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Cytological Studies in the Genus *Medicago* Linn.

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Medicago belongs to the tribe Trifolieae of family Fabaceae. It is represented by about 100 species which are indigenous to subtropical and temperate regions, but restricted to northern hemisphere of Old World. Though a large number of species (about 95%) have been cytologically examined for chromosome counts, only few reports are available for karyotype studies (Fryer 1930, Lesins and Lesins 1961, 1963a, 1963b, Simon and Simon 1965). In view of this, a cytological study in the genus has been undertaken and the results of meiosis and root tip mitosis (including karyotypes) involving 18 species of *Medicago* are reported in this communication.

Material and methods

Seed material for different species of *Medicago* was obtained from different agencies through correspondence (Table 1). Plants were raised in pots and in flower beds at Meerut University Experimental Farm. Dormancy of seeds was broken by rubbing them with glass fibre paper. Voucher specimens were deposited in the Department of Ag. Botany, Meerut University, Meerut.

For meiosis, flower buds were collected in forenoon, fixed for at least 24 hours in Carnoy's fluid (absolute alcohol: chloroform: acetic acid, 6: 3: 1) and stored in 70% ethyl alcohol. Anthers were squashed in 2.0% acetocarmine.

For mitosis young and healthy root tips were pretreated with saturated solution of α-bromonaphthalene for about one and a half to two hours, fixed in acetic alcohol (1: 3) for 24 hours and squashed in 2.0% acetocarmine.

Photomicrographs and camera lucida drawings were made from temporary preparations and measurements (length and width of chromosomes) were made with the help of Olympus micrometer eye piece. TC1% and TF% were calculated as earlier done in *Crotalaria* (Gupta and Gupta 1978).

Results

1. *Meiosis*

Meiosis was studied in 14 species of *Medicago*. Data on chromosome numbers and chiasmata frequencies are presented in Table 1. Most of the species were diploids having $n=7$ or $n=8$ (Figs. 1–6) and only two species namely *M. sativa* and *M. scutellata* were tetraploids having $n=16$ (Figs. 7 and 8). Due to inadequacy of material and stickiness of chromosomes at metaphase I, chiasmata frequencies

