

**G & BM**

**Tome IV**

**Iași, 2003**

## **THE CHROMOSOMAL NUMBER FOR RARE SPECIES FROM ROMANIA**

**ION I. BĂRA, RODICA RUGINĂ, CSILLA IULIANA BĂRA**

**Key words:** rare species, chromosomes number, endemic species

**Abstract:** The cytogenetic data for rare and endemic species from Romanian flora are uncertain and contradictory. It is necessary to establish the role of polyploidy and aneuploidy for survival and evolution of these species

### **INTRODUCTION**

In Romanian flora there exist many rare species, some times subjected to disappear. The investigation of these species chromosomes is very interesting and very useful for some correlations between karyotype and environment. We have considered very interesting to mention the areas of each species because we must know if a species is in the situation to become extinct. It is important the fact that investigated species are from many and very different families.

### **MATERIAL AND METHODS**

It was done a list with rare species existing in Romanian flora and, after that, for each of them it was indicated the  $x$  and  $2n$  numbers of chromosomes. On this basis we tried to establish if a population have individuals with some degrees of polyploidy and/or aneuploidy.

### **RESULTS AND DISCUSSIONS**

We found in formations for 95 rare species existing in the our flora but, in many cases, the informations are uncomprehensive and very different from one author to the other. Examining the table we may see that from the 95 registered species, for 45 of them is not a concordance between  $x$  and  $2n$ . For instance at *Allium flavum* ssp. *tauricum* there are mentioned the numbers  $x = 7, 8$  and  $9$  (Levan, 1929, 1931, 1935, 1937; Ono, 1935; Delay 1947; Tschermak-Woess, 1947) and  $2n = 16$ . Of course  $x=8$  is concordance with  $2n=16$  but we can not find a concordance between  $x=7$  or  $x=9$  and  $2n=16$ . A concordance between  $x$  and  $2n$  we have found only for 39 species (from those mentioned in table). We must underline that 17 of them present polyploide forms and the rest of 22 are diploide only. For instance the species *Acanthus balcanicus* Heyw et Richardson, *Asphodeline lutea* (L.) Rchb., *Beta trigyna* W et K, *Clypeola Jonthlaspi* L., *Crambe maritima* L., *Dianthus spiculifolius* Schur, *Digitalis ferruginea* L., *Galanthus elwesii*

Hook., *Helleborus odoratus* Wet. K., *Laburnum alpinum* (Mill) Gris., *Laburnum anagyroides* Medik, *Menyanthes trifoliata* L., *Peltaria alliacea* Jacq., *Petrorhagia saxifraga* (L) Link, *Prunus tenella* Batsch., *Pulsatilla vulgaris* Mill. Ssp. *grandis* (Wend) Zam., *Ruscus hypoglossum* L., display different degrees of polyploidy.

The species *Adonis vernalis* L., *Alyssum borzeanum* Nyar., *Angelica arhangelica* L., *Colutea arborescens* L., *Dianthus nardiformis* Janka, *Globularia punctata* Lapeyr, *Glaucium flavum* Cr., *Larix decidua* Mill., *Opopanax bulgaricus* Velen., *Paeonia daurica* Andr., *Paeonia tenuifolia* L., *Paliurus spina christi* Mill., *Polemonium coeruleum* L., *Potentilla rupestris* L., *Pyrus deagrifolia* Pall., *Scandix pecten-Weneris* L., *Schivierkia podolica* (Bess) Andr., *Scorzonera purpurea* L., ssp. *rosea* (W et K) Nym., *Silene nutans* L., ssp. *dubia* (Herb) Zapal, *Sorbus aria* (L) Cr., *Veratrum nigrum* L., *Waldsteinia geoides* Willd are diploide (they present a concordance between  $x$  and  $2n$ ).

On the other hand, we have met a great variability for the diploide chromosome number and unconcordance between  $x$  and  $2n$ . For instance, for *Anthericum liliago* L. Species are mentioned  $2n = 30, 32, 60$  and  $64$  and  $x = 8$  and  $15$ . May be a case of aneuploidy  $8$  being the basis number. From it could appeared the tetraploid  $2n=32$  and the octoploid  $2n=64$  From the diploid  $2n=16$  could appeared the monosomic  $2n=15$  after that the tetraploid  $2n=30$ . But all these are only suppositions. For other species the situation is more unclear. For instance at *Sedum album* L. are mentioned the numbers  $x = 6, 7, 8, 10, 11, 17$  și  $2n = 32, 64$ . We may find correspondence only between  $8$  and  $32$  or  $64$ , the others  $x$  values being without correspondence with  $2n$ . For these situations are necessary new investigations.

