Spontaneous Polyploidization in Rumex Hybrids

Z. ŚWIETLIŃSKA

The occurrence of polyploid forms in the subgenus Acetosa of the genus Rumex has long been known. The problem has been studied mainly by Japanese workers. Tri- and tetraploid forms found in natural conditions have been described by Ono (1935). Among the progeny of such plants there were individuals with a chromosome number amounting to 50 (in the subgenus Acetosa all species are dioecious their diploid chromosome number being 2n=14 in females and 2n=15 in males). Yamamoto (1938) lists numerous polyploids found in the course of his own investigations and reported by other workers.

The data in the literature indicate that within the subgenus Acetosa polyploidy is a rather frequent occurrence in natural conditions. However, there has been no investigation on the mechanism underlying the multiplication of the chromosome number of normally diploid sorrel species or on the causes of this increase. The workers mentioned above always started their experiments with polyploid plants and by appropriate crosses obtained derived forms with varying chromosome numbers.

In the course of an investigation on cytogentic relations between three species from the subgenus *Acetosa* growing in Poland — i.e. *R. acetosa*, *R. thyrsiflorus*, and *R. arifolius* — spontaneous polyploidy developing in some interspecific hybrids has been observed.

During experiments, in which various populations of the three above mentioned sorrel species were crossed, in the case of crosses between definite populations of R. thyrsiflorus and R. acetosa characteristic disturbances of meiosis occurred in F_1 causing the multiplication of the chromosome number in the F_2 generation. The problem arose as a side issue in an investigation centred on other questions and for this reason it has not been studied in all details. Nevertheless, the data assembled up to now throw some light on the nature of the processes causing polyploidy in the genus Rumex and have thus been thought to deserve a preliminary report.

MATERIAL AND METHODS

The original material consisted of plants collected from natural habitats and grown under uniform conditions on experimental plots at Skierniewice. The hybrids were obtained by crossing previously isolated individuals of the particular species.

The cytological examinations were made on fixed preparations. Root tips were fixed with Navashin's method according to Müntzing's modification. Frequently before fixing the preparations were treated with 8-oxy-quinoline according to Levan's method. The thickness of the microtome sections was 12—17 μ . The buds for examinations of meiosis were fixed in Navashin's fluid with previous treatment for 5—10 minutes in Carnoy's solution. The thickness of sections was 14—17 μ . The preparations were stained with crystal violet according to Newton.

The parental species

1. Rumex acetosa — The plants were collected from a damp meadow in the Białystok region. The composition of chromosome sets was 2X+12A in female individuals and X+2Y+12A in male individuals. The X-chromo-



Fig. 1. Metaphasal plates from root tip cells. a-R. acetosa, female plant, 2n=14; b-R. thyrsiflorus, male plant, $2n=15\times 2700$

somes were with median constriction and the longest in the set; the Y-chromosomes were also with two arms but somewhat shorter. All the autosomes were rodshaped, had subterminal centromeres, and belonged to type i* (fig. 1.a.).

The course of meiosis in microsporogenesis was on the whole regular.

^{*} Kihara and Yamamoto (1931) established that the set of autosomes in the subgenus Acetosa consisted of four types of chromosomes: type i — rodshaped with subterminal centromere; type v — short, with almost median centromere; type j — 2-armed with one arm much shorter; and type T — rodshaped with satellites.

In diakinesis there were six bivalents and one trivalent formed of sex chromosomes (fig. 2.a.). Chromatin bridges formed because of difficulties in the separation of bivalents were sometimes visible in anaphase I (fig. 2.b, c), undivided bivalents were also frequent (fig. 2.d). In anaphase the Y—

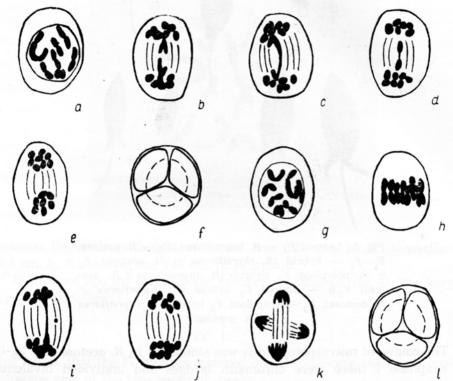


Fig. 2. Meiosis in PMCs, a-f-R. acetosa: a — diakinesis, b-e — anaphase I, f — tetrad; g-l-R. thyrsiflorus: g — diakinesis, h — metaphase I, i, j — anaphase I, k — anaphase II, l — tetrad. imes 1200

chromosomes moved to one pole and the X-chromosome to the other, consequently in each anaphase group there were either 7 or 8 chromosomes (fig. 2.e). After the second meiotic division regular tetrads were formed (fig. 2.f).

The proportion of fertile pollen grains in preparations of mature pollen stained with aceto-carmine was 94 to 98 per cent.

