Cytotaxonomical Studies in South Indian Apiaceae

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The family, Apiaceae consists of approximately 200 genera and 2900 species in the world (Lawrence 1974) and in South India, there are 16 genera and about 38 species much widespread in the plains as well as in the hills (Gamble 1967). Most of the species of South India has not been cytologically studied so far. Even where the chromosome numbers have been determined (Fedorov 1974), the knowledge concerning the details of karyotype pertaining to chromosome measurements, which are vital to the understanding of karyological affinities, is still meagre. It was, therefore, felt that detailed and critical karyological work in this family was desirable. In the present communication, cytological studies have been made in 21 species coming under 11 genera.

Materials and methods

The wild species in this present investigation were collected from different localities of South Indian plains and hills, particularly Ooty, Kodaikanal, Courtallum and Tirupathi, the voucher specimens of which have been deposited in the hortis siccus of the Botany Department, Annamalai University. The seeds of *Daucus carota* were obtained from M/S Soundarya Nursery, Teynampet, Madras. Some of the species were identified with the help of the officials of Government Botanical Survey of India, Southern Circle, Coimbatore.

For the study of the somatic chromosomes, several squash methods in vogue have been tried as the chromosomes of most of the taxa happen to be rather small. The exercised root tips were pretreated in 0.002% *n* hydroxyquinoline kept at 4°C for 3 hours. After thorough washing in distilled water the root tips were fixed in 1:3 acetic alcohol for a period of 24 hours. Then, they were squashed following the schedule of iron alum haematoxylin squash technique described by Marimuthu and Subramaniam (1960). From the diagram drawn with camera lucida at stage level the chromosome measurements pertaining to short/long arms were made with a standardized scale improvised for the purpose, the details of which are presented in Table 3. For the assessment of the morphology of the chromosomes, twelve metaphase plates in each species were studied critically for the sake of confirmation.

Observations

In all the three species of *Hydrocotyl* studied namely 1) *H. javanica* 2) *H. conferta* and 3) *H. rotundifolia*, the somatic chromosome number is $2n=18$. Sharma and

Table 1. The present and previous records of chromosome numbers

Taxa studied	Present record of chromosome number 2n	Previous report of chromosome number (2n), author and year
<i>Hydrocotyle javanica</i> Thunb.	18	72, Sharma and Bhattacharya 1959 96, Liu <i>et al.</i> 1961 24, Liu <i>et al.</i> 1961
<i>H. conferta</i> Wt.	18*	_____
<i>H. rotundifolia</i> Roxb.	18*	_____
<i>Centella asiatica</i> Urban	18	18, Sharma, A. K. and Ghosh 1954, Sharma and Bhattacharyya 1950, 1960 22, Sharma and Sharma 1957
<i>Pimpinella pulneyensis</i> Gamble	18*	_____
<i>P. candolleana</i> W. and A.	18*	_____
<i>Heracleum rigens</i> , Wall	40*	_____
<i>H. ceylanicum</i> Gardn.	44*	_____
<i>H. sprengelianum</i> W. and A.	44*	_____
<i>H. courtallense</i> Gamble	40*	_____
<i>H. hookerianum</i> W. and A.	46*	_____
<i>H. aquilegifolium</i> C. B. Clarke	40*	_____
<i>H. candolleana</i> Gamble	40*	_____
<i>H. pedatum</i> Wt.	46*	_____
<i>Carum copticum</i> Benth	18	18, Shah, 1953, Sharma and Ghosh 1954,
<i>Trachyspermum roxburghianum</i>	18	18, Sharma and Ghosh 1954,
<i>Cuminum cyminum</i> L.	18	14, Sharma and Ghosh 1954,
<i>Coriandrum sativum</i> L.	22	22, Sharma and Ghosh 1954, Suzuka 1956, Sharma and Datta 1957, Sharma and Battacharyya 1959, Bell and Constance 1960, 1966
<i>Foeniculum vulgare</i> Mill	22	22, Sharma and Ghosh 1954, Bell and Constance 1957, Mitsukuri and Kurakori 1959, Sharma and Battacharyya 1959, Carter 1960, Raghuvanshi and Joshi 1966
<i>Anethum graveolens</i> L.	22	22, Reese 1950, Suzuka 1956
<i>Daucus carota</i> L.	18	18, Sharma and Ghosh 1954, Schwanity and Pirson 1955, Sharma and Bhattacharyya 1959, Bell and Constance 1960, Mulligam 1961

* First record of somatic chromosome numbers in the respective species.