For the endemic species we have found the following situation: 11 of them (*Dianthus spiculifolius* Schur., *Erysumum witmanii* ZaW., *Erysumum comatum* Panc., *Minuartia graminifolia* (Ard) JaW., *Pinus nigra* Am., ssp. *banatica* (Borb) Novak., *Salvia transilvanica* (Schurr ex Griseb) Schur., *Serratula bulgarica* Acht et Stoj., *Silene nutans* L. ssp. *dubia* (Herb) Zapal., *Silene zavadzki* Herb., *Syringa josikaea* Jack. F. Ex. Reichenb.) are endemic species in Romania (some of them only in Carpathian mountains), an argument more for new investigations.

Ones from these 95 species are spread in Europe, North part of Africa and in Western Asia.

## CONCLUSIONS

The cytogenetic data for rare and endemic species from romanian flora are uncertain and contradictory.

We consider that are necessary new investigations for establish the real number of chromosomes for rare and endemic species from romanian flora (a condition for cytogenetic characterisation of these species).

Is necessary to establish the role of polyploidy and aneuploidy for survival and evolution of these species.

## BIBLIOGRAFIE

**Băra, I.I., Truță, E., Albu, I., 1990.** *The diversification of the evolution in the Papaveraceae family.* Anale UniW. "Al.I.Cuza" Iași, XXXVI, s. II-a., 99-102.

**Băra, I.I., Maniu, Marilena, Vană, Sofica, Surugiu, Csilla Iuliana, 2001.** *The karyotype of *Vicia narbonensis* L. and *Vicia sativa* L. species.* Analele Științifice ale Universității "Al.I.Cuza" - Iași, secțiunea II, a. Genetică și Biologie Moleculară, tom II, 38-41.

**Bolkoviskih, Z., Grif, W., Matvejeva, T., Zaharieva, O., 1969.** *Hromozomnue cista pvetovih rastenii.* Nauka, Leningrad.

**Cesmendiev, I. W., 1971.** *Ţitosistematiceskie issledovanie nekotorih vidov iz rodov Allium L. I Nectaroscondum Lindl.* Bot. Journ.SSR., 11, 1644-1657.

**Kuzmanov, A., 1993.** *Chromosome number of Bulgarian angiosperms: An introduction to a chromosome atlas of the Bulgarian flora.* Flora Mediteranea, 3, 19, 19-163.

**Morariu, Aliona, Băra, I.I., 2001.** *Cytogenetic effects of alkylant agents administration on *Glaucium flavum* Cr., *Papaver somniferum* L., and *Papaver rhoeas* L. species.* Analele Științifice ale Universității "Al.I.Cuza" - Iași, secțiunea II, a. Genetică și Biologie Moleculară, tom II, 86-92.

**Sveșnikova, L.I., 1965.** *Hromozomnue cista nekotorih vidov roda Galanthus.* Bot. Journ. SSSR, 52, 3, 359-362

**Sveșnikova, L. I., 1967.** *Hromozomnue cislane nekotorih vidov roda Galanthus iz iujnoi Evropi i maloi Azii.* Bot. Jum., SSSR, 52, 3, 359-363.

**Szabo, T., 1972/1994.** *Date carilogice pentru *Adonis* L., secția Consiligo D.C. din Transilvania (România).* Lucr. Șt. Seria Agr., Inst. Agr. "Petru Groza"-Cluj, 28, 75-80.