2. Rumex thyrsiflorus — The plants of this species had been collected on flood dams along the Vistula near Pulawy. In the autosome set, besides the rodshaped chromosomes with subterminal centromeres (type i), there were also two pairs of 2-armed chromosomes, one short with median centromere (type v), the other longer (type j), with arms of different length (fig. 1.b. and plate I_1).

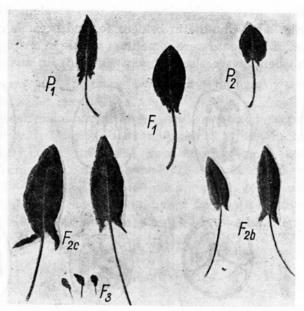


Fig. 3. Leaves: $P_1 - R$. thyrsiflorus, $P_2 - R$. acetosa, $F_1 - F_1$ — hybrid (R. thyrsiflorus \times R. acetosa), F_2 b — polyploid F_2 hybrid (R. thyrsiflorus \times R. acetosa) F_2 b — diploid F_2 hybrid (R. thyrsiflorus \times R. acetosa), F_3 — polyploid F_3 hybrid (R. thyrsiflorus \times R. acetosa)

The course of microsporogenesis was similar as in *R. acetosa* (fig. 2g-l). In anaphase I there were chromatin bridges and undivided bivalents. Moreover, in a few cells inversion bridges with acentric fragments were formed (fig. 2.i). Pollen fertility was about 97 per cent.

Morphologically the two species differed greatly. In R. acetosa (fig. 3.P₂) the leaves were wider, thinner, and less pointed at the base than in R. thyrsiflorus (fig. 3.P₁). In R. acetosa the branching of the inflorescence was simple, while in R. thyrsiflorus it was compound and more compact. The former species flowers in the second half of May and the latter in the second half of June, so that the flowering times of the two species did not coincide.

The first hybrid generation (F₁)

The first hybrid generation was obtained from the cross: $\circ R$. thyrsiflorus (No. 46) * x $\circ R$. acetosa (No. 3) The seeds from interspecific pollination for F_1 were set abundantly.

^{*} The figures indicate the numbers of individuals.

The germination rate of these seeds was nearly 80 per cent. The F_1 population was very uniform and morphologically intermediate, but manifested a marked heterosis with regard to the parental species (fig. $3.F_1$). For



Fig. 4. Metaphasal plate from root tip cell in F_1 hybrid (R. thryrsiflorus x R. acetosa), female plant 2n=14. x 2700

instance the average height of stems was 127 cm. in R. thyrsiflorus, 88.4 cm. in R. acetosa, and 146 cm. in F₁ hybrid.

In the root tips the chromosome number was diploid (fig. 4). The karyotype was as follows:

$$\left.\begin{array}{c} \circ 2X \\ \circ X+2Y \end{array}\right\}+j+v+10i.$$

This means that the F_1 hybrids received one chromosome each from every autosome pair of the two parental species.

Microsporogenesis was examined in four male individuals.

Plants No. 2 and 15: — In metaphase I rare isolated univalents were lying outside the plate (fig. 5.a) and, on the whole, in spite of the presence of the heteromorphic chromosome pair (v + j) conjugation was regular. Undivided bivalents (fig. 5.b) and chromatin bridges were frequently seen in anaphase I, (fig. 5.c), but even so the separation of chromosomes was usually regular. In anaphase groups there were 7 and 8 chromosomes (fig. 5.d). The second meiotic division usually proceeded normally (fig. 5.e), though semetimes there were chromosomes outside the plate (fig. 5.f). The appearance of the tetrads was normal. Pollen fertility vadier over a wide range. In some buds the percentage of seemingly fertile pollen grains could amount to 80, while in most buds of the same plant no normally developed grains were found.

Plants No. 16 and 29 — The conjugation during diakinesis was normal (fig. 5.g), but rare single univalents also occurred in anaphase I, besides

the chromatin bridges observed in the parental species (fig. 5.h), a high proportion of cells had bridges with acentric fragments, (fig. 5.i, j and plate I_i). Undivided bivalents were frequently seen in the spindle and

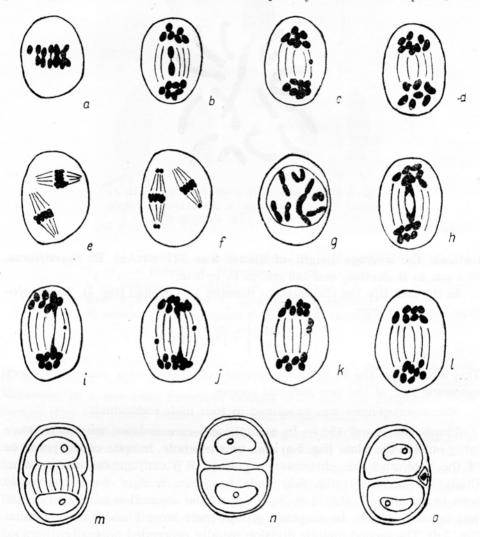


Fig. 5. Meiosis in PMC's in F_1 hybrids (R. thyrsiflorus \times R. acetosa), a-f — plant No 2: a — metaphase I, b-d — anaphase I, e-f — metaphase II; g-o — plant No. 16: g — diakinesis, h-l — anaphase I, m-o — diads. \times 1200

they sometimes moved to one of the cell poles (fig. 5.k). During anaphase I the chromosomes were divided into groups of 7 and 8 (fig. 5.l and plate I_5), but possibly in some cases undivided bivalents were also included in one of the anaphase groups (fig. 5.k).