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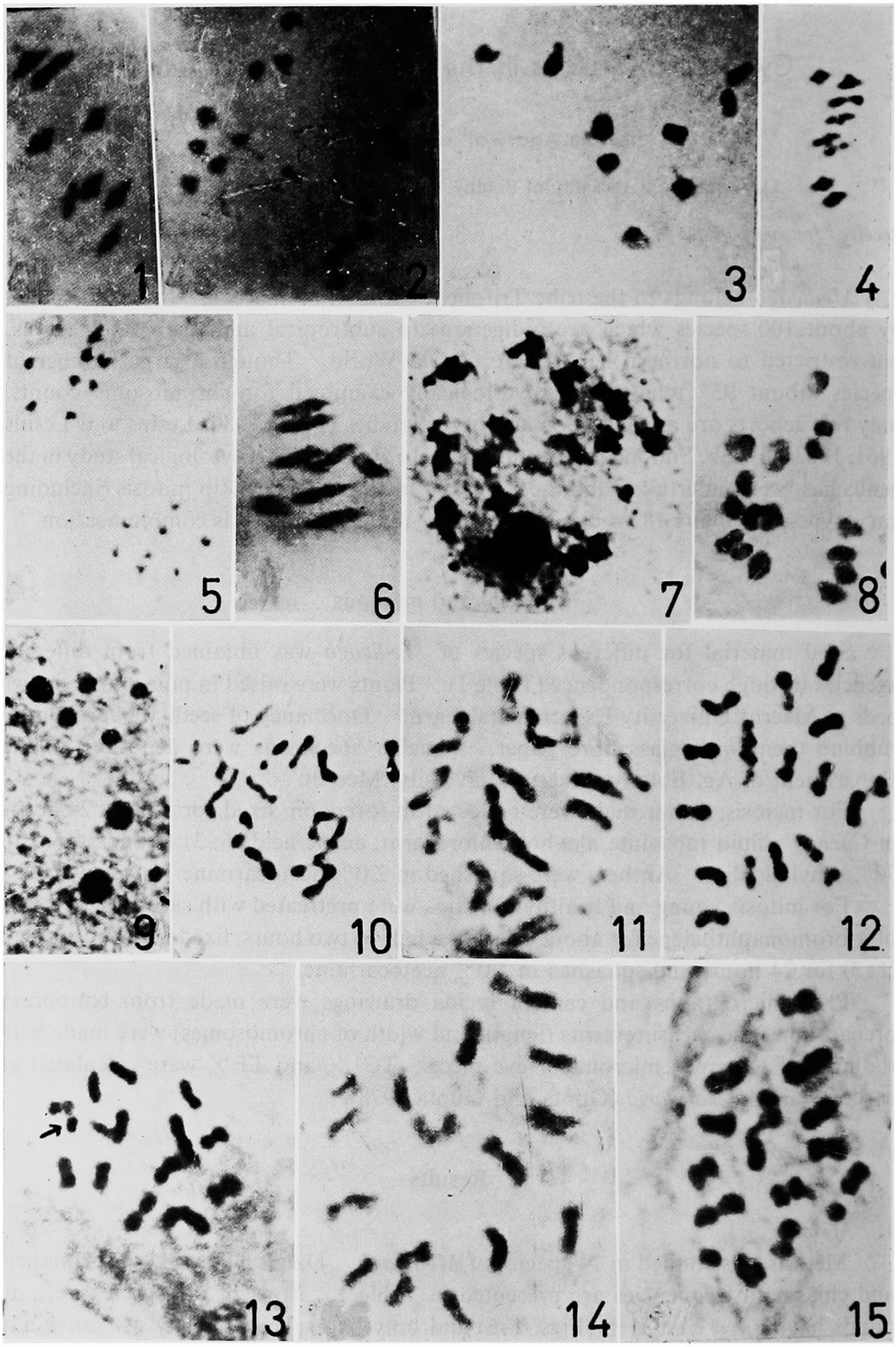
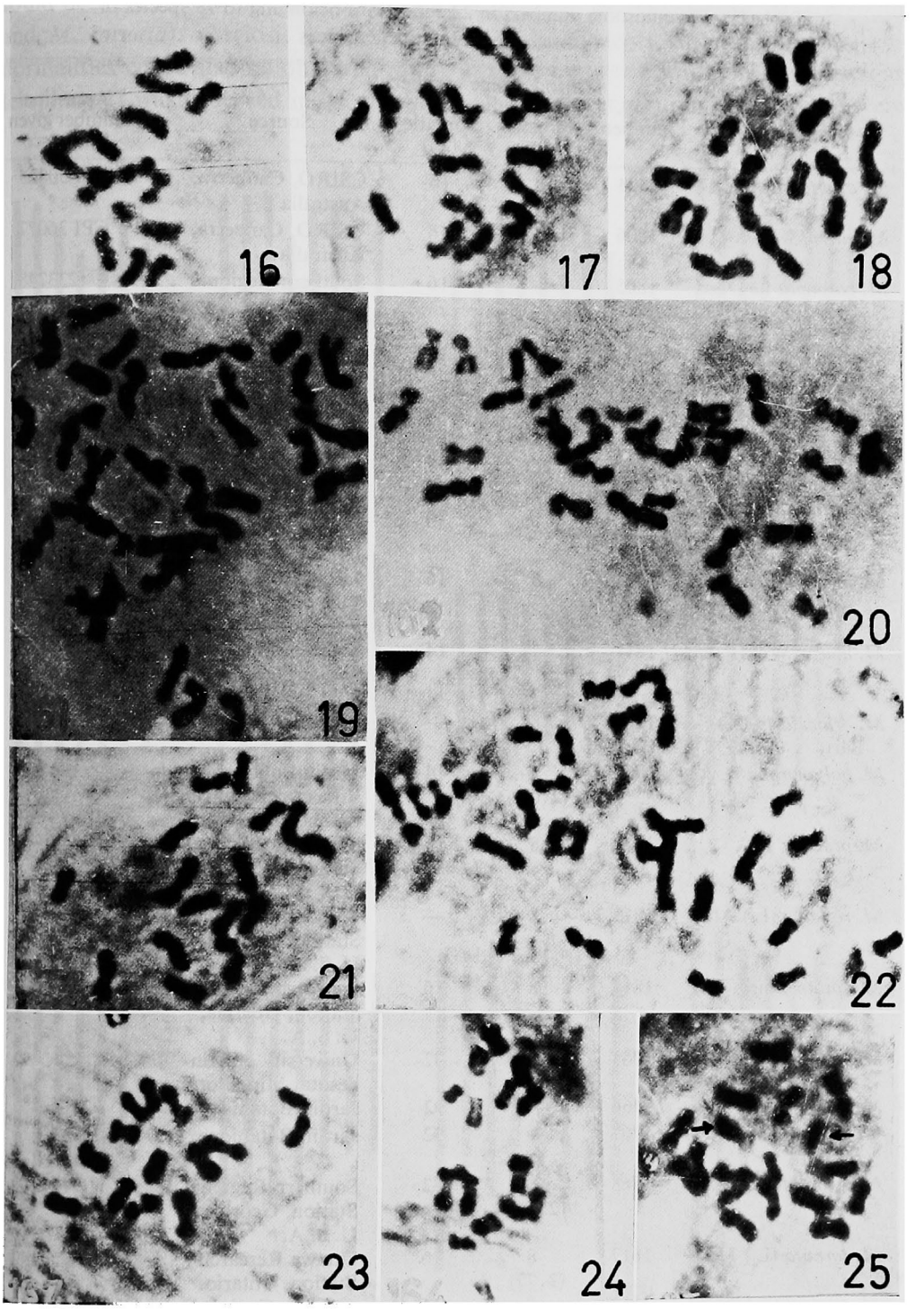


Figs. 1–15. 1–9: Meiosis in different *Medicago* species. 1, *M. arabica*, metaphase I, 8^{II}, 2, *M. arabica*, anaphase I, 8:8, 3, *M. intertexta* var. *ciliaris* (1808) metaphase I, $7^{II}+2^{I}$, 4, *M. lupulina*, metaphase I, 8^{II}, 5, *M. rigidula*, anaphase I, 8:8, 6, *M. rotata*, metaphase I, 8^{II}, 7, *M. sativa* (1765), diakinesis, 16^{II}, 8, *M. scutellata*, metaphase I, 16^{II}, 9, *M. turbinata*, telophase I, multinucleolate condition. 10–15: Mitotic metaphase plates in different *Medicago* species. 10, *M. arabica*, $2n=16$, 11, *M. coerulea*, $2n=16$, 12, *M. intertexta* var. *ciliaris* (1808), $2n=16$, 13, *M. littoralis*, $2n=14+1B$, 14, *M. lupulina*, $2n=16$, 15, *M. murex*, $2n=14$.