Bhattacharyya (1959) have recorded $2n=72$ chromosomes and Liu *et al.* (1961), $2n=96$ and $2n=24$ chromosomes respectively in *H. javanica*. This shows that the populations in different parts of the world would reveal aneuploid and euploid variations in this species. In *Centella asiatica* Sharma and Ghosh (1954) and Sharma and Bhattacharyya (1959 and 1960) have shown $2n=18$ chromosomes and

the present study confirms this number for this species. On the other hand, Sharma and Sharma (1957) have again shown $2n=22$ chromosomes in this species and perhaps, this latter population may be a different cytotype. In *Pimpinella pulneyensis*, *P. candolleana*, *Carum copticum*, *Trachyspermum roxburghianum* and *Cuminum cyminum*, the somatic chromosome number is $2n=18$. The population of *C. cyminum* studied by Sharma and Ghosh (1954) shows $2n=14$ chromosomes.

Among the species of *Heracleum* studied, 3 numbers of somatic chromosomes have been observed, that is, $2n=40$, $2n=44$ and $2n=46$. Besides the chromosomes are also larger in size, when compared to the chromosomes of other species included in the present investigation.

In *Coriandrum sativum*, *Foeniculum vulgare* and *Anethum graveolens*, there are $2n=22$ chromosomes in the root tip cells and this number is confirmed by the

Figs. 1-21. 1, *Hydrocotyle javanica* $2n=18$. 2, *H. conferta* $2n=18$. 3, *H. rotundifolia* $2n=18$. 4 and 5, *Centella asiatica*, prophase and metaphase $2n=18$. 6, *Pimpinella pulneyensis* $2n=18$. 7, *P. candolleana* $2n=18$. 8, *Heracleum rigens* $2n=40$. 9, *H. ceylanicum*, prophase $2n=44$. 10, *H. sprengelianum*, prometaphase $2n=44$. 11, *H. courtalense* $2n=40$. 12, *H. hookerianum* $2n=46$. 13, *H. aquilegifolium* $2n=40$. 14, *H. pedatum* $2n=46$. 15, *Carum copticum* $2n=18$. 16, *Trachyspermum roxburghianum* $2n=18$. 17, *Cuminum cyminum* $2n=18$. 18, *Foeniculum vulgare* $2n=22$. 19, *Coriandrum sativum* $2n=22$. 20, *Anethum graveolens* $2n=22$. 21, *Daucus carota* $2n=18$.

Table 2. Particulars about the 2n number (1), range of size (2), secondary constricted (3), submedian (4), median (5), subterminal (6), absolute, total chromosome length (7) and average chromosome length (8) of various species studied

Taxa	1	2	3	4	5	6	7	8
<i>Hydrocotyle javanica</i>	18	1.6 to 2.6	2	4	6	6	35.6	1.97
<i>H. conferta</i>	18	1.4 to 2.4	2	—	8	8	34.4	1.91
<i>H. rotundifolia</i>	18	1.2 to 2.0	2	—	10	6	27.2	1.51
<i>Centella asiatica</i>	18	1.5 to 2.6	4	—	8	6	34.2	1.88
<i>Pimpinella pulneyensis</i>	18	1.5 to 2.6	2	2	8	6	35.0	1.89
<i>P. candolleana</i>	18	1.5 to 2.6	4	—	10	4	36.4	2.02
<i>Heracleum rigens</i>	40	1.5 to 2.8	6	—	24	10	76.2	1.91
<i>H. candolleianum</i>	40	2.0 to 4.0	6	2	16	16	110.6	2.77
<i>H. pedatum</i>	46	2.0 to 5.0	8	6	16	16	140.6	3.06
<i>Carum copticum</i>	18	1.7 to 2.8	4	—	8	6	36.8	2.04
<i>Trachyspermum roxburghianum</i>	18	1.5 to 2.6	2	2	8	6	34.4	1.91
<i>Cuminum cyminum</i>	18	1.6 to 2.6	4	4	6	4	37.6	2.08
<i>Coriandrum sativum</i>	22	1.4 to 2.3	4	2	6	10	39.6	1.80
<i>Anethum graveolens</i>	22	1.5 to 3.0	4	4	4	10	49.4	2.23
<i>Foeniculum vulgare</i>	22	1.6 to 2.6	4	—	10	8	43.2	1.95
<i>Daucus carota</i>	18	2.0 to 4.0	2	4	6	6	52.0	2.88

previous studies of chromosome numbers in these species (Fedorov 1974). In *Daucus carota*, as well, the previous studies confirm the present record of somatic chromosome number ($2n=18$). The karyotypes of the species studied are furnished in Figures 1 to 21. The chromosomes of most of the taxa of Apiaceae are rather small. The size of them ranges from 1.0 to 5.0 μm . Based on the length, the chromosomes have been grouped into shorter, short, medium sized and long chromosomes. Under each of these groups, the following categories of chromosomes have been recognized.