More serious disturbance occurred in the second meiotic division. Cytokinesis took place directly after the end of the first meiotic division and as a result two daughter cells — i.e. dyads — were formed (fig. 5 m-o and plate I₆). The chromatin threads in the nuclei of the dyads were dedifferentiated and dyads either divided into two pollen grains or degenerated. The formation of tetrads was observed in neither of these two plants. This means that the second meiotic division was completely abolished. Thus, if the separation of chromosomes in anaphase I and the division of chromatids in the nuclei of the dyads were normal, the unreduced chromosome number in the resulting gametes would be 14 or 16.

Pollen fertility in the two individuals varied over a wide range. In some buds seemingly fertile pollen grains amounted to 90 per cent. These pollen grains were much larger than the pollen in the parental species. However, in most buds there were only a few large, seemingly fertile pollen grains, while the great majority were undeveloped and completely plasmolysed.

The second hybrid generation (F₂)

Because of the reduced fertility of male F_1 plants pollination for obtaining the F_2 generation was carried out in various combinations and the progeny of each parent pair was studied separately.

Pollination for the F2 generation:

♀ No 14	Xo	No 16	seeds di	d not	germina	ate		
17	X	16	,, ,,	,,	,,			
12	X	16	germination	rate	81.20/0;	104	plants	survived
12	×	15	,,	,,	75.90/0;	124	,,	,,
12	×	8	,,,	,,	$56.7^{0}/_{0};$	179	,,	,,

In the first and the second combinations there must have been some kind of antagonism between the parent plants making impossible the development of embryos and the germination of seeds.

F₂ progeny (12 x 16)

In this F_2 progeny about half of the seedlings manifested developmental disturbances already in the cotyledon stage. The cotyledons of such seedlings were small, thick, and frequently dis- or trisected; in some the growing points of stems and roots withered and the plants perished. The plants that survived developed normally and manifested much vigour. The leaves were large, thick, and frequently asymmetrical (fig. 3. F_2c); their shape as well as the shape of the inflorescence rather resembled

R. thyrsiflorus. In the progeny of 104 individuals only four plants were male and one was monoecious.

The somatic chromosome number was counted in 9 female plants. All these plants proved to be polyploids, but in the plates the chromosomes were so crowded that it was impossible to establish exactly their number and their morphology. In one plant 25 chromosomes were counted (fig. 6 and plate I₂) and in others the chromosome number was estimated at 24—27. Similar chromosome numbers were found in meiosis of the four male individuals. Since all the 13 examined plants had a polyploid

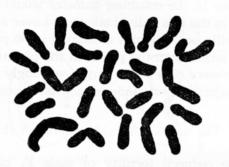


Fig. 6. Metaphasal plate from root tip cell F_2 hybrid R. thyrsiflorus \times R. acetosa (12 \times 16), female plant $2n=25.\times2700$

chromosome number and in view of the obvious morphological uniformity of all this F_2 progeny, the supposition that the whole population consisted of polyploid forms seems fully justified.

The production of unreduced gametes by the male F_1 parent and the supposed lack of disturbance in macrosporogenesis led to the conclusion that the expected chromosome number in F_2 plants should have been triploid according to the following pattern:

. 0	Norma	meiosis	Abolished second meiotic division		
•	X + 6A	2Y + 6A	2X + 12A	4Y + 12A	
normal meiosis X + 6A	2X + 12A ♀ 2n = 14	$X + 2Y + 12A$ $0^{\circ} 2n = 15$	3X + 18A $9 2n = 21$	X + 4Y + 18A $O 2n = 23$	

The occurrence in F_2 of chromosome numbers higher than triploid and lower than tetraploid is difficult to understand. The simplest explanation seems to be that in the male F_1 individuals the chromosomes were segregated irregulary during anaphase I. This supposition may be supported

to some extent by the presence of undivided bivalents which sometimes might possibly move to one of the poles. If such was the case, the additional assumption should be made that only the gametes with the extra

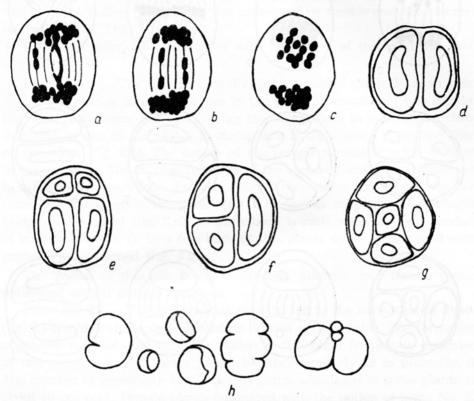


Fig. 7. Meiosis in PMC's of F_2 hybrid R. thyrsiflorus \times R. acetosa (12×16), plant No. 6: a-c — anaphase I, d — diad, e — tetrads, f — triad, g — pentad, \times 1200; h — pollen grains, \times 300, explanation in text p. 89 \times 300

chromosomes were capable of fertilization. However, since the chromosome number in F_2 has not been established exactly in a large number of individuals the above supposition has no major significance.

