CYTOGENETIC STUDIES IN SPERGULARIA

I. CYTOLOGY OF SOME OLD WORLD SPECIES

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During the past years the writer has been carrying out biosystematic studies in *Spergularia* (Pers.) Presl and prior to the publication of these results it is necessary to devote a short paper to the cytology of the species studied. There are about 20 species of the genus in Europe and North Africa but those studied have been limited by availability to the eleven which are listed below.

S. fimbriata Boiss.	S.W. Spain, S. Portugal, Morocco, Canary Islands. Perennial.
S. rupicola Lebel ex Le Jolis	Atlantic coasts of Europe, northwards to C. 58° N. in Scotland. Perennial.
S. media (L.) C. Presl (*S. marginata* (DC.) Kittel)	Cosmopolitan. Perennial.
S. marina (L.) Griseb. (*S. salina* J. & C. Presl)	Throughout Northern Hemisphere. Annual to short-lived perennial.
S. diandra (Guss.) Boiss. (*S. salsuginea* Fenzl)	S. of Europe & Mediterranean region, N. Africa. Annual.
S. purpurea (Pers.) G. Don (*S. longipes* Rouy)	Mediterranean region, N. Africa. Annual.
S. rubra (L.) J. & C. Presl (*S. campestris* Aschers.)	Throughout Northern Hemisphere. Annual to short-lived perennial.
S. nicaeensis Sarato ex Burnat	Mediterranean region. Biennial to perennial.
S. capillacea Willk. & Lange	Iberian peninsula. Annual to short-lived perennial.
S. bocconii (Scheele) Aschers. & Graebn. (*S. atheniensis* (Heldr. & Sart.) Aschers. & Schweinf.)	S.W. Europe and Mediterranean region. Annual to biennial.
S. heldreichii (Foucaud) Simon & Monnier	Mediterranean region extending to the Atlantic coast of France. Annual.

Five of these species *S. rupicola*, *S. media*, *S. marina*, *S. rubra* and *S. bocconii* are members of the British flora and both British and non-British material of them has been used in the investigation.

MATERIAL AND METHODS

The experimental plants used in the investigation were obtained from field collections made by the writer and other collectors (particularly

Dr. P. Monnier of the University of Montpellier) and from seeds sent by various Botanic Gardens. The source of all material is given in Table I.

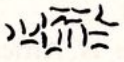
Most of the plants used for cytological study were grown in a cool greenhouse but some fixations of flower-buds were also made from plants in an outdoor plot.

Flower buds to be used for squash preparations were fixed in 3:1 ethanol/acetic acid and stored in the fixative in a "deep-freeze" at —10° C; iron acetocarmine was used for staining. Root-tips were fixed in Navashin's fluid to which a little saponin had been added and, after embedding, sections were cut at 10μ and stained using Johansen's crystal violet method (1934).

At least a dozen chromosome counts were made for every collection and in those which were analysed for occurrence of multivalents a minimum of fifty first meiotic metaphases were examined.

Results

Chromosome numbers recorded in this investigation are listed in Table 1 and compared with those made by other workers. Meiosis in the pollen mother cells is illustrated in Pl. 11 and mitosis in the root-tip in Figs. 1 and 2. The size of the mitotic chromosomes in the root tips is 0·75–2·0μ which is too small for any useful analysis of chromosome morphology.



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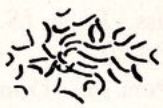

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Figs. 1 and 2.

Root tip mitoses of *Spergularia* ×1800. 1. *S. media*, 2n=18. 2. *S. rupicola*, 2n=36.

Chiasma frequency is low (only one or two chiamata per bivalent) but despite this, occasional quadrivalents were observed in some of the tetraploid species. The number of quadrivalents was, however, never more than one per cell and the percentage of cells in which they occurred always very low. Insufficient pollen mother cells were examined to give an accurate percentage for quadrivalent formation; Table 2 gives the actual numbers observed. It is often difficult to distinguish whether a body is a quadrivalent or two bivalents lying together and where there has been doubt the latter explanation has been taken. Single pollen mother cells with sixteen bivalents, one trivalent and one univalent were observed in both *S. nicaeensis* (Hèrault collection) and *S. purpurea* (Cordoba collection).

In one flower bud of *S. purpurea* from Coimbra many metaphases were seen with one univalent present and anaphases were observed where there was one lagging chromosome, presumably corresponding to this univalent. Unfortunately none of these figures was good enough for detailed analysis.