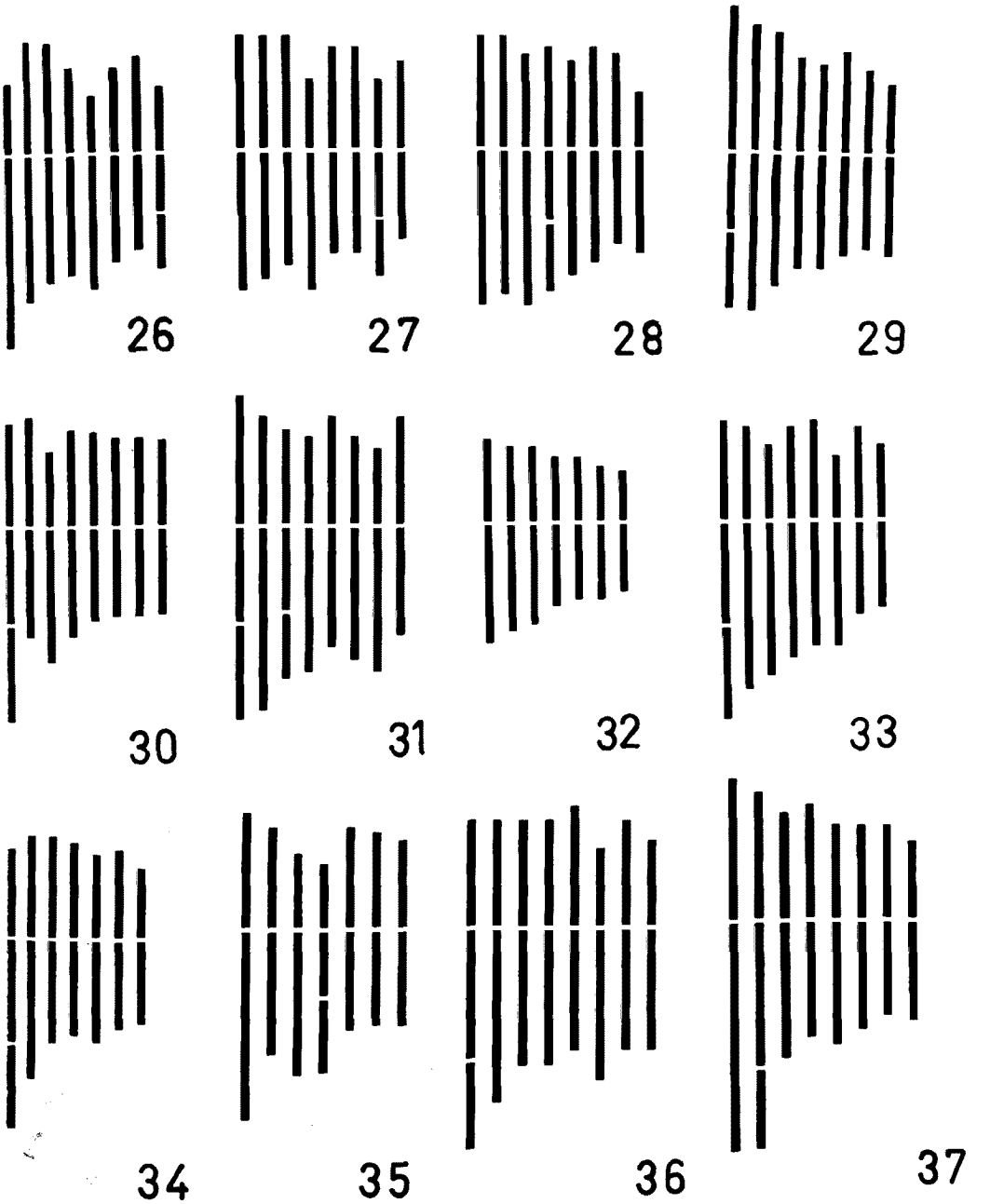
Figs. 16–25. 16, *M. polymorpha*, $2n=14$, 17, *M. praecox* $2n=16$, 18, *M. rotata*, $2n=16$, 19, *M. sativa* (1766) $2n=32$, 20, *M. sativa* subsp. *falcata*, $2n=32$, 21, *M. truncatula*, $2n=16$, 22, *M. scutellata* $2n=32$, 23, *M. tuberculata*, $2n=14$, 24, *M. tornata*, $2n=16$, 25, *M. tornata*, $2n=16+2B$.

Table 1. Chromosome numbers in 22 collections belonging to 18 species of *Medicago* along with their sources

Species	Coll. number	Chromosome number		Source	Accession number given by source
		Gametic (*n*)*	Somatic (2*n*)		
M. arabica L. Huds	1804	8 (2.00)	16	CSIRO, Canberra, Australia	CPI 50825
M. coerulea Less	1806	—	16	CSIRO, Canberra, Australia	CPI 36077
M. coronata L. Desr.	1805	—	16	Southern Regional Station, Georgia, U. S. A.	233248
M. intertexta L. Mill var. *ciliaris*	1808	8	16	CSIRO, Canberra, Australia	CPI 32818
M. intertexta L. Mill var. *ciliaris*	1809	—	16	RBG, Kew, England	171/07
M. intertexta L. Mill var. *intertexta*	1810	—	16	RBG, Kew, England	172/01
M. littoralis Rohde	1807	—	14	Southern Regional Station, Georgia, U. S. A.	255381
M. lupulina L.	1771	8	16	Jardin Botanique de la Ville et de l'Université Besançon, France.	
M. murex Willd.	1811	—	14	Southern Regional Station, Georgia, U. S. A.	197859
M. orbicularis (L.) Bart.	1761	8	—	RBG, Kew, England	120/13
M. polymorpha L.	1770	7 (2.04)	14	Jardin Botanique de la Ville et de l'Université, Besançon, France	
M. praecox DC.	1812	8	16	Southern Regional Station, Georgia, U. S. A.	212948
M. rigidula (L.) All.	1813	8	—	Southern Regional Station, Georgia, U. S. A.	
M. rotata Boiss	1814	8	16	Southern Regional Station, Georgia, U. S. A.	
M. sativa L.	1765	16	32	University of Minnesota, Minnesota	
M. sativa L.	1766	—	32	Jardin Botanique	
M. sativa L. subsp. *falcata*	1767	—	32	Jardin Botanique	
M. scutellata L.	1768	16 (2.18)	32	Southern Regional Station, Georgia, U. S. A.	197819
M. tornata (L.) Mill.	1817	8 (1.77)	16	Ottawa Research Station, Ontario.	M44
M. truncatula Gaertn.	1815	8	16	CSIRO, Canberra, Australia	V357
M. tuberculata Willd.	1816	—	14	Botanischer Garten Der, Leipzig	
M. turbinata (L.) All.	1772	8	—	RBG, Kew, England	30/01