Meiosis was analysed in all four polyploid individuals.

Plant No. 6 — Analyses of diakinesis were made very difficult by the numerous chromosomes. Besides bi- and trivalents, and figures difficult to recognize, some univalents were present both in diakinesis and in anaphase I.

The proportion of undivided bivalents and of chromatin bridges in anaphase I was high (fig. 7.a, b and plate I_{7.8}). The number of chromosomes on each of the poles after the parting of chromosomes in anaphase I was about 11—14 (fig. 7.c and plate I₉).

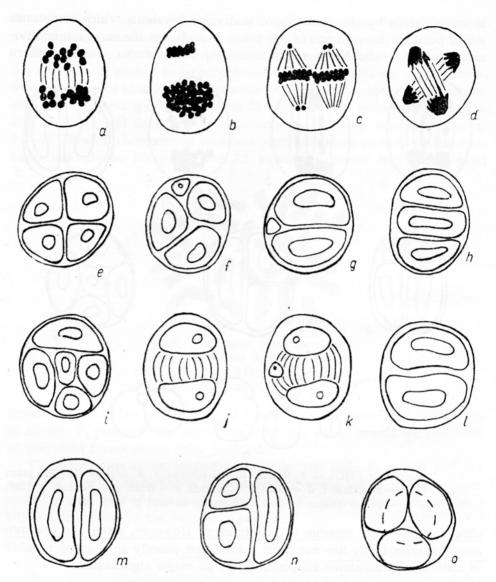


Fig. 8. Meiosis in PMC's of F_2 hybrid R. thyrsiflorus \times R. acetosa (12 \times 16), a-i- plant No. 7: a — anaphase I, b-c — metaphase II, d — anaphase II, e-f — tetrads, g — diad, h — triad, i — pentad; j-l — plant No. 21: formation of diad, explanation in text p. 89; m-o — plant No. 48: m — diad, n — triad, o — tetrad. \times 1200

The second meiotic division was disturbed by the formation of dyads immediately after the first division (fig. 7.d and plate I_{10}). In some buds triads, tetrads, and even pentads were also found (fig. 8.e-g), but the process of their formation has not been observed.

No normally developed pollen grains have been found in aceto-carmine preparations. The pollen grains differed in size, their content was completely plasmolysed, and their shape was frequently irregular. Sometimes two or three pollen grains were not separated by membranes and formed one whole (fig. 7.h).

In female individuals pollinated with the pollen of this plant no seeds were set.

Plant No. 7 — The first meiotic division proceeded similarly as in plant No. 6. The parting of chromosomes to the poles in anaphase I was regular (fig. 8.a). Sometimes, however, owing to disturbances in meiosis the chromosomes in one of the plates in metaphase II were more numerous than on the other (fig. 8.b). The course of the second meiotic division differed in various buds. The successive stages of the second division were observed in numerous preparations. In metaphase II there were sometimes chromosomes not included in the plates (fig. 8.c) and in anaphase II chromatin bridges were noted (fig. 8.d). In such buds tetrads were the end product of meiosis (fig. 8.e, f). In a few buds of this plants dyads, triads, and even pentads were formed (fig. 8.g,i).

The pollen of this plant was completely abortive and female plants pollinated with it did not set seeds.

Plant No. 21 — The first meiotic division led to the formation of dyads (fig. 8.j-l) which either degenerated or formed pollen grains. The formation of tetrads was not observed. The pollen of this plant frequently consisted of two or three grains partly joined together similarly as in plant No. 6. The number of seemingly fertile pollen grains amounted in some plants to about 50 per cent. Female plants pollinated with the pollen of plant No. 21 developed large well formed seeds, but the fertility of the plants, as compared to F_1 , was markedly reduced.

Plant No. 48 — The course of meiosis was here similar as in plant No. 21. Dyads were usually formed in large numbers (fig. 8.m), but in some buds there were triads (fig. 8.n) and normal tetrads (fig. 8.o). The pollen grains were large and well developed, though sometimes they consisted of dyads, were degenerated and plasmolysed. The percentage of seemingly fertile pollen grains in some buds amounted to 50. Female plants pollinated with the pollen of this plant developed large well formed seeds, but the seeds were few.

The monoecious plant was completely sterile. Meiosis was probably inhibited already in the course of microsporogenesis at the stage of diakinesis, since later stages of division were not observed.

All freely pollinated female plants developed large but very few and usually empty seeds.