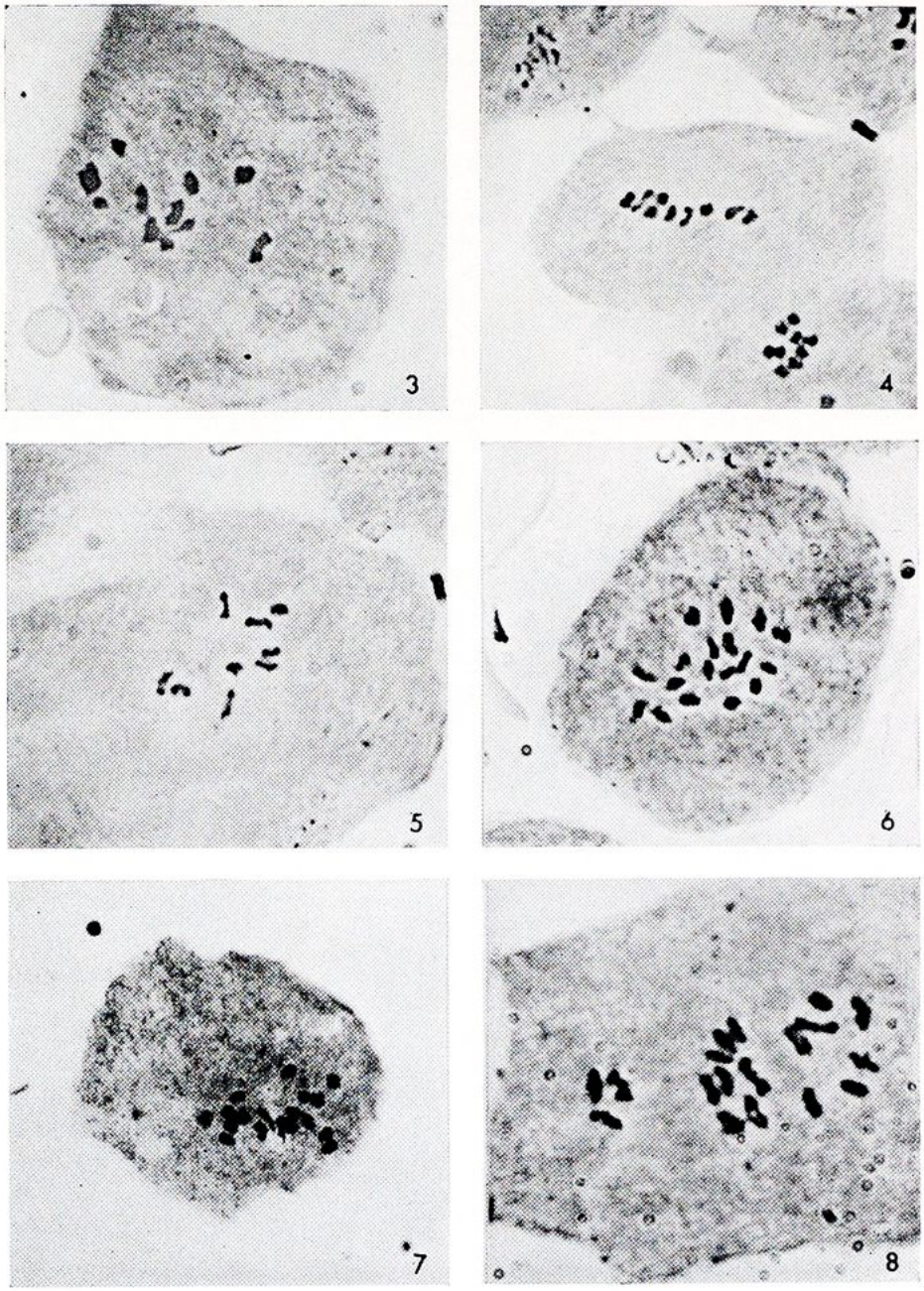

PLATE 11. FIGS. 3-8. 1st meiotic metaphase in P.M.C. ×1,000.
3. *S. fimbriata*, 9 bivalents (1 separated). 4. *S. media*, 9 biv. 5. *S. diandra*, 9 biv.
6, *S. marina*, 18 biv. 7. *S. bocconii*, 18 biv. 8. *S. heldreichii*, 18 biv.