* Figures in parentheses are chiasmata/bivalent.

could be worked out only in four species (*M. arabica*, *M. polymorpha*, *M. scutellata* and *M. tornata*). Meiosis was regular with only bivalents, although meiotic abnormalities could also be observed rarely in some cases. Formation of pentads at telophase II was observed in *M. tornata*. Univalents at metaphase I (Fig. 3) were



Figs. 26–37: Idiograms prepared from mitotic metaphase in different *Medicago* species. 26, *M. arabica*, 27, *M. coerulea*, 28, *M. coronata*, 29, *M. intertexta* var. *ciliaris* (1809), 30, *M. intertexta* var. *ciliaris* (1808), 31, *M. intertexta* var. *intertexta*, 32, *M. littoralis*, 33, *M. lupulina*, 34, *M. murex*, 35, *M. polymorpha*, 36, *M. praecox*, 37, *M. rotata*.

Table 2. Analysis of mitotic chromosomes in different diploid species of *Medicago* (In each species, the first row represents absolute length of chromosome in μ, the second row gives arm ratios; the third row gives relative chromosome length and the fourth row gives TCI%)

Species and collection no.	Chromosome pairs							
	1	2	3	4	5	6	7	8
M. arabica	2.83	2.83	2.55	2.21	2.07	2.07	2.07	1.86
(2*n*=16)	2.72	1.28	1.18	1.46	2.33	1.31	1.00	1.70
	100.00	100.00	90.24	78.05	73.17	73.17	73.17	65.85
	15.30	15.30	13.81	11.94	11.19	11.19	11.19	10.08
M. coerulea	2.76	2.62	2.48	2.28	2.21	2.21	2.07	1.93
(2*n*=16)	1.22	1.11	1.00	2.00	1.00	1.00	1.73	1.00
	100.00	95.00	90.00	82.50	80.00	80.00	75.00	70.00
	14.87	14.12	13.38	12.26	11.89	11.89	11.15	10.41
M. coronata	2.97	2.83	2.76	2.62	2.35	2.35	2.07	1.79
(2*n*=16)	1.39	1.28	1.67	1.37	1.42	1.12	1.00	1.89
	100.00	95.35	93.02	88.37	79.07	79.07	69.77	60.46
	15.03	14.34	13.99	13.29	11.89	11.89	10.49	9.09
M. intertexta	3.24	3.11	2.76	2.28	2.21	2.21	1.93	1.86
var. *ciliaris*	1.04	1.25	1.11	1.20	1.29	1.00	1.15	1.45
Coll. no. 1809	100.00	95.74	85.11	70.21	68.08	68.08	59.57	57.45
(2*n*=16+1B)	16.55	15.84	14.08	11.62	11.27	11.27	9.86	9.51
M. intertexta	3.17	2.35	2.21	2.21	2.00	1.93	1.93	1.86
var. *ciliaris*	1.88	1.00	1.91	1.13	1.00	1.00	1.00	1.00
Coll. no. 1808	100.00	73.91	69.56	69.56	63.04	60.87	60.87	58.70
(2*n*=16)	17.97	13.28	12.50	12.50	11.33	10.94	10.94	10.55
M. intertexta	3.45	3.17	2.62	2.55	2.48	2.42	2.35	2.35
var. *intertexta*	1.50	1.71	1.53	1.64	1.12	1.50	1.83	1.00
(2*n*=16+1B)	100.00	92.00	76.00	74.00	72.00	70.00	68.00	68.00
	16.13	14.84	12.26	11.94	11.61	11.29	10.97	10.97
M. littoralis	2.21	2.00	1.93	1.59	1.52	1.45	1.31	
(2*n*=14+1B)	1.46	1.42	1.33	1.30	1.20	1.33	1.37	
	100.00	90.62	87.50	71.87	68.75	65.62	59.37	
	18.39	16.67	16.09	13.22	12.64	12.07	10.92	
M. lupulina	3.24	2.90	2.55	2.55	2.48	2.07	2.07	1.79
(2*n*=16)	1.94	1.80	2.08	1.47	1.25	2.00	1.00	1.17
	100.00	89.36	78.72	78.72	76.60	63.83	63.83	55.32
	16.49	14.74	12.98	12.98	12.63	10.53	10.53	9.12
M. murex	2.97	2.62	2.21	2.07	2.00	1.93	1.66	
(2*n*=14)	2.07	1.37	1.00	1.00	1.23	1.00	1.18	
	100.00	88.37	74.42	69.77	67.44	65.12	55.81	
	19.20	16.96	14.29	13.39	12.95	12.50	10.71	
M. polymorpha	2.90	2.14	2.07	1.93	1.93	1.79	1.73	
(2*n*=14)	1.63	1.21	2.00	2.11	1.00	1.00	1.08	
	100.00	73.81	71.43	66.67	66.67	61.91	59.52	
	20.00	14.76	14.29	13.33	13.33	12.38	11.91	
M. praecox	3.11	2.69	2.35	2.35	2.35	2.21	2.21	2.00
(2*n*=16)	2.00	1.60	1.27	1.27	1.00	1.91	1.13	1.42
	100.00	86.67	75.55	75.55	75.55	71.11	71.11	64.44
	16.13	13.98	12.19	12.19	12.19	11.47	11.47	10.39