Table 3. Particulars about karyotype analysis of the species studied

Chromosome type	No.	IC	Long arm in μm	Short arm in μm	Ratio L/S	Total chr. length	Relative chr. length
1 <i>Hydrocotyle javanica</i>							
F	2	42.31	1.1	1.1	—	2.6	7.30
G	4	27.27	1.4	0.6	2.33	2.2	6.18
H	6	45.00	0.9	0.9	—	2.0	5.62
K	6	12.5	1.2	0.2	6.00	1.6	4.49
2 <i>Hydrocotyle conferta</i>							
F	2	41.67	1.0	1.1	—	2.4	6.98
H	4	45.45	1.0	1.0	—	2.2	6.40
H	4	45.00	0.9	0.9	—	2.0	5.81
K	4	11.11	1.4	0.2	7.0	1.8	5.23
K	4	14.29	1.0	0.2	5.0	1.4	4.07
3 <i>Hydrocotyle rotundifolia</i>							
F	2	40.00	0.8	0.8	—	2.0	7.35
J	10	43.75	0.7	0.7	—	1.6	5.88
K	6	16.67	0.8	0.2	4.0	1.2	4.41

Table 3. (cont.)

Chromosome type	No.	IC	Long arm in μm	Short arm in μm	Ratio L/S	Total chr. length	Relative chr. length
4 <i>Centella asiatica</i>							
F	4	42.31	1.1	1.1	—	2.6	7.60
H	2	45.00	0.9	0.9	—	2.0	5.85
J	6	44.44	0.8	0.8	—	1.8	5.26
K	6	13.33	1.1	0.2	5.5	1.5	4.39
5 <i>Pimpinella pulneyensis</i>							
F	2	42.31	1.1	1.1	—	2.6	7.43
G	2	25.00	1.6	0.6	2.67	2.4	6.86
H	8	45.00	0.9	0.9	—	2.0	5.71
K	6	13.33	1.1	0.2	5.50	1.5	4.29
6 <i>Pimpinella candolleana</i>							
F	4	42.31	1.1	1.1	—	2.6	7.14
H	10	45.00	0.9	0.9	—	2.0	5.49
K	4	13.33	1.1	0.2	5.50	1.5	4.12
7 <i>Heracleum rigens</i>							
F	4	42.86	1.2	1.2	—	2.8	3.67
F	2	42.31	1.1	1.1	—	2.6	3.41
H	8	45.00	0.9	0.9	—	2.0	2.62
J	16	44.44	0.8	0.8	—	1.8	2.36
K	10	13.33	1.1	0.2	5.50	1.5	1.97
8 <i>Heracleum candolleianum</i>							
A	2	35.00	2.2	1.4	1.57	4.0	3.62
C	4	31.43	2.0	1.1	1.82	3.5	3.16
D	2	37.14	2.0	1.3	1.53	3.5	3.16
E	10	46.67	1.4	1.4	—	3.0	2.71
H	6	46.15	1.2	1.2	—	2.6	2.35
I	8	8.00	2.1	0.2	10.5	2.5	2.26
I	8	10.00	1.6	0.2	8.0	2.0	1.81
9 <i>Heracleum pedatum</i>							
A	2	46.00	2.3	2.3	—	5.0	3.56
B	6	25.00	2.6	1.0	2.60	4.0	2.84
D	6	27.78	2.4	1.0	2.40	3.6	2.56
E	16	46.67	1.4	1.4	—	3.0	2.13
I	10	8.00	2.1	0.2	10.50	2.5	1.78
I	6	10.00	1.6	0.2	8.00	2.0	1.42
10 <i>Carum copticum</i>							
F	2	42.86	1.2	1.2	—	2.8	7.61
F	2	42.31	1.1	1.1	—	2.6	7.07
H	8	45.00	0.9	0.9	—	2.0	5.43
K	6	11.76	1.0	0.2	6.50	1.7	4.62
11 <i>Trachyspermum roxburghianum</i>							
F	2	42.31	1.1	1.1	—	2.6	7.56
G	2	30.43	1.4	0.7	2.00	2.3	6.69
H	6	45.00	0.9	0.9	—	2.0	5.81
J	2	44.44	0.8	0.8	—	1.8	5.23
K	6	13.33	1.1	0.2	5.50	1.5	4.36

Table 3. (cont.)