TABLE 1

Chromosome numbers in Spergularia

Species	*Source*	*Herbarium Specimen Number*	*Mitosis root tip* 2n	*Meiosis PMC* n	*Previous Counts* *Source*	*Authority*
S. fimbriata	Rabat, Morocco. (Institut Scientifique Cherifien)	C571	18	9^{II}		
S. rupicola	Cornwall, v.c.2	C591		18^{II}		
	Pembrokeshire, v.c.45	C592		18^{II}		
	Anglesey, v.c.52	C572	36	18^{II}		
	Denbighshire, v.c.50	C593		18^{II}		
	Cheshire, v.c.58	C561	36	18^{II}		
				$16^{II}\ 1^{IV}$		
	Co. Antrim, v.c.H39	C594		18^{II}		
				$16^{II}\ 1^{IV}$		
+	University College of North Staffordshire	C573		18^{II}		
+	Jardin Botanique de L'Université et de la Ville de Lausanne, Switzerland	C574	36	18^{II}		
	Estaçao Agronomica Nacional Sacavém, Portugal	C595		18^{II}		
			36		B	Blackburn & Morton, 1957
S. media	Anglesey, v.c.52	C575	18	9^{II}		
	Parkgate, Cheshire v.c.58	C576	18	9^{II}		
	West Kirby, Cheshire, v.c.58	C577		9^{II}		
	Glasson, West Lancs., v.c.60	C578	18	9^{II}		
	Wyre Estuary, West Lancs., v.c.60	C579		9^{II}		
	Gironde, West of Bordeaux, France	C611		9^{II}		
	Palavas, South of Montpellier, France	C612		9^{II}		
	Aragon, Spain	C581	18	9^{II}		
	Valencia, Spain	C582		9^{II}		
	South of Tunis, Tunisia	C613		9^{II}		
	Jardin Botânico, Universidade de Coimbra, Portugal	C596		9^{II}		
	Sacavém, Portugal	C597		9^{II}		

TABLE 1 (*Continued*)

Species	*Source*	*Herbarium Specimen Number*	*Mitosis root tip* 2n	*Meiosis PMC* n	*Previous Counts* *Source*	*Authority*
			18		G	Wulff, 1937
			18		S	Nordenskiold 1942
			18		P	Castro & Fontes, 1946
			18		P	Rodriquez, 1953
			18		Co, F, NA, P, Sp.	Monnier,1955 & 1960B
			18		P & B	Blackburn, & Morton, 1957
			36		B	Blackburn, & Morton, 1957
				9^{II}	NA	Reese, 1957
Race with no seed-wing. (*S. marginata* var. *aptera* Marshall)	Glasson, West Lancs., v.c.60	C5710	18	9^{II}		
	Loch Eil, v.c.97	C614		9^{II}		
	Rum, v.c.104	C598		9^{II}		
	West Ross, v.c.105	C599		9^{II}		
			18		F & NA	Monnier,1955
S. marina	N. Norfolk, v.c.28	C5711		18^{II}		
	Anglesey, v.c.52	C5712		18^{II}		
	Burton, Cheshire, v.c.58	C5713		n=18 2nd mei. met.		
	Frodsham, Cheshire, v.c.58	C5714	36	18^{II} 16^{II} 1^{IV}		
	Glasson, West Lancs., v.c.60	C5715	36	18^{II}		
	Marsh Houses, West Lancs., v.c.60	C5716	36	18^{II}		
	West Ross, v.c.105	C5910		18^{II}		
	Nr. Montpellier, France	C615		18^{II}		
	Alicante, Spain	C583		18^{II}		
	Barcelona, Spain	C584		18^{II}		

TABLE 1 (*Continued*)