F₂ progenies (12 x 8) and (12 x 15)

The F_2 seedlings of these progenies (280 individuals) developed normally (see leaves fig. 3. F_2 b). The segregation of morphological traits characteristic for the two parental species was strongly marked. The somatic chromosome number in the 12 plants examined was diploid. This was the anticipated chromosome number since the course of meiosis in the male F_1 plants No. 8 and 15 had been normal. Moreover, in the female F_1 plants normal haploid gametes must have been produced, because the female plant No. 12 gave a diploid progeny after pollination with pollen from male plants with regular meiosis. The two F_2 progenies now considered differed only by the proportion of male individuals. In F_2 (12x8) out of 160 plants 25 were male individuals and in F_2 (12x15) there were only 4 male plants in a population of 120.

The course of meiosis during microsporogenesis was examined only on smears with aceto-carmine. It is of interest that also in this F_2 progeny

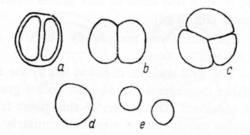


Fig. 9. Pollen grains in F_2 hybrid R. thyrsiflorus \times R. acetosa (12 \times 8) and (13 \times 15). Explanation in text p. 90×300

dyads were formed in some individuals after the first meiotic division, although the trait had not been manifested by the F_1 male parents. Most of the dyads degenerated (fig. 9.a), but sometimes they developed into large well formed pollen grains (fig. 9.d), which in some cases remained joined in pairs retaining the original arrangement of the dyads (fig. 9.b). In some plants the tendency not to separate was displayed also by pollen grains developed from tetrads. Such tetrads had then the appearance of enormous pollen grains but on closer examination the walls separating the particular grains were visible (fig. 9.c). Finally, in this progeny there was also a certain proportion of pollen grains that were smaller and these probably had developed from normal tetrads (fig. 9.e).

Out of 30 male plants 17 produced seemingly normal pollen the fertility of which ranged 30 to 75 per cent. Ten plants produced, besides normal

pollen, also pollen grains in the form of dyads or tedrads and single very large grains. In these ten plants the proportion of seemingly fertile pollen ranged 5 to 80 per cent. In the three remaining plants the very few pollen grains were all completely degenerated.

The feritility of the female plants in this F_2 population is still unknown since controlled pollinations for obtaining the F_3 generation have not as yet been carried out. The female individuals which flowered on the first year after sowing out (none of the male plants flowered in that year) developed seeds from free pollination, but the seeds were very few and malformed, which seems to indicate that their fertility was greatly reduced.

The third hybrid generation (F₃)

The third hybrid generation was obtained from the F_2 progeny (12 \times 16). Owing to the high sterility of F_2 plants the seeds for F_3 were obtained only from the following crosses:

F ₂ ♀ No 29	$9 \times $ \circ No	21,	240	seeds	of	which	52	germinated
5	$6 \times$	21,	10	,,	,,	,,	3	,,
	\times	48,		,,			4	,,
4	4×	48,				,,	4	,,

All the seedlings of the third hybrid generation manifested pronounced developmental anomalies. All the cotyledons were small, thick, dis- or



Fig. 10. Metaphasal plate from root tip cell of F_3 hybrid, $2n = 41 \times 2700$

trisected, and had a very irregular shape. The few leaves which developed were very small, thick, and strongly deformed (fig. 3. F_3). The plants perished without forming flower shoots.

In four plants of this F3 progeny it was possible to examine a few mitotic plates. One plant had 41 chromosomes (fig. 10) and one about 40. In the other two plants the chromosome number was estimated at 26—27 and 35-37. This means that in the third hybrid generation as compared * to F₂ a further increase of the chromosome number had occurred. However, cytological examinations in F3 as well as in F2 were insufficient to analyse fully this process. Observations lead only to the conclusion that in F2, similarly as in F1, functional male gametes must have been unreduced. The property of producing unreduced gametes, consisting in the abolishment of the second meiotic division, was thus transferred from F₁ to F2. The first stage of the process led to the appearance of the polyploid F₂ generation manifesting much vegetative vigour and fertility strongly reduced with regard to the parental species or the F1 hybrids. The next stage of the polyploidization caused a further increase of the chromosome number in the F3 generation, but the plants were very weak and actually non-viable.

