THE IDENTIFICATION OF FOUR NEW MONOSOMICS IN COMMON OATS *AVENA SATIVA* L.

BY

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ABSTRACT

Eight monosomic lines produced by X-irradiation and one derived spontaneously from a population of common oats of variety Garry were studied to identify the missing chromosome. Enlarged photomicrographs of well-spread mitotic figures of each line provided the basis for critical analysis of the missing chromosome. The chromosomes were cut out from the photographs, matched and mounted on metric paper. A total of four new monosomic lines were added to those already identified by McGinnis in the variety Garry.

Line R 176-376, deficient for chromosome 14, exhibited a distinct phenotype, i.e., white striations in the leaves of seedlings which were more pronounced on nullisomic plants than on monosomics. In addition they were subintermediate fatuoid and had abaxial curling of leaves. A hemizygous ineffective dominant gene (or genes) appeared to condition white striations on leaves, while the genes for subintermediate fatuoid and abaxial curling of leaves acted as complete dominants. One plant monotelosomic for the short arm was similar to nullisomics suggesting that genes conditioning all characteristics were located on the long arm of chromosome 14.

Sister lines R 252-452, -453, -456, and -457 were

deficient for the same chromosome 14, but lines -452, -453, and -456 all had a heterozygous deletion in the upper part of the short arm of chromosome 19, while -457 was homozygous for the deletion. The missing part of the short arm had an effect on the fatuoid expression when chromosome-14 was lost.

R 524-575 was deficient for chromosome 6. Hybrids between this line and R 176-376, had 19 pairs and two univalents. The two univalents were not distributed randomly but were oriented in close proximity to each other, indicating homoeology is probably shared by these two chromosomes.

Line R 364-535, deficient for chromosome 18, displayed a kinky neck phenotype when it was nullisomic. A high frequency of monotelosomics for the long arm seemed to be specific for this line. It is suggested that the gene or genes for normal neck are on the long arm of chromosome 18 since these monotelosomics appeared normal.

In line R 355-508, the critical chromosome was number 4; nullisomics were late in flowering and extremely slow-growing.

R 222-413, R 263-477, and 21-139-136 were all identified as mono-15 and R 267-492 was monosomic-21. Both these lines had been obtained prior to this study.

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INTRODUCTION

Common oats is an allohexaploid comprised of the A, C, and D genomes (24). Because of the polyploid nature of this species, the deficiency of a single chromosome and even a pair of chromosomes is tolerated, so that it should be possible to produce a series of all 21 monosomics. Such a series has been completed by Sears (26) in common wheat which is similar to oats in chromosome number and ploidy level. The value of this monosomic series has been widely recognized.

It is reasonable to assume that a monosomic series in common oats would prove equally useful in cytogenetic studies. Two main sources were found productive by Sears, namely the progeny of a haploid plant pollinated with normal pollen and the progeny of a partially asynaptic nullisomic (3B). Unfortunately such sources have not been so productive in common oats. Only one haploid has been obtained thus far (19) and asynaptic nullisomics are sterile both when selfed and crossed.

As pointed out by Rajhathy (22) the idiogram of hexaploid oats shows considerable variation in chromosome size and in position of centromere, and many of the different monosomics can be identified from karyotype studies. Thus lack of progress in establishing a complete monosomic series has been mainly due to the lack of a promising

source of aneuploids. Recently a large number of monosomics have been produced at this institution by X-irradiation. The present study was conducted on a number of these monosomics in order to identify them.

LITERATURE REVIEW

SPONTANEOUS OCCURRENCE OF ANEUPLOIDS

Huskins (8) in 1927 was the first one to observe a monosomic plant in oats which was characterized by a fatuoid phenotype. He classified the oat fatuoids into three series α, β, r (9). The heterozygous fatuoid of series β was associated with a deficiency for the "C" chromosome. Normal types and heterozygous types were found to segregate in ratios of 1:5 to 1:10 plus a few nullisomics which were homozygous fatuoids. The 40-chromosome fatuoids had irregular meiosis and were dwarf and sterile.