**Tarnavski, I., 1948.** *Die chromozomenzahlen der Antophyten. Flora von Rumänien mit einem Ausblick aus das Polyploidie Problem.* Buletinul. Grădinii Botanice Cluj, 27 (supliment 1), 1-30

No.	The species	The chromosomes number		The author	Area
		x	2n		
1	<i>Acanthus balcanicus</i> Heywet Richrdson	14	56	Grant, W.F., 1955	Balcani
2	<i>Adonis vernalis</i> L.	8	16	Langlet, 1927, Tureson 1938, Larsen 1954, Jaworska 1964, Skalinska, 1964	Central Europe, Siberia, Western Asia
3	<i>Adonis vologensis</i> Steve	4, 8	16	Szabo, 1972	România, Hungary, SSSR, Central Asia
4.	<i>Allium flavum</i> L. Ssp. <i>tauricum</i> (Besser et Reichenb.), Stearn	7, 8, 9	16, 32	Levan, 1929, 1931, 1935, 1937; Ono, 1935; Delay, 1947; Tschermak- Woess, 1947.	S-E Europe
5	<i>Allium obliquum</i> L.	7, 8, 9	16	Levan, 1931, 1935; Geitler, 1935; Sato, 1942	Eurasia
6	<i>Alyssum borzeanum</i> Nyar.	8	16	Manton, 1932	România, Bulgaria, SSSR
7	<i>Angelica archangelica</i> L	11	22	Schulz-Goebel, 1932; Love et Loves, 1948, 1956; Garde et Malheiros- Garde, 1949; Jørgensen, et al., 1958	Central Europe, Siberia
8	<i>Anthericum liliago</i> L.	8, 15	30, 32, 60, 64	Bowdwn, 1940, 1945; Delay, 1947; Strandhede, 1963; Eldvers, 1932; Tischler, 1934	Central and South Europe, Asia Minor, North Africa.
9	<i>Asparagus verticillatus</i> L	10	20, 24	Bozzini, 1959; Delay, 1947	România, Balcani, Ukraine, Asia Minor
10	<i>Asphodeline lutea</i> (L) Rehb	7	14, 28, 56	Sussenguth, 1921; Prozina, 1936; Sat, 1942; Delay, 1947; La Cour, 1952; Mitra, 1964; Kozumanov et Popova, 1968; Borhidi, 1968	Mediterranean, Algeria
11	<i>Astragalus dasyanthus</i> Pall.	8, 12	16	Cehov, 1935; Pavlova, 1988	Balcani, SSSR
12	<i>Belvalia sarmatica</i> (Pall) Woron	4, 5	9	Buvat, 1945; Zaharieva, Makusenko, 1968	România, Bulgaria, SSSR
13	<i>Beta trigyna</i> W et K	9	36, 54	Zasimovici, 1934, 1938, 1965; Bleier, 1928; Sirotina, 1934, 1936; Walther, 1963	Rusia, România, Galicia
14	<i>Campanula romanica</i> SăW.	8, 10, 17	34	Podlech, Danboldt, 1963	Central Europe, Siberia
15	<i>Centaurea marschalliana</i> Spreng	8, 9, 10, 11, 12	20	Ciukanova et al., 1968	SSSR, România, Galicia