Backrosses

Because of the differences in the time of flowering only backcrosses with *R. thyrsiflorus* were possible. The backrosses were as follows:

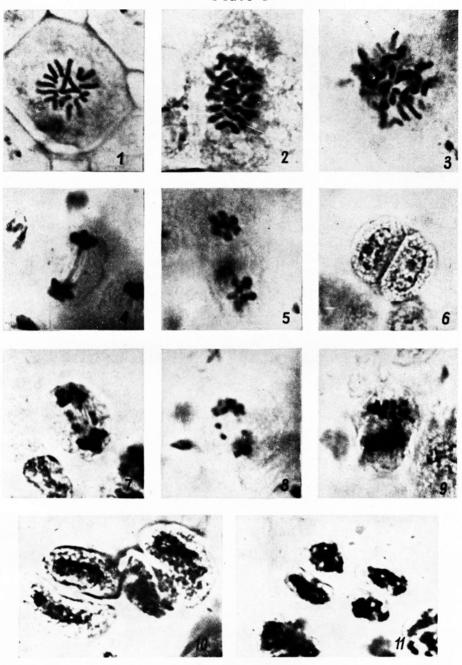
- ♀ R. thyrsiflorus No. 46 × ♂ F₁ No. 8
- Q R. thyrsiflorus No. 46 × F₁ No. 15
- $^{\circ}$ F₁ No. 12 \times $_{\circ}$ R. thyrsiflorus No. 43.

The following plants of the F_1 generation were used for the backcrosses: 9 No. 12, \circlearrowleft Nos. 8 and 15. The same hybrids were also the parent plants of the F_2 generation described above. The female plant No. 46 of R. thyrsiflorus was the original parental form of the first hybrid generation. The attempted backcross with the male F_1 plant No. 16 was unfortunately unsuccessful.

Backcrosses \circ R. thyrsiflorus $\times_{\circlearrowleft}$ F_1

The progeny from the backcrosses of the male F_1 plants Nos. 8 and 15 differed greatly, though the other parent was in both cases the same female R. thyrsiflorus plant.

The progeny from the backcross with the F_1 plant No. 15 was on the whole similar to the F_2 population, but individuals morphologically similar to R. thyrsiflorus predominated. In this backcross, similarly as in the F_2 progeny (12×15), the proportion of male individuals was very low (6 in 105). The backcross with the F_1 plant No. 8 also produced individuals



predominantly similar in their morphology to R. thyrsiflorus, though the plants differed from the preceding backcross progeny in some characteristic features. The most remarkable of these traits was the greatly reduced number of leaves and flower shoots. Every plant developed an average of about 10 thick and strong flower shoots, whereas on the plants of the preceding backcross the number of flower shoots was about 30. The few leaves that developed were larger and thicker so that this backcross had a more vigorous appearance. The proportion of male individuals was relatively high (40 in 200) similarly as in the F_2 progeny (12×8). There was also one monoecious plant which was completely sterile.

The somatic chromosome number in the four examined individuals of the backcross with plant No. 15 was 2n=14 or 15. In the backcross with plant No. 8 there was one triploid plant (2n=21) for 25 individuals with the normal diploid chromosome number. This means that the male plant No. 8 must have produced a small proportion of unreduced functional gametes.

The backcross \circ $F_1 \times \circ R$. thyrsiflorus

This backcross manifested no special features. The plants morphologically resembled R. thyrsiflorus and their growth habit was more uniform than in the two other backcrosses described above. The proportion of male individuals (10 in 204) approached the average. The somatic chromosome number counted in 15 plants was 2n = 14 or 15.

Microsporogenesis in the backcrosses

The most important property of microsporogenesis in all the three backcrosses consisted in the formation of dyads observed in male individuals after the first meiotic division, similarly as was the case in \mathbf{F}_1 and \mathbf{F}_2 .

The course of meiosis was investigated in 12 male plants. In seven individuals no serious disturbances were observed. The conjugation during diakinesis was regular (fig. 11.a) and in anaphase I, in spite of the frequent inversion bridges (fig. 11.b, c) and the presence of undivided bivalents (fig. 11.d), the chromosomes segregated so that 7 and 8 of them moved to either pole (fig. 11.e), The second meiotic division proceeded normally (fig. 11.f, g) and led to the formation of normal tetrads (fig. 11.h).

In some buds of the other five plants a transversal wall was formed between the nuclei after anaphase of the first meiotic division (fig. 11.i). However, after the appearance of the separating wall the nuclei of the

czasowe badania w zakresie eksperymentalnej ekologii roślin dotyczą najczęściej roślin kultywowanych i nie były przeprowadzane w naturalnych siedliskach.

Z tego rodzaju badań, przeprowadzonych w naturalnych siedliskach, należy wymienić ogólna w tym zakresie prace W. A. Kowdy (1956) i S. W. Zonna (1956) o wzajemnym oddziaływaniu gleb i leśnej roślinności. Na uwage w tych pracach zasługuje dynamiczne ujęcie zagadnienia funkcji mineralnego składu roślin zielnych i drzew w procesach glebowych w biocenozie leśnej. Autorzy ci wysuwają postulat badań składu mineralnego roślin w powiązaniu z dynamiką procesów glebowych. Praca J. Wehrmanna (1955), przeprowadzona na poletkach doświadczalnych, mówi o ilościowej zależności występowania manganu, miedzi i kobaltu w roślinach od typu gleby, czasu pobierania prób do analiz i składu gatunkowego zbiorowiska roślinnego. Jest to praca najbliższa założeniom chemicznej ekologii roślin. W dotychczasowych pracach o zawartości mikroelementów w roślinach tylko niewielu badaczy uwzględnia w rozważaniach czas zbioru materiału do analiz, jakkolwiek niektórzy autorzy zwracają uwagę, że zawartość mikroelementów w roślinach zmienia się w okresie wegetacyjnym wraz z rozwojem roślin. O konieczności uwzględniania stadium rozwoju rośliny w tego rodzaju badaniach mikroelementów pisał S. Macko (1956).

