CYTOTAXONOMIC STUDIES IN CARDAMINE PRATENSIS L. IN THE NETHERLANDS

C. C. BERG

(Botanical Museum and Herbarium, Utrecht)

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ABSTRACT

In material of *Cardamine pratensis* from many localities in the Netherlands, the following somatic chromosome numbers were counted: 28, 30, 31, 32, 34, 36, 38, 39, 45, 46, 52, 53, 54, 56, 58, 59, 60, 62, 64, 64 à 66, 67, 68, 69, 70, 72, 73, 74, 75, 76, 78, (80), 84, and (118) [1]). Many numbers were due to intra-individual deviation of the normal number. As main groups tetraploids with 28–32, octoploids with 56–64, and decaploids with 70–80 chromosomes could be distinguished; the normal numbers of the ploidy levels were 30, 60, and 74–76, respectively. Two subspecies could be distinguished on the basis of morphological, ecological, and cytological characters.

INTRODUCTION

In the morphologically and ecologically very variable species *Cardamine pratensis* L. a large number of cytotypes has become known from many countries. In most cases only one or a few individuals were examined. In investigations by BANACH (1951), HUSSEIN (1955), and LÖVKVIST (1947, 1956) a larger number of individuals was involved. Moreover, Banach studied the cytological differentiation in connection with ecology, and Lövkvist in connection with morphology, ecology, and distribution, completed with crossing experiments. These studies showed regional differences in the cytological differentiation of *C. pratensis*.

LÖVKVIST (1956) distinguished in the temperature zone of Europa 6 species in the *C. pratensis* complex. Only *C. pratensis* L. s. str. and *C. palustris* Petermann could be expected in the Netherlands. These species could be distinguished cytologically, ecologically, and morphologically; moreover, restrictions in compatibility were evident. In *C. pratensis* L. s. str. common chromosome numbers were 16, 30, and 44 (being at the di-, tetra-, and hexaploid level, respectively). Chromosome numbers of *C. palustris* Petermann proved to be 56 or higher (being at the octo-, deca-, and dodecaploid level). There are indications that the two taxa cannot be separated satisfactorily in all parts of the distribution area, while the morphological discontinuity does not always appear to be correlated with the same ploidy level (see SCHULZ, 1903; LINDMAN, 1914; BANACH, 1951; HUSSEIN, 1955; LÖVKVIST, 1956; CLAPHAM, TUTIN and WARBURG, 1962).

[1]) Numbers in parentheses could not be determined exactly.

A positive correlation between the chromosome number and the water content of the substrate was evident (BANACH, 1951; LÖVKVIST, 1956).

The primary object of this study was to get some impression of the cytological differentiation of *C. pratensis* in the Netherlands and to investigate if and how the above mentioned taxa could be distinguished.

MATERIAL AND METHODS

From several parts of the Netherlands one or some individuals from a population were collected. Most of them were grown in experimental gardens. Chromosome counts were made on metaphase plates of roottip cells. The roottips were fixed in Karpechenko, embedded in paraffin, sectioned at 15 μ, and stained according Heidenhain's haematoxylin method. In each preparation at least 3 metaphase plates were examined. Voucher specimens of the investigated plants have been deposited in the Herbarium of the State University of Utrecht.

CYTOLOGYCAL RESULTS

The plants often showed intra-individual variation of the chromosome number, usually within the limits of the ploidy level (see LÖVKVIST, 1956). In most of these plants, however, one or possibly 2 numbers can be considered to be the normal numbers of the ploidy level, as they are usually present in the plant or within the population.

The following somatic chromosome numbers were counted, as the only number found in the plant or together with other numbers: 28, 30, 31, 32, 34, 36, 38, 39, 45, 46, 52, 53, 54, 56, 58, 59, 60, 62, 64, 64 à 66, 67, 68, 69, 70, 72, 73, 74, 76, 78, (80) [1]), 84, and (118).

Tetraploids (**2*n* = 28–32**)

30 was counted in plants from about 50 localities; it is the normal number at this ploidy level and the commonest one in *C. pratensis* L. s.l. in the Netherlands. 28 was found three times together with 30 and/or 32; 31 twice, and 32 fourteen times as the only number or together with other numbers at this level. There were indications that the numbers 31 and 32 had been caused by chromosome fragmentation. Sometimes 60 or c. 60 has been found as an endopolyploid number.