16	<i>Centaurea ruthenica</i> Lam.	8, 9, 10, 11, 12	30	Podubnaia-Arnoldi, 1931	Central Asia, Central Europe
17	<i>Chypeola Jonthlaspi</i> L.	8	16, 32	Jaretski, 1928; Bonnet, 1963	Mediterranean
18	<i>Colutea arboreescens</i> L.	8	16	Cehov, 1930, 1935; Hindacova, 1967	Central Europe, S-W Asia
19	<i>Convolvulus persicus</i> L.	10, 11	20, 22	Cezmedziev, 1976	Black Sea and Caspian
20	<i>Coronilla emerus</i> L. Ssp. <i>emeroides</i> (Boiss et Sprun) Hay.	5, 6, 7	18	Ciukanova, 1967	Mediterranean
21	<i>Coronilla scorpioides</i> (L.) Koch	5, 6, 7	12	Larsen, 1955	Mediterranean
22	<i>Coronilla vaginalis</i> Lam	5, 6, 7	12	Baksav, 1956	Central Europe
23	<i>Corylus columa</i> L.	7, 11	14	Woodworth, 1929	South Europe, Asia Minor, Caucasus, Himalaya, China
24	<i>Grambe maritima</i> L.	15	30, 60	Tarnavski, 1948; Jaretski, 1932; Manton, 1932; Tischler, 1934; Wulf, 1937; Litardiere, Doulat, 1942	Europe
25	<i>Crocus chrysanthus</i> (Herb) Herb	4, 5, 7	8, 9, 10	Darlington, 1937; Karasawa, 1937, 1961; Mather, 1932; Bowles, 1952	Balkans
26	<i>Cytissus ratisbonensis</i> (Schaf) Rothm	6, 8, 10	48	Matick, 1950	Central Europe, South Russia, Siberia
27	<i>Dianthus nardifolius</i> Jan ka	15	30	Rohweder, 1934; Gentscheff, 1937; Carolin, 1957	Bulgaria
28	<i>Dianthus spiculifolius</i> Schur.	15	60	Rohweder, 1934; Gentscheff, 1937	Endemic in Carpathian mountains
29	<i>Dictamnus albus</i> L.	9	30, 36, 72	Negodi, 1939; Bowden, 1940, 1945; Desai, 1960	Eurasia
30	<i>Digitalis ferruginea</i> L.	7	56, 70	Buxton, Dark, 1934; Yakar, 1945	Balkans
31	<i>Echbalium ellaterium</i> (L.) A. Rich	12	18, 24	Kojuhov, 1934; McKay, 1930, 1931.; Whitaker 1933; Delay, 1947	Mediterranean
32	<i>Echinops banaticus</i> Roch. Ex Schrad	8	30	Move, Frankton, 1962	Balkans, Asia Minor, Caucasus
33	<i>Ephedra distachya</i> L.	10, 12	16, 24, 28	Jaccard, 1894; Berridge et Sanday, 1907; Sigrianki, 1913; Florin, 1932; Resende, 1937	Eurasia
34	<i>Erysimum wittmanii</i> ZaW.	7, 8	14	Favarger, 1964; Jankun, 1965; Polatschek, 1966	Endemic in Carpathian mountains
35	<i>Erysimum comatum</i> Panc.	7, 8	14	Polatschek, 1966	Endemic in Carpathian mountains
36	<i>Ferula sadleiriana</i> Ldb	10, 11	22	Baksav, 1956	Romania, Bulgaria

37	<i>Galanthus elwesii</i> Hook	12	24, 48	Heitz, 1926; Sato, 1937, 1938, 1942; Svešnikova, 1967	Balcani
38	<i>Genista radiata</i> (L.) Scop.	6, 7	48	Santos, 1945	Balcani
39	<i>Globularia punctata</i> Lapeyr.	8	16	Schwarz, 1964; Murin, Uhríkova, 1967; Hindakova, 1967	Central and South Europe, Caucasia
40	<i>Glaucium flavum</i> Cr.	6	12	Sugiura, 1931, 1936; Smith, 1950; Rodrigues, 1953; Larsen, 1954; Băra et al., 1990	Balcani, Mediterana, Asia Minor, Crimea, Caucasia
41	<i>Gymnospermium altaicum</i> (Pallas) Spach.	6, 8	16	Toren, 1962	România, Bulgaria
42	<i>Helieborus odoratus</i> Wet. K.	8	32	Langlet, 1932	S-E Europe
43	<i>Hyacinthella leucophæa</i> (C. Koch) Schur	9, 10	18, 20	Buvat, 1945; Popova, 1972	South Europe, Russia, Asia Minor
44	<i>Iris graminea</i> L. Ssp. <i>brandze</i> (Prod) D. Web. Chater	5, 6, 7, 11, 17	10	Tarnavschi, 1938	Central and South Europe
45	<i>Iris halophila</i> Pall.	5, 6, 7, 11, 17	44, 46, 48	Simonet, 1934; Lenz, 1963; Day, 1963; Tarnavschi, 1938	S-E Europe
46	<i>Iris sibirica</i> L.	5, 6, 7, 11, 17	28	Kazao. 1928, 1929; Simonet, 1928, 1932, 1934; Randolph, 1934; Woisk, 1964.	Eurasia
47	<i>Iris sintenisii</i> Jan ka	5, 6, 7, 11, 17	16	Randolph & Randolph, 1956.	România, Balcani, Asia Minor
48	<i>Laburnum alpinum</i> (Mill.) Gris	6	48	Cehov, 1931; Gilot, 1965.	Central Europe
49	<i>Laburnum anagyroides</i> Medik	6	48	Gilot, 1965	Central Europe
50	<i>Larix decidua</i> Mill.	12	24	Nemec, 1910; Sax, 1932, 1933; Hruby, 1933; Smolska, 1927	Alpi, Silezia
51	<i>Menyanthes trifoliata</i> L.	9	54	Matsuura, Sato 1935; Palmgren, 1943; Love & Love, 1944, 1956; Delay, 1947; Rork, 1949	Europe, Mediterana