Nishiyama (17,18) reported that monosomic heterozygous fatuoids segregated into normal types, heterozygotes and dwarf, asynaptic homozygotes. In the progeny of heterozygous fatuoids, some off-types were found which were monotelosomic for either the long arm or the short arm of the submedian "C" chromosome. Plants monotelosomic for the short arm were heterozygous fatuoids, semidwarf, asynaptic and highly sterile. Plants monotelosomic for the long arm were homozygous fatuoids, dwarf, synaptic and partially fertile. Nishiyama concluded that the gene or genes for fatuoid characteristics are located on the short arm and the gene or genes for synapsis are on the long arm of the "C" chromosome.

Philp (20) was able to demonstrate that chlorophyll deficiency is associated with a nullisomic condition. In the advanced generation of a cross between Avena sativa and Avena fatua, one family segregated 1 green: 6 albino seedlings. The green plants were monosomic, whereas the albinos were all nullisomics.

Philp (21) reported on a nullisomic with a narrow leaf width which was found in the progeny of the same interspecific cross between Avena sativa and Avena fatua. Measurements and cytological observations showed that seedlings whose leaf width was less than 3 mm had 40 chromosomes and the others had 41 chromosomes. The ratio was approximately 1 broad: 2 narrow. This indicated that a gene for normal leaf width is located on this chromosome.

McGinnis and Taylor (15) showed that albinism in an intervarietal cross of Avena sativa resulted from the loss of a whole chromosome pair. Monosomic and disomic plants were green. In the progeny of selfed monosomics, a high frequency of nullisomic plants resulted (63.9%) indicating that the deficient pollen grains underwent little or no certation and competed successfully in fertilization. They identified the chromosome as the shortest one of the complement, namely chromosome 21, on the basis of the idiogram prepared by Rajhathy (22). An albino plant monotelosomic for the short arm was later isolated which

indicated that the gene for chlorophyll production is located on the long arm of this chromosome (16).

McGinnis and Andrews (4) identified a second chromosome involved in chlorophyll production. The nullisomic plants derived from a selfed monosomic segregated in a ratio of 3 green: 1 albino. This meant the gene constitution of the monosomic plant was VvV-. The chromosome carrying the gene for chlorophyll production was found to be the second shortest one, namely chromosome 15. One albino was monotelosomic for the short arm making it possible to locate the gene for chlorophyll production on the long arm of this chromosome also.

Joshi and Howard (10) noted that meiosis was not completely regular in Avena sativa, homologous chromosomes occasionally failing to form a bivalent. Riley and Kimber (25) checked the chromosome numbers of 631 seedlings of the variety Sun II, and found seven plants with 41 chromosomes and one with 34 chromosomes. This represented a frequency of spontaneous aneuploids of 1.3%. They suggested that the extraction of a whole monosomic set by selecting spontaneous monosomics might be possible in the cultivated oat. McGinnis (13) in an attempt to produce an aneuploid series in hexaploid oats looked for spontaneous aneuploids in the variety Garry. Out of 4,023 seedlings, he found 24 aneuploids comprised of 2 nullisomics, 17 monosomics, and

5 trisomics. This represented a frequency of spontaneous aneuploids of 0.6%.

In 1963 Hacker and Riley(6) obtained 6 nullisomics, 40 monosomics and 4 trisomics from 3,453 seedlings of the variety Sun II, a frequency of about 1.2%. In a study of the 40 monosomics, Hacker and Riley (7) grouped them into 13 distinct classes based on morphology of their nullisomic progeny and karyotype differences. Three categories of asynaptic nullisomics were also reported.

Lafever and Patterson (11) found a male sterile oat plant in a sib line of Clintland 60, which was analyzed later to be due to a nullisomic condition. From crosses of the nullisomic line with disomics, monosomic F_1 hybrids were obtained. Univalent transmission rate in F_1 hybrids was similar in both the male and female, about 6%. No certation effect for the 20-chromosome gametes was detected.