Chromosome type	No.	IC	Long arm in μm	Short arm in μm	Ratio L/S	Total chr. length	Relative chr. length
12 <i>Anethum graveolens</i>							
C	4	33.33	1.6	1.0	1.60	3.0	7.98
D	4	33.33	1.8	1.0	1.80	3.0	7.98
H	4	46.15	1.2	1.2	—	2.6	6.91
K	10	13.33	1.1	0.2	5.50	1.5	3.99
13 <i>Foeniculum vulgare</i>							
F	4	42.31	1.1	1.1	—	2.6	6.57
H	10	45.00	0.9	0.9	—	2.0	5.05
K	8	12.50	1.2	0.2	6.0	1.6	4.04
14 <i>Cuminum cyminum</i>							
F	4	42.31	1.1	1.1	—	2.6	5.26
G	4	22.73	1.5	0.5	3.0	2.2	4.45
H	6	45.00	0.9	0.9	—	2.0	4.05
K	4	12.50	1.2	0.2	6.0	1.6	3.24
15 <i>Coriandrum sativum</i>							
C	4	21.74	1.2	0.5	—	2.3	5.32
D	2	22.73	1.5	0.5	3.0	2.2	5.09
H	6	45.00	0.9	0.9	—	2.0	4.63
K	10	14.29	1.0	0.2	5.0	1.4	3.24
16 <i>Daucus carota</i>							
A	2	25.00	2.6	1.0	2.60	4.0	7.69
D	4	37.14	2.0	1.3	1.53	3.5	6.73
E	6	46.67	1.4	1.4	—	3.0	5.77
I	6	10.00	1.6	0.2	8.00	2.0	3.85

Long (4.0 to 5.0 μm)

Type A : Chromosome with median primary and subterminal secondary centromeres.

Type B : Chromosome with submedian primary and subterminal secondary centromeres.

Medium (3.0 to 3.9 μm).

Type C : Chromosome with submedian primary and subterminal secondary centromeres.

Type D : Chromosome with submedian centromere.

Type E : Chromosome with median centromere.

Short (2.0 to 2.9 μm).

Type F : Chromosome with median primary and subterminal secondary centromeres.

Type G : Chromosome with submedian centromere.

Type H : Chromosome with median centromere.

Type I : Chromosome with subterminal centromere.

Shorter (1.0 to 1.9 μm)

Type J : Chromosome with median centromere.

Type K : Chromosome with subterminal centromere.

The total number of secondary constricted chromosomes has been found to

Fig. 22. Idiograms of various species studied.

vary from species to species. The present and previous records of chromosome numbers are given in Table 1. The details of chromosome measurements are given in Table 2. In this table the particulars about the karyotype analyses of the species and the descriptions of the various columns are as follows:

1. Chromosome number (2n).
2. Size range in millimicrons.
3. Number of chromosomes with secondary constrictions.
4. Number of chromosomes with submedian constrictions.
5. Number of chromosomes with median constriction.
6. Number of chromosomes with subterminal constriction.
7. Absolute chromosome length in millimicrons.
8. Average chromosome length in millimicrons.

From the karyotype analysis it is clear that all the species of Apiaceae included in the present investigation are characterised by asymmetrical karyotypes. The idiograms (Fig. 22) shows the karyotypes analyses of the various species studied.