52	<i>Minuartia graminifolia</i> (Ard.) A.W.	8, 9, 10, 11, 12, 13, 15, 23	32	Favarger, 1962	Endemic in România
53	<i>Narcissus angustifolius</i> Curt	7, 10, 11	14, 14+1-2B	Geitler, 1935; Grif unpublished	Europe
54	<i>Nectaroscordium siculum</i> (Ucrâ.) Lindt. Ssp. <i>bulgaricum</i> (Janka.) Stearn	9	16, 18	Mensincai, 1939, 1940; Cesmedziev, 1970	România, Balcani, Crimeea, Asia Minor
55	<i>Oporanax bulgaricus</i> Velen	11	22	Garde, 1957	România, Bulgaria
56	<i>Ornithogalum fimbriatum</i> Wild	6, 7, 8, 9	12, 14	Delaunay, 1926; Neves, 1956; Narkova, Radenkova, Ivanova, 1972, 1974	Balcani, Crimeea, Asia Minor
57	<i>Paeonia daurica</i> Andr.	5	10	Barber, 1941	Crimeea, Caucasia
58	<i>Paeonia peregrina</i> Mill. Var. <i>romanica</i> Brândză	5	20, 16	Gajewski, 1948; Wefelscheid, 1911; Tarnavski unpublished	România (Dobroudja)
59	<i>Paeonia tenuifolia</i> L.	5	10	Gajewski, 1948	Balcani, Ucraina, Crimeea, Caucasia
60	<i>Paliurus spina-Christi</i> Mill	12	24	Dolcher, 1947, 1955	South Europe, Western Asia
61	<i>Peltaria alliacea</i> Jacq.	7	28	Mantbn, 1932	Alpes, Balcani
62	<i>Periploca graeca</i> L.	11, 12	22, 24	Bowden, 1940, 1945; Pardi, 1933; Lopane, 1951	Mediterran
63	<i>Perorhagia saxifraga</i> (L.) Link	15	60	Blackburn, 1931; Favarger, 1946; Larsen, 1954; Nagl, 1962	Balcani
64	<i>Pinus nigra</i> Arn. ssp. <i>banatica</i> (Borb.) Novak		24	Ferguson, 1924	Endemic in România
65	<i>Polygonum caeruleum</i> L.	9	18	Winge, 1923; Clausen, 1931; Heitz, 1932; Sakai, 1935; Flory, 1937; Greiesinger, 1937; Turesson, 1938; Love & Love, 1956	Europe, Caucasia, Siberia, Japan, North America.
66	<i>Potentilla rupestris</i> L.	7	14	Tischler, 1929; Simotomei 1930; Clausen et al., 1932, 1939, 1940; Popov, 1935, 1939; Gagnieu, 1959	Eurasia, North America.
67	<i>Pninus tenella</i> Batsch.	8	16	Koebel, 1927, 1928	Central Europe, Transcaucasia, Central Asia, East Siberia.
68	<i>Pulsatilla vulgaris</i> Mill. Ssp. <i>grandis</i> (Wend.) Zam.	8	32	Tarnavski, 1948	Europe.
69	<i>Pyrus eleagnifolia</i> Pall.	17	34	Rabin, 1926; Adate, 1933	S-E Europe, Asia Minor, Caucasia