Gauthier and McGinnis (5) studied a spontaneous monosomic of Garry oats, which exhibited a kinky panicle neck. This character was exaggerated in the nullisomic condition. The chromosome involved was identified as chromosome 20 on the basis of the standard karyotype of Rajhathy (22). Two plants monotelosomic for the short arm exhibiting the exaggerated kinkiness of the nullisomics suggested that the long arm carried a gene for normal neck.

INDUCED ANEUPLOIDS

(a) X-irradiation

Costa-Rodrigues (4) facilitated the production of monosomics by X-irradiating young oat panicles at a dose of 300 r. He obtained 20 monosomics out of 279 progeny analysed. Such a high frequency of monosomics (7.2%) indicated that irradiation is an efficient method of inducing them. McGinnis (1962, unpublished) irradiated the variety Garry and obtained 41 monosomics out of 506 progeny analysed (8.1%). Costa-Rodrigues (4) suggested that X-irradiation induces a break in the two chromatids of a chromosome which by illegitimate reunion would form a dicentric chromosome. Such a chromosome is eventually lost in subsequent chromosome divisions.

In view of the success realized by the earlier investigations, a project to determine the most efficient level of X-irradiation was conducted by Andrews and McGinnis (1). Preflowering panicles of the varieties Garry and Rodney of Avena sativa were X-irradiated at 75 r, 150 r, 300 r, and 600 r. They found the frequency of aneuploids increased with the level of irradiation. The 600 r treatment produced the most aneuploids, but the greatest degree of chromosome damage as well. As a result the monosomic frequency was not increased above that obtained from the

300 r dose. The 150 r treatment produced fewer aneuploids than the higher doses, but all were normal monosomics. It was concluded that the most effective level was from 150 r to 300 r. A total of 53 monosomics were produced in Garry and 60 in Rodney by this method.

Rajhathy and Dyck (23) using low doses of X-irradiation to treat preflowering panicles of oats, found 15.5% aneuploids from irradiated panicles. They suggested the use of X-irradiation is the most efficient way to produce oat monosomics.

Chang and Sadanaga (3) obtained six monosomics (A to F) in the variety Cherokee by X-irradiating at 600 r. The breeding behaviour, morphology, karyotype, and intercrossing results were studied. They failed to differentiate the 6 monosomes by karyotype analysis. However, morphologically, monosomes C and F were obviously different from others, while A, B, D, and E differed less obviously as far as morphology was concerned.

(b) Haploids

Rajhathy and Dyck (23) attempted to screen haploids from a large population of common oats. From 672,000 seedlings in three oat varieties they found 148 twins but no haploid was found.

In a similar study in Kanota, a variety of Avena

byzantina which produced a high frequency of twin seedlings, Nishiyama (19) found a haploid. By pollinating the haploid with normal pollen, six monosomic lines were obtained.

(c) Interspecific Hybridization

Rajhathy and Dyck (23) found that late generations of backcrossed pentaploid hybrids produce a high frequency of aneuploids. The authors suggested that because of the high degree of heterozygosity and heterogeneity in these lines, this method is unsatisfactory. In addition, since the A genome is common to all species of Avena, theoretically only 14 different monosomics could be produced.

(d) Transmission Rate

The production and maintenance of monosomic lines depends largely on the behaviour of the univalent chromosomes. Lin (12) carried out an investigation of the influence of genotype and environment on univalent transmission in two monosomics, mono-15 and mono-21 of Avena sativa. It was shown that when meiosis and fertilization occurred under the lowest temperature level tested (50°F), the influence of seed source and variety on the univalent transmission rate of three different lines of mono-21 was not obvious, but at 60, 70, and 80°F the transmission rate showed a positive difference among them. The difference was especially manifest at 80°F. In general the monosomic

lines obtained from X-irradiation had a higher transmission rate than the same monosomics obtained spontaneously.

MATERIALS AND METHODS

The variety Garry was developed at the Canada Department of Agriculture Research station, Winnipeg, Manitoba, from the cross Victoria X (Hajira X Banner) X Victory. This variety was distributed widely in Manitoba and North Eastern United States.