Species	*Source*	*Herbarium Specimen Number*	*Mitosis root tip* 2n	*Meiosis PMC* n	*Previous Counts* *Source*	*Authority*
S. marina	Denia, Spain	C585		18^{II}		
	Coimbra, Portugal	C5911	36	18^{II}		
	Sacavém, Portugal	C5912	36	18^{II}		
			36		B	Blackburn, 1933
				18^{II}	G	Rohweder, 1939
			36		S	Nordenskiold 1942
			36		—	Tarnavschi, 1943
			36	18^{II} 16^{II} 1^{IV} 14^{II} 2^{IV}	P	Rodriquez, 1953
			36		B & P	Blackburn & Morton, 1957
				n=18 2nd mei. met.	NA	Reese, 1957
			36		Co, F, It, NA, P, Sp.	Monnier, 1960a
S. diandra	Aknoul, Morocco	C615		9^{II}		
			18		P	Blackburn, & Morton, 1957
S. purpurea	Madrid, Spain	C586		18^{II}		
	Cordoba, Spain	C5716		18^{II} 16^{II} 1^{III} 1^{I}		
	Coimbra, Portugal	C587		18^{II*}		
	Lisbon, Portugal	C588		18^{II} 16^{II} 1^{IV}		
	Marrakesh, Morocco	C616		9^{II}		

TABLE 1 (*Continued*)

Species	*Source*	*Herbarium Specimen Number*	*Mitosis root tip* 2n	*Meiosis PMC* n	*Previous Counts* *Source*	*Authority*
S. purpurea			18		P	Rodriquez, 1953
			36			Blackburn, & Morton, 1957
			18		NA	Monnier,
			36		F, P, Sp.	1962
S. rubra	Cornwall, v.c.1	C5717		18^{II}		
	Hereford, v.c.36	C589		18^{II}		
	Cheshire, v.c.58	C5718		18^{II}		
	Lancashire, v.c.59	C5719		18^{II}		
	Kuopio, Finland	C618		18^{II}		
	Turku, Finland	C619		18^{II}		
	Coimbra, Portugal	C5913		18^{II}		
+	Auderghem, Brussels (Jardin Exp. Jean Massart)	C5720		18^{II}		
+	Nantes, France	C5810		18^{II}		
	Hérault, France	C6110		n=27 2nd mei. met.		
	Zagreb, Jugoslavia (Institum Botanicum, Facultatio Agronom. Forest. Universitatis Zagreb)	C5721		n=27 2nd mei. met.		
			36		S	Nordenskiold 1942
			36		F	Monnier,1956
			36		B	Blackburn & Morton, 1957
			54		F	Monnier,1962
var. alpina Willk.			36		F	Favarger, 1956
S. nicaeensis	Hérault, France	C6111		18^{II} 16^{II} 1^{III} 1^{I}		
	Barcelona, Spain	C5811		18^{II} 16^{II} 1^{IV}		
			36		F	Monnier,1959

TABLE 1 (*Continued*)

Species	*Source*	*Herbarium Specimen Number*	*Mitosis root tip* 2n	*Meiosis PMC* n	*Previous Counts* *Source*	*Authority*
S. capillacea	Coimbra, Portugal	C5914	18	n=19 2nd mei. met.	P	Blackburn & Morton, 1957
S. bocconii	Cornwall, v.c.2	C5722	36	18^{II}		
	Barcelona, Spain	C5813		18^{II}		
	Denia, Spain	C5814		18^{II}		
	Valencia, Spain	C5815		18^{II}		
	Coimbra, Portugal	C5916		18^{II}		
	Sacavém, Portugal	C5917		18^{II}		
	Pozzallo, Italy	C6112		18^{II}		
	Palermo, Sicily	C6113		18^{II}		
	Oran, Algeria	C6114		18^{II}		
			36		B & P	Blackburn & Morton, 1957
S. heldreichii	Tangier, Morocco	C6115	36	18^{II}	NA	Simon & Monnier,1958

When the source of seed is a Botanic Garden the full title is only given in the first case, thereafter it is referred to only by its locality.

^{II}=bivalent, ^{III}=trivalent, ^{IV}=quadrivalent, ^{I}=univalent.

+=seed of unknown origin from a Botanic Garden or other source.

*=in one flower bud many figures showed a single univalent but none were good enough for detailed analysis.

B=Britain; Co=Corsica; F=France; G=Germany; It=Italy; NA=North Africa; P=Portugal; S=Scandinavia; Sp.=Spain.