70	<i>Ranunculus constantinopolitanus</i> (DC) D'UrW.	7, 8	14	Langlet, 1936, 1942; Laster, 1932	Balceni
71	<i>Rumex scutatus</i> L.	4, 5, 7, 8, 9, 10	20	Noda, 1926; Jaretsky, 1928; Ono, 1935; Love, 1942; Mattick 1950	Europe, Asia Minor, Caucasia, Transcaucasia, North Africa
72	<i>Ruscus aculeatus</i> L.	10	36, 40	Fernandes, 1929, 1930, 1931; Nakajima, 1936; Sato, 1942; Martinoli, 1951	Europe, North Africa.
73	<i>Ruscus hypoglossum</i> L.	10	40	Martinoli, 1951	Central Europe, Balceni, East Africa, Asia Minor
74	<i>Salvia ringens</i> Sibth et Sm	6, 8	12, 14	Scheel, 1931; Iakovleva, 1933; Markova et Ivanova, 1982	Balceni, Asia Minor
75	<i>Salvia transsilvanica</i> (Schur et Griseb.) Schur	6, 8	16, 18	Scheel, 1931; Iakovleva, 1933; Hrubý, 1935; Benoist, 1937	Endemic
76	<i>Scandix pecten-Weneris</i> L.	8	16	Melderis, 1930; Schulz-Gaebel, 1930; Vanscher, 1931; Tischler, 1934; Delay, 1947; Vitaker, 1948; Kordium, 1967	Europe, Mediterana
77	<i>Schivereckia podolica</i> (Bess.) Andz.	8	16	Manton, 1932	România, Russia till Ural
78	<i>Scorzonera pupurea</i> L ssp. <i>rosea</i> (W et K) Nym.	7	14	Favarger et Huynh, 1964; Favarger, 1965; Ciuksanova et al., 1968	Central Europe, Balceni, România
79	<i>Scutellaria orientalis</i> L ssp. <i>sosnovskyi</i> (Takht.) Fed.	8, 11, 17	16	Quezel, 1957	S-E Europe, Caucasia, Asia Minor, Siberia
80	<i>Sedum album</i> L.	6, 7, 8, 10, 11, 17	32, 64	Baldwin, 1935, 1939	Europe, Asia Minor, N-W Asia, N.Africa
81	<i>Serratula bulgarica</i> Acht. et Stoj.	10, 11	22, 24	Narkova et al., 1972	Endemic in România
82	<i>Serratula radiata</i> W et K.) M.B.	10, 11	30, 60	Podubnaia-Arnoldi, 1939; Baksay, 1956, 1957	România, Balceni, Caucasia, W. Siberia
83	<i>Serratula wolffi</i> Andrae.	10, 11	22	Tarnavski, 1938; Arano, 1964, 1965; Ciuksanova et al., 1968	România, Siberia, W. Russia.
84	<i>Silene nutans</i> L. Ssp. <i>dubia</i> (Herb.) Zapal.	12	24	Blackburn, 1928, 1929; Tischler, 1934; Rockweder, 1939; Vladesco, 1941	Endemic in Carpathian mountains



85	<i>Silene zawadzki</i> Herb.	6, 7, 8, 10, 11, 17	24	Heitz, 1926	Endemic in Carpathian mountains
86	<i>Sorbus aria</i> (L.) Cr.	17	34	Sax, 1931; Pouques, 1951; Lijefors, 1953; Baksay, 1956; Moffet, 1931	Europe
87	<i>Spiraea crenata</i> L.	5, 9	36	Sax, 1936	Europe, Russia
88	<i>Spiraea salicifolia</i> L.	5, 9	36	Sax, 1936	Central Europe., Siberia, N-W. America.
89	<i>Syringa josikaea</i> Jacq. f. ex Reichenb.	11, 23	44, 46, 48	Tischler, 1930; Sax et Abbe, 1932	Endemic in Carpathian mountains
90	<i>Taxus baccata</i> L.	8, 12	16, 24	Overton, 1892; Strasburger, 1904; Newbn, 1927; Darlington, 1930; Dark 1932; Sax, 1933;	Europe, W. Asia., N. Africa.
91	<i>Veratrum nigrum</i> L.	8	16	Miller, 1930	Alpes, Central Europe, Balcani, Siberia
92	<i>Vicia narbonensis</i> L.	5, 6, 7	14	Svešnicova, 1927; Heitz, 1931; Senecianova-Korceaghina, 1932; Kumar, 1960; Srivastava, 1963; Martin et Shanks, 1966; Ciukanova, 1967; Băra et al., 2001	Europe
93	<i>Vicia tenuifolia</i> Roth. Ssp. <i>stenophylla</i> Velen.	5, 6, 7	12	Baksay, 1954	Balcani, Crimea, Caucasus
94	<i>Waldstenia geoides</i> Willd.	7	14	Polva, 1949; Reese, 1925	România, Balcani, Crimea
95	<i>Zyzyphus jujuba</i> Mill.	12	24, 40, 48, 60, 72, 96	Morinaga et al., 1929; Bowdwn, 1940, 1945; Srinivasachar, 1940; Srinivasan, 1952; Khoshoo et Singh, 1963	