Discussion

In 21 species of South Indian Apiaceae, detailed cytotaxonomical studies have been made, of which first record of chromosome number has been made in 12 species namely, *Hydrocotyle conferta*, *H. rotundifolia*, *pimpinella pulneyensis*, *P. candolleana*, *Heracleum rigens*, *H. ceylanicum*, *H. sprengelianum*, *H. courtallense*,

Table 4. Frequency distribution of variously sized chromosomes among the species studied

Taxa studied	A	B	C	D	E	F	G	H	I	J	K
1 <i>Hydrocotyle javanica</i>	—	—	—	—	—	2	4	6	—	—	6
2 <i>H. conferta</i>	—	—	—	—	—	2	—	8	—	—	4
3 <i>H. rotundifolia</i>	—	—	—	—	—	2	—	—	—	10	6
4 <i>Centella asiatica</i>	—	—	—	—	—	4	—	2	—	6	6
5 <i>Pimpinella pulneyensis</i>	—	—	—	—	—	2	2	8	—	—	6
6 <i>P. candolleana</i>	—	—	—	—	—	4	—	10	—	—	4
7 <i>Heracleum rigens</i>	—	—	—	—	—	6	—	8	—	16	10
8 <i>H. candolleum</i>	2	2	4	2	10	—	—	6	16	—	—
9 <i>H. pedatum</i>	2	6	—	6	16	—	—	—	16	—	—
10 <i>Carum copticum</i>	—	—	—	—	—	4	—	8	—	—	6
11 <i>Trachyspermum roxburghianum</i>	—	—	—	—	—	2	2	6	—	2	6
12 <i>Cuminum cyminum</i>	—	—	—	—	—	4	4	6	—	—	4
13 <i>Coriandrum sativum</i>	—	—	4	2	—	—	—	6	—	—	10
14 <i>Anethum graveolens</i>	—	—	4	4	—	—	—	4	—	—	10
15 <i>Foeniculum vulgare</i>	—	—	—	—	—	4	—	10	—	—	8
16 <i>Daucus carota</i>	2	—	—	4	6	—	—	—	6	—	—

Fig. 23. Histogram showing the absolute chromosome lengths of the species studied.

H. hookerianum, *H. aquinlegifolia*, *H. candolleum* and *H. pedatum*.

In the present study, when the karyotypes of 16 taxa were examined, it was obvious that karyotype asymmetry was of a high order. From the works of Löve and Löve (1942) on boreal plants, Babcock (1947) in *Crepis*, Sato (1942) in Liliaceae, Stebbins *et al.* (1953) in Cichoreae and Sharma and Chatterji (1964) in Araliaceae,

it is understood that chromosomes of uniform size with symmetrical karyotypes is a primitive feature. Therefore, the presence of high frequency of karyotypic asymmetry is an advanced character. Besides, excepting *Daucus carota* and species of *Heracleum* studied, all the other species studied possess smaller chromosomes and this is another advanced cytological character of this family.

The more commonly available somatic chromosome numbers of the family are $2n=16, 18, 20, 22, 24, 32, 36, 44, 48, 64$ and 72 . Of these, $n=8$ may be the primary basic number of the family and the other numbers, $9, 10, 11, 12$ and 24 are derived basic numbers and these show aneuploid variations. The numbers $2n=32, 36, 44, 48, 64$ and 72 are polyploids, particularly euploid derivatives during evolution. Therefore, it is concluded that both aneuploidy and euploidy together with karyotypic asymmetry play important role in the origin and evolution of the family, Apiaceae. It is further evidenced by the considerable variations of the absolute chromosome lengths of the various species under investigation as shown by histogram (Fig. 23) representation. Bell and Constance (1957, 1960), having studied a large number of species of Apiaceae concluded that aneuploid series appeared to occur in *Eremocharis*, *Eryngium*, *Oenanthe* and *Ptilimnium* and that polyploidy discovered in *Bowlesia* and confirmed in *Pimpinnella* would appear to play important roles in the origin and evolution of the family.

Summary

Karyotype analyses were made using root tip mitosis in 21 species of South Indian Apiaceae. First record of chromosome numbers have been made in *Hydrocotyle conferta* ($2n=18$), *H. rotundifolia* ($2n=18$), *Pimpinnella pulneyensis* ($2n=18$), *P. candolleana* ($2n=18$), *Heracleum rigens* ($2n=40$), *H. ceylanicum* ($2n=44$), *H. sprengelianum* ($2n=14$), *H. courtallense* ($2n=40$), *H. hookerianum* ($2n=46$), *H. aquilegifolium* ($2n=46$), *H. candolleianum* ($2n=40$) and *H. pedatum* ($2n=46$). The most commonly occurring somatic chromosome number is $2n=18$ and $2n=22$. It is evident from the present investigation and those data available from earlier studies in this family that aneuploidy and euploidy together with karyotypic asymmetry of chromosomes play important role in the origin and evolution of the species studied.

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