TABLE 2

Observed occurrence of quadrivalents in tetraploid species

Species	*Source*	*No. of PMC examined*	*No. of cells containing quadrivalents*
S. rupicola	Cheshire, v.c.58	52	2
	Co. Antrim, v.c.H39	68	5
S. marina	Cheshire, v.c.58	63	1
	Montpellier, France	50	0
	Barcelona, Spain	55	0
S. purpurea	Coimbra, Portugal	92	0
	Lisbon, Portugal	51	2
S. rubra	Hereford, v.c.36	60	0
	Cheshire, v.c.58	54	0
	Turku, Finland	50	0
S. nicaeensis	Hérault, France	54	0
	Barcelona, Spain	55	2
S. bocconii	E. Cornwall, v.c.2	56	0
	Denia, Spain	c.60	0
S. heldreichii	Tangiers, Morocco	61	0

Discussion

A comparison of the chromosome counts obtained in this investigation with those of other workers (Table 1) shows a high degree of correspondence.

The basic number of the genus is x = 9 and of the species investigated, *S. fimbriata*, *S. media*, *S. diandra* and *S. capillacea* are diploid, whilst *S. rupicola*, *S. marina*, *S. nicaeensis*, *S. bocconii* and *S. heldreichii* are tetraploid. *S. purpurea* includes both diploid and tetraploid races[1] and *S. rubra* tetraploid and hexaploid races. The tendency for northern distribution of polyploidy in Europe already shown in the Caryophyllaceae by Blackburn and Morton (1957) is well demonstrated by *Spergularia* since *S. fimbriata*, *S. diandra*, *S. capillacea* and diploid *S. purpurea* are exclusively southern (see notes on distribution p. 293) whilst, apart from the cosmopolitan *S. media*, only tetraploids extend into North Europe. Furthermore Monnier (1960a, b) has discovered two other diploid species in N. Africa.

Blackburn and Morton (1957) have recorded a root-tip count of 2n = 36 from British *S. media*. The writer has examined herbarium specimens of the population from which this count was made and has confirmed the identification. Seeds removed from one of these specimens, however, grew into normal diploid plants suggesting the possibility that the tetraploid might have been an isolated plant (or a part of a plant) in the population; it is interesting to note in this connection that the occurrence of occasional polyploid cells in roots of *S. marina* and *S. purpurea* has

[1] Dr. P. Monnier, University of Montpellier, is at present working on the distribution of these races.

been observed by Blackburn and Morton (1957). All other counts of *S. media* from many European and North African localities have been diploid and there is no doubt that this is the normal chromosome level of the species.

The hexaploid form of *S. rubra* is the highest polyploid recorded in the genus. The collections from which counts have been made originate from Jugoslavia and the South of France so the form may well be widespread in the Mediterranean region. The plants are noticeably more robust than tetraploid *S. rubra*.

The observation of quadrivalents in *S. rupicola*, *S. marina*, tetraploid *S. purpurea* and *S. nicaeensis* is interesting even although they occur in relatively few of the pollen mother cells and are never more than one per cell. In *S. marina* quadrivalents were also recorded by Rodriquez (1953), working on Portuguese material, and he observed two in some cells (one of which he figures). The formation of quadrivalents indicates a high degree of homology in chromosomes with as low a chiasma frequency as occurs in these species. There are alternative explanations of this homology, (a) it is the result of segmental interchange, or (b) it is the result of autopolyploid or segmental allopolyploid origin of the species. Evidence relating to this problem obtained from chromosome pairing in interspecific hybrids will be presented in a later paper. The observation in *S. purpurea* and *S. nicaeensis* of cells with a trivalent and univalent is presumably the result of competition for pairing. Multivalent formation has not been observed in any of the other polyploid species but more investigation is necessary to discover if a very low percentage does in fact occur. Absence of multivalents does not, however, preclude the possibility of their autopolyploid or segmental allopolyploid origin since competition in pairing of chromosomes with low chiasma frequency would tend to cause bivalent formation. Furthermore "diploidization" might have occurred during the course of evolution so that the formation of multivalents which occurred in the "raw" polyploid is now eliminated.

Summary

Eleven species of *Spergularia* from Europe and North Africa have been investigated cytologically. The base number of the genus is $x=9$ and of the species studied *S. fimbriata*, *S. media*, *S. diandra* and *S. capillacea* are diploid whilst *S. rupicola*, *S. marina*, *S. nicaeensis*, *S. bocconii* and *S. heldreichii* are tetraploid. *S. purpurea* contains both diploid and tetraploid races and *S. rubra* both tetraploid and hexaploid races. Quadrivalents have been observed in a low percentage of pollen mother cells in the tetraploids *S. rupicola*, *S. marina*, *S. purpurea* and *S. nicaeensis*.

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