Octoploids (**2*n* = 56–64**)

Octoploids are known from 4 localities listed in Table 1. They always occurred together with tetraploids and sometimes with decaploids, too. 60 appeared to be the normal number at the octoploid level. This number usually occurred together with other numbers

[1]) Numbers in parentheses could not be determined exactly.

TABLE 1

Origin of the material and collection numbers	Somatic chr. numbers [1])
Monnikendam: 483C, 487C	30
489	32
475	36, 38
476	38
487E	45
478	46, (45)
482	52, 53
479, 481, 483D	60
484, 486D	68
485B	67, 68, 69
486C	72, 73, 74
480	74, 76, (72)
Between Keizersveer and Sleewijk: 429, 431, 433, 436, 437	30
432	28, 30, 32
435	32
430, 434A, 434B, 438	68
Meerkerk: 439	34, 36, 39
440	74, 76, 78, (68)
Brabantse Biesbos near Drimmelen:	
425, 526, 495, 498, 503F, 507B	30
500	28, 30, 32
494	58
496	58, 60, 64, (118)
505C	60, 62, (58)
499D	59, 60, 62, 64
502B	56, 64
503A	60, 62, (64)
Near Ulvenhout, loc. I: 414, 415, 416	30
413	32
loc. II: 418	30
417, 419	30, 31, 32
Near Bavel: 420, 421, 422, 423	30
424	60
Near Etten-Leur: 402, 407, 408, 505D, 506	30
403, 405, 504	74
411, 415	76
404, 511	72, 74, 78, (68)
410	74, 78
406	54, 84
Near Terheyden (N.B.): 398	30
401	32
399, 400	76
Near Oostvoorne: 253–255, 258–260, 266–268	30
256, 257	32
445B	64, 68
445G	68
445H	64 à 66
445I	68, 72, (64)
446A	58, 60, 62
446D	59, 60, 62
446E	60, 62, (58), (64)
Near Rockanje: 263, 264, 448	30
447E	68, 70, 74, 76
447A	70, 76
Near Kortenhoef: 452A, 452B	74, 76, 78
454A, 454B	76

[1]) Numbers in parentheses could not be determined exactly.

of this level, although in some plants 60 could not be found. Once c. 118 was found as an endopolyploid number.

Decaploids (**$2n = 70$–80**)

Decaploids were met with in 13 localities besides those mentioned in Table 1 near Utrecht, Westbroek, Eemnes, Lemmer, Oosterhout (N.B.), and Heelsum. 74 and 76 appeared to be the normal numbers; these numbers could occur together and/or in combination with other numbers at this level. Once 75 was counted as the only number.

Other chromosome numbers

In populations with decaploids plants were found with 34 and 36, or 38, or 36, 38, and 39 chromosomes. Plants with 45 or 46 chromosomes were met with together with tetraploids and octoploids; one plant with 52 and 53 chromosomes in a population with tetraploids and decaploids.
68 chromosomes were counted in individuals originating from some neighbouring localities, probably in pure breeding populations. The number of 68 occurred once together with numbers at the decaploid level.
68 chromosomes were also found to occur in combination with other numbers in plants originating from populations containing octoploids or decaploids and octoploids.
54 chromosomes were once found in a preparation in which all other counted metaphase plates showed the number $2n = 84$.

When roottips of the same plant were fixed at different moments, it often appeared that one preparation showed, e.g., only 32 chromosomes and the other only 30, or that the one appeared to have a varying chromosome number at the decaploid level, while in the other, for instance, only 76 were counted.

Table 1 gives a selected list of chromosome counts, with indication of the origin of the plants and their collection numbers.

Crossing experiments

Some crossings were made between plants of different ploidy levels. The results were partly investigated and appeared to be very confusing. In the framework of this publication should be mentioned that plants with c. 68 chromosomes were obtained from crossings between octoploids and decaploids and an individual with c. 45 chromosomes from a crossing between a tetraploid and an octoploid.

Restrictions in compatibility between octoploids and decaploids on the one and tetraploids on the other side appeared to be more or less evident.

Discussion

The somatic chromosome numbers counted by several workers up to 1956 are, according to tables given by Lövkvist (1956): 16, 28,

30, 32, 33, 34, 37, 38, 40, 43, 44, 45, 46, 48, 50, 52, 54–68, 70–78, 80, 84, 88, > 90, and c. 96. Other chromosome numbers were not known so far. New numbers obtained from the present study are 31, 36, 39, 53, 69.