Table 2. Continued

Species and collection	no.	Chromosome pairs							
		1	2	3	4	5	6	7	8
M. rotata		3.59	3.38	2.35	2.21	2.07	1.93	1.79	1.73
($2n=16$)		1.60	1.72	1.27	1.00	1.31	1.15	1.00	1.27
		100.00	94.23	65.38	61.54	57.69	51.92	50.00	35.48
		18.84	17.75	12.32	11.59	10.87	10.15	9.42	9.06
M. tornata		2.28	2.28	2.07	2.07	1.93	1.79	1.73	1.52
($2n=16+0-2B$)		2.30	1.54	1.15	1.14	1.33	1.17	1.28	1.20
		100.00	100.00	90.91	90.91	84.35	78.79	75.76	66.67
		14.54	14.54	13.22	13.22	12.34	11.45	11.01	9.69
M. truncatula		3.38	2.76	2.48	2.42	2.28	2.07	1.86	1.66
($2n=16$)		1.23	1.67	1.00	1.33	2.00	1.00	1.25	1.00
		100.00	81.63	73.47	71.43	67.35	61.22	55.10	48.98
		17.88	14.60	13.14	12.77	12.04	10.95	9.85	8.76
M. tuberculata		2.55	2.48	2.35	2.14	1.66	1.66	1.52	
($2n=14$)		1.85	1.77	1.00	2.10	1.00	1.00	1.00	
		100.00	97.30	91.39	83.78	64.86	64.86	59.46	
		17.79	17.31	16.35	14.90	11.54	11.54	10.58	

invariably observed in *M. intertexta* var. *ciliaris* and multinucleolate condition was observed in *M. turbinata* at telophase I (Fig. 9).

2. *Mitosis and karyotypes*

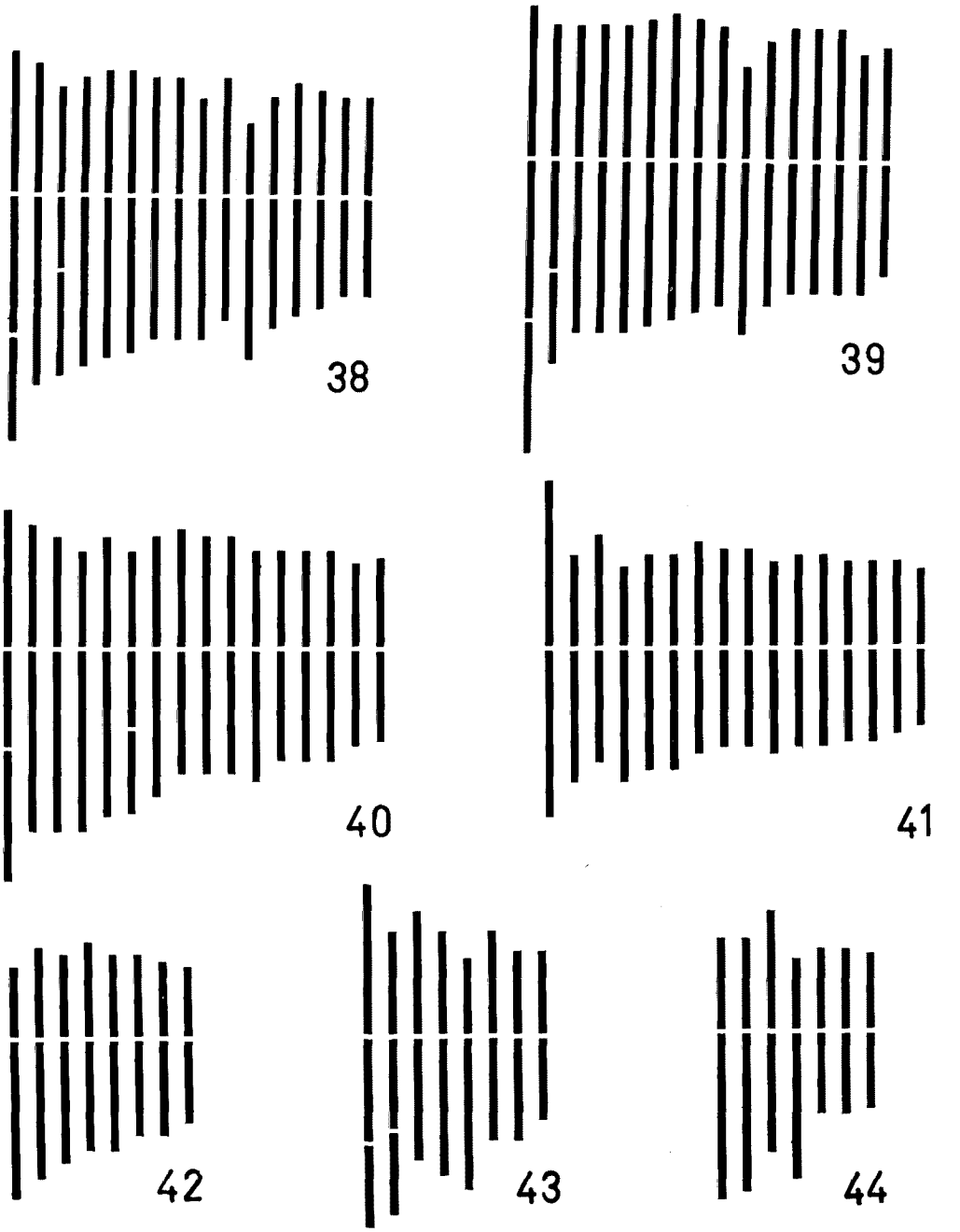
Mitosis was studied from 19 collections belonging to 15 species. Fifteen collections were diploids, and mainly had $2n=16$ except in four species (*M. littoralis*, *M. murex*, *M. polymorpha* and *M. tuberculata*) which had $2n=14$. Remaining four collections were tetraploids with $2n=32$. Data on chromosome measurements and other characteristics are presented in Tables 2–4. Corresponding mitotic metaphase plates and idiograms are presented in Figs. 10–25 and 26–44 respectively.