Na szczególną uwagę zasługuje praca Smitha i jego współpracowników (A. Maksimow 1954), którzy badali w ciągu dwóch lat zmiany w zawartości manganu przez cały okres wegetacyjny w liściach drzew wiecznie zielonych. Badacze ci stwierdzili, że zawartość manganu w młodych liściach była mała, natomiast w liściach, w których procesy rozwoju i wzrostu zostały zakończone, stężenie manganu było większe i na ogół stałe. Podobne badania nad zawartością manganu w roślinach uprawnych przeprowadził Snider (A. Maksimow 1954) i wykazał wzrost zawartości manganu w roślinach wraz z ich rozwojem wegetacyjnym. J. Wehrmann (1955) zaś zwraca uwagę na różnice w zawartości manganu, miedzi i kobaltu w roślinach w okresie kłoszenia się i w początkowym okresie kwitnienia. N. Karlsson i O. Svanberg (1952), badajac zawartość kobaltu w koniczynie w różnych okresach jej wzrostu, stwierdzili stopniowy wzrost zawartości tego mikroelementu w roślinie, aż do czasu kwitnienia. A. P. Szczerbakow i M. S. Turkowa (1956) wychodząc z założenia, że fizjologiczna rola manganu jest związana z procesami utleniającymi, które najbardziej intensywnie przebiegają w młodych rosnących tkankach i organach, uważali, że słuszne jest prześledzenie ilościowego rozmieszczenia manganu w różnych stadiach rozwojowych igieł drzew iglastych. Jest to jedyna praca specjalnie poświęcona

dyads did not pass immediately to the resting stage but entered the prophase and as if the metaphase of the second division. At that stage the chromosomes were already divided into two chromatids but were still connected at the centromere (fig. 11.j and plate I_{11}). Typical metaphase plates with division spindles were never observed in the dyads. Nevertheless, in older anthers, besides the dyads (fig. 11.k), there were also triads and tetrads usually arranged in one plane (fig. 11.l-m), which

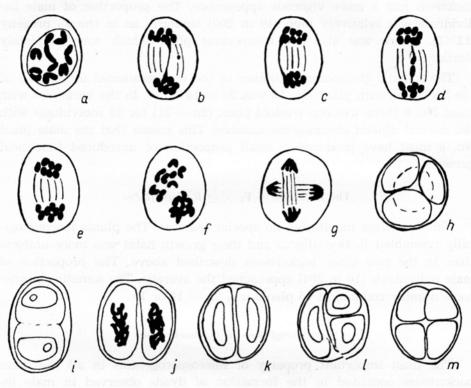


Fig. 11. Meiosis in PMC's in backcross (Q R. thyrsiflorus \times \bigcirc F_1), a-h — regular meiosis: a — diakinesis, b-e — anaphase I, f — metaphase II, g — anaphase II, h — tetrad, i-m — meiotic disturbances: i — formation of diad, j — prophase in the nucleus of a diad, k — diad, l — triad, m — tetrad; \times 1200

seems to indicate that the second meiotic division in such buds must have greatly diverged from the normal course, but the problem necessitates further, more detailed investigations.

In the plants manifesting meiotic disturbances described above in some buds the first and the second meiotic divisions had an absolutely regular course.

In the majority of plants from the backcrosses the proportion of seemingly well developed pollen grains ranged 25 to 87 per cent. In six plants

all the pollen was completely degenerated and in three individuals there were pollen grains connected in dyads and tetrads.

In the experiments carried out so far the backcrosses have proved to be completely sterile, in spite of the normal vegetative development and the relatively high, at least in some plants, percentage of seemingly well developed pollen grains. Well formed viable seeds were developed from none of the 13 controlled pollinations that have been carried out. Only a few malformed seeds were gathered and this seems to indicate that fertilization might have occurred in some cases, but the development of endosperm or embryos was inhibited.

DISCUSSION

From the cytological standpoint the most interesting observation made in the course of this investigation on the hybrids between R. acetosa and R. thyrsiflorus was the modification of meiosis during microsporogenesis in the hybrids. After the first meiotic division dyads were formed and the second meiotic division was omitted partly or completely. This property was first manifested in some male F_1 plants and was transferred to the F_2 generation as well as to the backcrosses by individuals with both modified and unmodified meiosis.

Numerous examples of the formation of dyads in natural Rumex acetosa polyploids were described by Ono (1935). In the progeny of such plants Ono observed the frequent further increase of the chromosome number. He assumed that the increase of the chromosome number must have been caused by unreduced gametes, but he did not investigate the process of their formation.

