, Cairo</td>
<td>12</td>
<td>6</td>
<td>3.39±0.44</td>
<td>1.31±0.08</td>
<td>42.77 0.88 0.32</td>
<td>1   5</td>
</tr>
<tr>
<td>19</td>
<td><em>S. speciosa</em> Taubert</td>
<td>Tanzania, Mwanza</td>
<td>12</td>
<td>6</td>
<td>3.21±0.23</td>
<td>1.37±0.10</td>
<td>42.68 0.88 0.18</td>
<td>1   5</td>
</tr>
</tbody>
</table>

2n = Diploid chromosome; x = basic chromosome number, MCL = mean chromosome length; SE = standard error, r-ratio = arm ratio TF% = total form percentage; A1 = intrachromosomal asymmetry index; A2 = interchromosomal asymmetry index; M = metacentric chromosomes; m = metacentric chromosomes and sm = submetacentric chromosomes.
while the count of $2n=12$ recorded in both *S. goetzei* and *S. keniensis* has previously been reported by Heering and Hanson (1993). On the other hand, the count of $2n=12$ recorded in the present study; for each of *S. greenwayi*, *S. hirtistyla*, *S. microphylla* and *S. quadrata* and the presence of $2n=4x=24$ in *S. formosa*, are new records for the species. The records of diploid chromosome number in all species studied; except two species; is in accordance with the statistics given by Bandel (1974) and Goldblatt (1981a) indicating that polyploidy is not common in the genus.

Mean chromosome length (MCL) values varies between species (Table 1). The highest value (4.20±0.50 μm) is recorded in *S. sesban* No. 16, while the lowest value (2.21±0.39 μm) is found in *S. sericea* No. 14. Short chromosomes was also recorded in *S. pachycarpa* (MCL=2.33±0.31 μm),
S. keniensis (2.64±0.21 μm), S. formosa (2.70±0.37 μm), S. rostrata (2.72±0.17 μm) and S. greenwayi (2.95±0.29 μm) while in the remaining species, chromosomes with MCL values ranging between 3 and 4 μm have been observed. The karyotypes of the examined species are considerably symmetric with regard to chromosome length, the most variable chromosomes in length are found in S. quadrata (SE of MCL=0.51 μm), whereas the most similar chromosomes are scored in S. rostrata (SE of MCL=0.17). The variation in length among chromosomes of the species studied is also reflected in the values of A₂. In general, higher A₂ values are scored in species with higher degrees of variation in length.

Most of the species studied have karyotypes comprised of metacentric to submetacentric chromosomes as indicated by their mean r-values, whereas the lowest value (1.22±0.02) is recorded in S. bispinosa, the highest value (2.12±0.21) is found in S. sericea No. 14. The degree of karyotype
asymmetry as indicated by TF%-values ranges between 31.67% in S. sericea No. 13 and 45% in S. bispinosa. SE of mean r-values also indicates the low degree of karyotype asymmetry in the majority of the species studied. A1 values show the high degree of karyotype symmetry in most species (Table 1).

The uniformity of the basic chromosome number \((x=6)\) in all; except S. pachycarpa; and the similarity among the species studied in karyotypic criteria, particularly in karyotype symmetry, are in agreement with the results of some previous studies (Parihar and Zadoo 1987, Joshua and Bhatia 1989, Salimuddin 1993, Vijayakumar and Kuriachan 1995). The similarity in karyotype features in the genus is reflected in the possibility of hybridization between the three species S. goetzei, S. keniensis and S. sesban (Heering and Hanson 1993).

Senn (1938) had reported that the basic chromosome number \((x=8)\) is the most frequent number in the subfamily Faboideae. He postulated that other numbers arose as a result of aneuploid loss or gain or duplication of this number. However, Goldblatt (1981a) suggested \(x=14\) for the subfamily and \(x=10\) or 11 for tribe Robinieae. He noted that the presence of only \(x=6\) and 7 in Sesbania is correlated with its special situation in the tribe; where \(x=8, 9, 10\) and 11 are the base numbers of other genera; and postulated that \(x=6\) or 7 is derived through aneuploid reduction processes.

Occurrence of \(x=6\) and 7 in the present study, and the presence of \(x=5, 6, 7\) and 8 in the previous reports on Sesbania, indicates that aneuploidy is common in the genus. This observation is supported by the score of \(2n=10, 12, 14, 16, 24\) and 28 in S. sesban and of \(2n=12, 13, 14, 16\) and 24 in S. bispinosa. However, this may not be considered to support the view of Goldblatt (1981a) that Sesbania is derived through aneuploid reduction from \(x=10, 11\) but can indicate that other genera in the tribe are derived from a primary base number of \(x=6\) as found in Sesbania. Within this genus \(x=6\) is found in the majority of species (66.67%). Polyploids derived from this number are recorded in 11.11% of the species whereas the haploid number of \(n=5, 10, 11, 13, 14\) and 16 are reported in the remaining species. From this statistics and the results of the present study, \(x=6\) may be confirmed as the basic number from which other numbers are derived by aneuploid changes of the diploid or polyploid numbers.

Stebbins (1974) and Moore (1987) had reported that the karyotype is varied in the degree of symmetry among the plant genera and species. Symmetrical karyotypes are considered more primitive than asymmetrical ones. The evolution of the latter from the former has been recorded in several genera, for example Crepis (Babcock 1947), Clarkia (Lewis and Lewis 1955), Leontodon (Rousi 1973), Crotalaria (Gupta and Gupta 1978), Phaseolus (Sarbhoy 1980) and several others. In South Indian material of the genus Sesbania, namely S. bispinosa, S. grandiflora, S. procumbens and S. sesban, Vijayakumar and Kuriachan (1995) reported karyotype features similar to those recorded in the present study. These authors argued that evolution of karyotype may be inferred from symmetry to asymmetry as a result of pericentric inversion or unequal translocation. However, in the light of the similarity in karyotype asymmetry among the 15 species studied here and the reports of karyotype homology in the genus (Salimuddin 1993) and the findings that interspecific hybridization is easily possible with little chromosomal irregularities (Heering and Hanson 1993) their view needs further support.

Based on the considerable interspecific similarity in karyotype criteria, recorded in the present study, and in previous reports (Baquar and Akhtar 1968, Bir et al. 1975, Lubis et al. 1981, Parihar and Zadoo 1987, Joshua and Bhatia 1989, Salimuddin 1993, Vijayakumar and Kuriachan 1995), interspecific crossing compatibility reported by Datta and Bagchi (1971) and Heering and Hanson (1993), it can be concluded that the genus Sesbania is a natural primitive monophyletic group in the tribe Robinieae from which other genera could have been evolved.
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References