Chromosomes were designated in the descending order of length, as 1–8 in diploids and 1–16 in tetraploids. Depending upon their absolute length, chromosomes were classified into four categories, namely A=more than 4.0 μ, B=3.0 μ to 4.0 μ, C=2.0 μ–3.0 μ and D=less than 2.0 μ.

Discussion

In the present study sixteen species (eighteen collections) out of eighteen (twenty two collections) studied were diploids, of which twelve species had $2n=16$ and four species had $2n=14$. Remaining two species representing four collections were tetraploid with $2n=32$. With the help of different chromosome indices available, Agarwal (1979) prepared a chromosome atlas for the genus *Medicago* which suggests that the genus is quite variable with $2n=14$, 16, 18, 28, 32 and 48. It was also shown that 76.84% of the species examined by different workers exhibited $2n=16$ and that 27.36% of the species exhibited polyploidy with $2n=32$. The earlier published

data and present results suggest that $x=8$ is the predominant base number in the genus, although in eight species out of 95 examined so far, $x=7$ is known. As obvious from the earlier published results, polyploidy in the genus is not infrequent and has, therefore, played some role in evolution of the genus which has a polyploid



Figs. 38–44: Idiograms prepared from mitotic metaphase in different *Medicago* species. 38, *M. sativa* (1765), 39, *M. sativa* (1766), 40, *M. sativa* subsp. *falcata*, 41, *M. scutellata*, 42, *M. tornata*, 43, *M. truncatula*, 44, *M. tuberculata*.

Table 3. Analysis of mitotic chromosomes in different tetraploid species of *Medicago* (In each species, the first row represents absolute length of chromosome; the second row gives arm ratios; the third row gives relative chromosome length and the fourth row gives TCl%)

Species and collection no.	Chromosome pairs															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
M. sativa	4.07	3.38	3.04	3.04	3.04	2.97	2.76	2.76	2.55	2.55	2.48	2.42	2.35	2.35	2.07	2.07
Coll. no. 1765	1.68	1.45	1.59	1.44	1.32	1.26	1.22	1.22	1.47	1.06	2.27	1.33	1.13	1.00	1.00	1.00
($2n=32$)	100.00	83.05	74.58	74.58	74.58	73.22	67.80	77.80	62.71	62.71	61.02	59.32	57.63	57.63	50.95	50.85
	9.28	7.70	6.92	6.92	6.92	6.76	6.29	6.29	5.82	5.82	5.66	5.50	5.35	5.35	4.72	4.72
M. sativa	4.62	3.52	3.17	3.17	3.17	3.17	3.17	3.04	2.09	2.76	2.76	2.76	2.76	2.76	2.48	2.35
Coll. no. 1766	1.91	1.55	1.30	1.30	1.30	1.19	1.09	1.10	1.10	1.88	1.86	1.22	1.00	1.00	1.25	1.00
($2n=32$)	100.00	76.11	68.66	68.66	68.66	68.66	68.66	65.67	62.69	59.70	59.70	59.70	59.70	59.70	53.72	50.75
	9.52	7.24	6.53	6.53	6.53	6.53	6.53	6.25	5.97	5.68	5.68	5.68	5.68	5.68	5.11	4.83
M. sativa subsp.	3.66	3.04	2.90	2.76	2.76	2.55	2.55	2.41	2.35	2.34	2.28	2.07	2.07	2.07	1.79	1.79
falcata	1.65	1.44	1.62	1.86	1.50	1.62	1.31	1.06	1.12	1.12	1.36	1.14	1.14	1.14	1.17	1.00
($2n=32$)	100.00	83.02	79.24	75.47	45.47	69.81	69.81	66.04	64.15	64.15	62.26	56.60	56.60	56.60	49.06	49.06
	9.28	7.71	7.35	7.00	7.00	6.48	6.48	6.13	5.95	5.95	5.78	5.25	5.25	5.25	4.53	4.53
M. scutellata	3.31	2.21	2.21	2.07	2.07	2.07	2.07	1.93	1.93	1.86	1.86	1.86	1.73	1.73	1.66	1.52
($2n=32$)	1.00	1.46	1.00	1.73	1.31	1.31	1.00	1.00	1.00	1.25	1.08	1.08	1.08	1.08	1.00	1.00
	100.00	66.67	66.67	62.50	62.50	62.50	62.50	58.33	58.33	56.25	56.25	56.25	52.08	52.08	50.00	45.83
	10.32	6.88	6.88	6.45	6.45	6.45	6.45	6.02	6.02	5.81	5.81	5.81	5.38	5.38	5.16	4.73