The failure of the second meiotic division in pollen development was for the first time observed by Levan (1936) in various Allium species and was called by him monokinetic meiosis. According to Levan the mechanism of monokinetic meiosis is not influenced by the structural properties of chromosomes but is controlled genically. As a result of this kind of meiosis diploids can give rise to triploids and, in turn, triploids give rise to pentaploids. Levan even described a form with 108 chromosomes (the diploid chromosome number in Allium is 2n=16) obtained from a hyperoctoploid (2n=68).

Another instance of polyploidy caused by the failure of the second meiotic division was reported in the hybrids of Musa (Dodds and Pittendrigh 1945). In this case the unreduced chromosome number was introduced into the crosses by female gametes. In a later investigation Dodds and Simmonds (1946) observed the omission of the second

or both meiotic divisions also in microsporogenesis. However, the dyads and the monads, with respectively the zygotic and the double zygotic chromosome number, were uncapable of functioning, since such male hybrids were completely sterile.

The process of the formation of dyads in the first meiotic division and of the abolishment of the second meiotic division observed in sorrel is quite certainly analogical to the monokinetic meiosis in *Allium* described by Levan. The genic nature of the mechanism involved in this process is indicated by that the property of forming dyads is transferred to the further generations even by male and female plants with a normal meiosis. In the further generations the mechanism is to some extent differentiated. For instance in F_2 the formation of dyads was sometimes restricted to some buds, while at the same time in the other buds of the same plant the second meiotic division proceeded without major disturbances. In the backcrosses and in the F_2 generation derived from a F_1 plant with normal meiosis the process was restricted to some plants only and even then did not affect all the buds in one plant.

It follows from all that has been said above that in the *R. thyrsiflorus* population from Puławy there must be a gene or a set of genes which, in combination with the genome of *R. acetosa*, cause the abolishment of the second meiotic division and the formation of functional unreduced pollen grains. These genes are probably restricted to some populations only, since a female plant of *R. thyrsiflorus* belonging to a different population, when pollinated with pollen from the same male plant of *R. acetosa* as the one used in the other cross, produced only a diploid progeny.

The genetic system causing meiotic disturbances is probably effective only during microsporogenesis. This is indicated by that the same female F_1 plant produced polyploids when it was crossed with a male plant producing unreduced gametes, and diploids when it was pollinated by pollen from a male plant with a normal meiosis. As is well known from numerous reports (Beadle, McClintock 1928, Beadle 1930, Rees, Thompson 1956, Lawrance 1958) the particular stages of meiotic divisions are controlled by various genes. The majority of these reports deal with the first meiotic division. The results of the present investigation indicate that also the course of the second division may be controlled by specific genes which in no way influence the first division.

The information assembled so far seems to suggest that the activities of these genes may be a source of polyploidy. It is possible that in the case of the genus *Rumex* the process has no phylogenetic significance, since the species of this subgenus are dioecious, and since polyploidy causes disturbances in the sex determination. At any rate the problem cannot be explained without further investigations, especially in view of

the serious differences in the proportion between the number of female and male plants and the occurrence of monoecious plants.

Very noteworthy is also the genetic system which automatically increases the chromosome number in every successive generation. The numerous cytological and genetic problems arising in this connection will be the subject of future researches.

SUMMARY

1. Crosses between certain populations of R. thyrsiflorus and R. acetosa gave in F_1 , alongside of plants with normal meiosis, some male individuals producing unreduced gametes. In the plants with meiotic disturbances the formation of dyads after the first meiotic division and the complete omission of the second division were observed.

2. F_2 plants and backcrosses obtained from F_1 individuals with normal meiosis had a diploid chromosome number (\circ 2n = 14 and \circ 2n = 15). The F_2 progeny of male individuals with modified meiosis consisted of

polyploid plants (the chromosome number was about 25).

3. The property of forming dyads after the first meiotic division was transferred to both the diploid and the polyploid F_2 progeny as well as to the backcrosses.

- 4. In F_3 plants obtained from the polyploid F_2 plants there was a further rise of the chromosome number up to 2n = 41. However these plants were non-viable.
- 5. The results obtained indicate that in some *R. thyrsiflorus* populations there are hereditary factors controlling the course of the second meiotic division.

My very sincere gratitude is due to Professor W. Gajewski for his kind guidance and valuable advice in the course of this work.

Department of Genetics, Polish Academy of Sciences,
Laboratory of Experimental Taxonomy, Warsaw. (Entered: 6.8.1959)

......

Beadle G. W., 1930, Genetical and cytological studies of Mendelian asynapsis in Zea mays, Cornell Univ. Agr. Exp. Sta. 129: 1—23.

REFERENCES

Beadle G. W., and B. McClintock, 1928, A genetic disturbance of meiosis in Zea mays, Science 68: 433.

Dodds K. S. and C. S. Pittendrigh, 1945, Genetical and cytological studies of Musa VII. Certain aspects of polyploidy, Journal of Genetics 47, 1.