8 is the basic number in *Cardamine*. According to LÖVKVIST (1956, 1963) 7 may be conceived as a second basic number. Combinations of these numbers can theoretically give several euploids at the each ploidy level (e.g. at the octoploid level 56, 58, 60, 62, and 64). This classification, suggested on account of chromosome numbers in *Cardamine* and the distribution of longer chromosomes in the cytotypes of *Cardamine pratensis*, has been taken over in this study, although not for the same reasons.

The presence of intra-individually variable chromosome numbers, called aneusomaty (DUNCAN, 1945), in tetra-, octo-, and decaploids, is a remarkable phenomenon which formerly was not known in *C. pratensis*. It might be caused by chromosome fragmentation and chromosome deletion. The latter possibility was suggested by LÖVKVIST (1956) for aberrant chromosome numbers in pure breeding populations of a high ploidy level. The possibility of chromosome fusion was also pronounced (LAWRENCE, 1931; LÖVKVIST, 1963).

LÖVKVIST (1956) regarded as pure breeding populations in the South of Sweden and Danmark those with $2n = 30$, (38), 56, (60), 64, 68, 72, 76, 80 and c. 96. In general most other numbers would have been caused by hybridization. In the Netherlands pure breeding populations with $2n = 30$, 60, 68 and 74–76 appeared to occur. The number of hybrids seems to be rather small. In the British Isles 30 and 56 are common somatic chromosome numbers, while 32, 38, 48, 57, 58, 64 and 72 have been counted incidentally (HUSSEIN, 1955; LÖVKVIST, 1956). In Poland 30, 32, 44, 50, 58, 64, 72, 76 and 78 have been counted (BANACH, 1951). Thus the cytological differentiation in *C. pratensis* in the Netherlands appears to differ more or less from that of other regions where tetraploids, octoploids, and decaploids occur.

As plants of each ploidy level showed aneusomaty, it is conceivable that these levels are connected by autoploidization. The distribution of the octoploids indicated the possibility that they are autoploids of tetraploids.

Plants with chromosome numbers between 32 and 40 might be subhaploid developments from octoploids or decaploids, as already suggested by LÖVKVIST (1956).

Individuals with $2n = 45$–46 and 52–53 probably are hybrids of tetraploids on the one side and octoploids or decaploids on the other side, whereas those with 68 or c. 68 chromosomes may be hybrids of octoploids and decaploids.

MORPHOLOGY

Several characters have been studied to find differences between octoploids and decaploids on the one side and tetraploids on the

other. Table 2 gives some more or less useful differentiating characters.

In most cases the characters of the uppermost cauline leaves are sufficient to distinguish both groups. It was impossible to distinguish octoploids from decaploids. Plants with chromosome numbers between the tetraploid level and the octoploid level mostly resembled the tetraploids.

In general these results confirm those of LÖVKVIST (1956).

TABLE 2

Characters	Octoploids and decaploids	Tetraploids
uppermost cauline leaves	pinnate; leaflets, except the terminal one, deciduous	pinnatifid
lateral racemes	mostly absent or few	mostly present and more numerous
(terminal) racemes	fewer flowers on the average	more flowers on the average
rosette-like lateral shoots	normally present	normally absent
flowers and most floral parts	larger on the average	smaller on the average
sepals	always longer than 4 mm	nearly always smaller than 4 mm
anthers	2.0–2.7 mm long	1–2.0 mm long
flowering time	as a whole later	as a whole earlier

ECOLOGY

Decaploids usually occurred in situations where the roots reach the (ground-) water always or for the greater part of the year. Once a population with $2n = c.70$ was found in a streamlet 40–50 cm below the water level (Heelsum).

Tetraploids were usually met with in dryer habitats. Their roots seldom reach the (ground-)water. They showed a rather great amplitude with regard to the water content of the soil. Although octoploids have been found in the same places as tetraploids, they usually prefer wetter habitats.

These records confirm those of BANACH (1951) and LÖVKVIST (1956).

TAXONOMY

The results obtained from this study demonstrate the presence of two groups of individuals in *C. pratensis* L. in the Netherlands. These groups are cytologically separable, nearly always morphologically distinguishable, and ecologically different, restrictions in compatibility are more or less evident. This is a confirmation of the results of LÖVKVIST (1956).

Instead of assigning specific status to the groups, the author prefers to treat them as subspecies, namely octoploids and decaploids as *C. pratensis* L. ssp. *palustris* (Wimm. and Grab.) Janch., and tetraploids as *C. pratensis* L. ssp. *pratensis*.

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