Table 4. Chromosome data in 19 collections of *Medicago*

Species and collection no.	Total chromatin length (μ)	Mean chromosome length (μ)	Longest/shortest ratio	TF%	Chromatin volume (μ^3)
M. arabica	18.49	2.31	1.52	39.55	12.54
M. coerulea	18.56	2.32	1.43	45.35	15.96
M. coronata	19.74	2.47	1.65	42.31	13.38
M. intertexta var. *ciliaris* (1809)	19.60	2.45	2.09	46.13	13.27
M. intertexta var. *ciliaris* (1808)	17.66	2.22	1.70	44.92	17.91
M. intertexta var. *intertexta*	21.39	2.67	1.47	40.64	16.79
M. littoralis	12.01	1.72	1.68	42.53	6.44
M. lupulina	19.65	2.46	1.81	38.95	12.78
M. murex	15.46	2.21	1.79	44.20	15.67
M. polymorpha	14.49	2.07	1.68	41.91	7.78
M. praecox	19.27	2.41	1.55	41.22	14.09
M. rotata	19.05	2.38	2.08	43.12	15.96
M. sativa (1765)	43.90	2.74	1.97	42.92	41.91
M. sativa (1766)	48.56	3.03	1.97	44.32	40.35
M. sativa subsp. *falcata*	39.39	2.46	2.04	42.73	38.40
M. scutellata	32.09	2.01	2.18	46.88	21.75
M. tornata	15.67	1.96	1.50	41.41	8.41
M. truncatula	18.91	2.36	2.04	43.80	8.49
M. tuberculata	14.36	2.05	1.68	42.31	12.03

Table 5. Karyotypic formulae of 19 collections belonging to 16 different species of *Medicago* (A, B, C and D represent length of the chromosomes in decreasing order; 'sc' used as subscript represents secondary constriction in long arm; superscripts 'm', 'sm' and 'st' represent respectively the median, submedian and subterminal position of centromeres)

S. no.	Species	Karyotypic formulae
1	*M. arabica*	$2C^{m}+3C^{sm}+2C^{st}+1scD^{sm}$
2	*M. coerulea*	$4C^{m}+1scC^{sm}+2C^{sm}+1D^{m}$
3	*M. coronata*	$2C^{m}+1scC^{sm}+4C^{sm}+1D^{sm}$
4	*M. intertexta* var. *ciliaris* (1809)	$1scB^{m}+1B^{sm}+3C^{m}+1C^{sm}+1D^{m}+1D^{sm}$
5	*M. intertexta* var. *ciliaris* (1808)	$1scB^{sm}+3C^{m}+1C^{sm}+3D^{m}$
6	*M. intertexta* var. *intertexta*	$1scB^{sm}+1B^{sm}+2C^{m}+2scC^{sm}+2C^{sm}$
7	*M. littoralis*	$2C^{sm}+1D^{m}+4D^{sm}$
8	*M. lupulina*	$1scB^{sm}+1C^{m}+4C^{sm}+1C^{st}+1D^{m}$
9	*M. murex*	$2C^{m}+2C^{sm}+1scC^{st}+2D^{m}$
10	*M. polymorpha*	$3C^{sm}+3D^{m}+1scD^{st}$
11	*M. praecox*	$1scB^{sm}+2C^{m}+5C^{sm}$
12	*M. rotata*	$1scB^{sm}+1B^{sm}+1C^{m}+2C^{sm}+2D^{m}+1D^{sm}$
13	*M. sativa* (1765)	$1scA^{sm}+1scB^{sm}+3B^{sm}+5C^{m}+5C^{sm}+1C^{st}$
14	*M. sativa* (1766)	$1scA^{sm}+1scB^{sm}+3B^{m}+3B^{sm}+4C^{m}+4C^{sm}$
15	*M. sativa* subsp. *falcata*	$1scB^{sm}+1B^{sm}+6C^{m}+1scC^{sm}+5C^{sm}+2D^{m}$
16	*M. scutellata*	$1B^{m}+2C^{m}+4C^{sm}+8D^{m}+1D^{sm}$
17	*M. tornata*	$2C^{m}+1C^{sm}+1C^{st}+1D^{m}+3D^{sm}$
18	*M. truncatula*	$1scB^{sm}+2C^{m}+1scC^{sm}+2C^{sm}+1D^{m}+1D^{sm}$
19	*M. tuberculata*	$1C^{m}+2C^{sm}+2C^{st}+2D^{m}$

series with $2n=16$, 32 and 48. The base number $x=8$ is found in several genera in Fabaceae. These genera include *Astragalus*, *Crotalaria*, *Indigofera*, *Melilotus*, *Trifolium* and *Trigonella*. It is also known that out of eight species having $2n=14$, only two species namely *M. constricta* and *M. globosa* have exclusively $2n=14$ while the remaining six species (*M. hispida*, *M. murex*, *M. polymorpha*, *M. praecox*, *M. rigidula* and *M. tuberculata*) have both $2n=16$ and $2n=14$.

Karyotype analysis in the genus has been attempted in the past by several workers (Fryer 1930, Heyn 1956, Lesins and Lesins 1961, 1962, 1963a, 1965, 1966, Simon and Simon 1965). Out of these, an important study on karyotype in *Medicago*

Table 6. Placement of different *Medicago* species in 12 categories proposed by Stebbins (1971), on the basis of karyotypes

Longest/Shortest	Proportion of chromosomes with arm ratio 2:1			
	0.0	0.01–0.50	0.51–0.99	1.00
<2:1	1A *M. coronata* *M. intertexta* var. *ciliaris* Coll. no. 1808 *M. intertexta* var. *intertexta* *M. littoralis* *M. sativa* Coll. no. 1766	2A *M. arabica* *M. coerulea* *M. lupulina* *M. murex* *M. polymorpha* *M. praecox* *M. sativa* Coll. no. 1765 *M. tornata* *M. tuberculata*	3A	4A
2:1–4:1	1B *M. intertexta* var. *ciliaris* Coll. no. 1809 *M. rotata* *M. sativa* ssp. *falcata* *M. scutellata*	2B *M. truncatula*	3B	4B
>4:1	1C	2C	3C	4D

is that of Simon and Simon (1965), who analysed 37 accessions belonging to 24 species. In the present study 15 species of *Medicago* were analysed, which is largely a reinvestigation of species earlier utilized either by Fryer (1930) or by Simon and Simon (1965). However, while Fryer (1930) emphasized on chromosome size mainly, Simon and Simon (1965) tried to determine the degree of karyotype asymmetry on the basis of gradient index (smallest chromosome/largest chromosome $\times 100$). It should be realized that gradient index is only one of the criteria used for determining karyotype asymmetry. Stebbins (1971) also used gradient in the form of ratio of longest chromosome to shortest chromosome. It is obvious thus that Simon and Simon (1965) used only part of the information in determining karyotype asymmetry. TF% included in our data also gives an idea about proportion of chromosomes with unequal arms.

The different species investigated during the present study were classified according to Stebbins' (1971) classification, which is shown in Table 6. If the results are compared with those of Simon and Simon (1965), only nine species are common which are *M. arabica*, *M. coronata*, *M. intertexta* var. *ciliaris*, *M. intertexta* var. *intertexta*, *M. littoralis*, *M. murex*, *M. rotata*, *M. tornata* and *M. truncatula*. It can be realized that gradient index of less than 50% will fall in categories of class A and those with a gradient index of more than 50% should fall in categories of class B. Our results agree with those of Simon and Simon (1965) with respect to *M. rotata*, *M. coronata*, *M. littoralis* and *M. murex*. It will be seen that even among these, *M. murex* falls in 2A showing higher degree of asymmetry which can not be inferred from gradient index. This species, namely, *M. murex* is, therefore, an advanced species.

Accessory chromosomes, also described as B-chromosomes were also recorded in four taxa belonging to three species namely *M. intertexta* var. *ciliaris*, *M. intertexta* var. *intertexta*, *M. littoralis* and *M. tornata*, in root tip cells as examined in the mitotic metaphase. In the present study of the three species, exhibiting the presence of B-chromosome only one species, namely *M. intertexta* was earlier known to have B-chromosome (Heyn 1963). Therefore, the presence of B-chromosome in *M. littoralis* and *M. tornata* is reported for the first time during the present study.

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Summary

1. In the genus *Medicago* male meiosis was studied in 14 species and mitosis with the help of root tips was studied from 19 collections belonging to 15 species. Most of the species were diploids having $n=7$ or $n=8$, only two species (*M. sativa* and *M. scutellata*) were tetraploids having $n=16$.
2. Chiasmata frequency/bivalent varied from 1.77 (*M. tornata*) to 2.18 (*M. scutellata*).
3. Total chromatin length varied from 14.36 μ (*M. tuberculata*) to 48.56 μ (*M. sativa* Coll. no. 1766).
4. On the basis of karyotypes, species were classified according to the classification of Stebbins (1971). Only four of the 12 categories were represented by the species examined, indicating that the degree of asymmetry was low.
5. Accessory chromosomes were recorded from root tip cells in four taxa belonging to three species (*M. intertexta*, *M. littoralis* and *M. tornata*).

References

Agarwal, K. 1979. Cytogenetic studies in Trifolieae and Galegeae with special emphasis on *Indigofera* L. Ph. D. Thesis, Meerut University, Meerut.

Fryer, J. R. 1930. Cytological studies in *Medicago*, *Melilotus* and *Trigonella*. Can. J. Res. **3**: 3–50.

Gupta, R. and Gupta, P. K. 1978. Karyotypic studies in the genus *Crotalaria* Linn. Cytologia **43**: 357–369.

Heyn, C. C. 1956. Some chromosome counts in the genus *Medicago*. Caryologia **9**: 160–165.

— 1963. The annual species of *Medicago*. Scri. Hierosol. Publ., Hebrew Univ., Jerusalem **12**: 1–154.

Lesins, K. and Lesins, I. 1961. Some little known *Medicago* species and their chromosome complements. Can. J. Genet. Cytol. **3**: 7–9.

— and — 1962. Trueness to species in seed samples of *Medicago* with a note on $2n=14$ species. Can. J. Genet. Cytol. **4**: 337–339.

— and — 1963a. Some little known *Medicagos* and their chromosome complements, II. Species from Turkey. Can. J. Genet. Cytol. **5**: 133–137.

— and — 1963b. *Medicago saxatilis* M. S., a second high-ploidy species in the genus. Can. J. Genet. Cytol. **5**: 348–350.

— and — 1965. Little known *Medicago* and their chromosome complements, III. Some Mediterranean species. Can. J. Genet. Cytol. **7**: 97–102.

— and — 1966. Little known *Medicago* and their chromosome complements, IV. Some mountain species. Can. J. Genet. Cytol. **8**: 8–13.

Simon, J. P. and Simon, A. 1965. Relationships in annual species of *Medicago* L. Number and morphology of chromosomes. Aust. J. Agric. Res. **16**: 37–50.

Stebbins, G. L. 1971. Chromosomal Evolution in Higher Plants. Edward Aronold (publ.) Ltd., London.