The monosomic lines used in this study were produced by X-irradiation at a rate of 300 r (McGinnis, 1962, unpublished). Karyotype analysis was conducted on eight of them in order to identify the deficient chromosome. In addition, one monosomic, 21-139-156, obtained spontaneously was also studied. These monosomics were grown in a growth cabinet in 1962 and in the field in 1963 and 1964. The irradiated monosomic lines employed are as follows:

Monosomic	Origin
R 176-376	Garry, X-irradiation 300 r (McGinnis, 1962, unpublished)
R 222-413	"
R 252-452	"
-453	
-456	
-457	
R 263-477	"
R 267-492	"
R 355-508	"
R 364-535	"
R 524-575	"
21-139-156	Garry, Spontaneous (McGinnis, 1961)

Seeds of each monosomic line were treated with Arason, put in plastic containers in vermiculite saturated with water

and kept at 0-2°C for two days to break dormancy. The germination boxes were then left at room temperature until the roots were 2-3 cm in length. Root tips about 1 cm were excised, pretreated in ice water for 24 hours at 0-2°C and fixed in acetic-alcohol. After three days the root tips were hydrolyzed in 1N HCl at 60°C for 10 minutes, stained with Feulgen and examined by the aceto-carmine squash technique. Chromosome counts were made on mitotic metaphase. Monosomics, nullisomics, disomics and telosomics were sorted out and grown in 5-inch pots in the greenhouse. A panicle was collected from each plant prior to anthesis in order to confirm the chromosome counts made from the seedling root tips, and at the same time to observe if any chromosome aberrations had been induced by X-irradiation. Any observed morphological differences between the monosomics or nullisomics and the disomics were assumed to be the result of the deficiency of a particular chromosome.

A critical analysis of the karyotype of each monosomic line was conducted. Well-spread mitotic figures were selected for chromosome measurements which were made by camera lucida drawings, microprojection drawings and on enlarged photomicrographs. Enlarged photomicrographs were found to be particularly valuable in identifying the monosomics. Chromosomes were cut out from the photographs, matched, and mounted on metric paper according to the

groupings of the standard karyotype proposed by Rajhathy (22).

By this method, certain of the chromosomes could be readily identified. Lines found to be monosomic for chromosomes belonging to the same group, and similar in chromosome morphology, were intercrossed and the 40-chromosome progeny analysed at metaphase I of meiosis. Configurations of 20 bivalents in inter-line hybrids would indicate that the parent lines were deficient for the same chromosome, but 19 bivalents plus two univalents would show that the parent lines were deficient for different chromosomes.

RESULTS AND DISCUSSION

For clarity and brevity, each monosomic line examined critically will be dealt with separately. Emphasis is placed on morphology, karyotype, and univalent transmission in each monosomic line.

R 176-376

(a) Morphology

In this line the nullisomics were shorter than normal, reaching 3/4 normal height under field conditions, but only slight difference observed in the greenhouse (Plate I, Fig 1.). The leaves were narrower and panicles smaller. The plants were so highly sterile that a seed set of only 0.5% was obtained. These morphological differences were common to the other nullisomics in this study and to some of the nullisomic lines studied by Hacker and Riley (7).

White striations in the leaves of seedlings, appeared to be specific for this aneuploid line and could be used as a marker to detect the presence of monosomics and nullisomics. On nearly all the leaves of nullisomic plants, the white stripes were present. Striations did not appear on all the leaves of monosomic plants, and when present were not so pronounced as on the nullisomic plants. They were however sufficiently

PLATE I. Characteristic morphology of monosomic line R 176-376.

Figure 1. Comparison in height of a monosomic and nullisomic plant.

Figure 2. White striations on leaves of 41- and 40-chromosome plants.

Figure 3. Abaxial curling of a leaf of a nullisomic plant.

Figure 4. Subintermediate fatuoid seed with a slightly twisted awn and pubescent sucker mouth.

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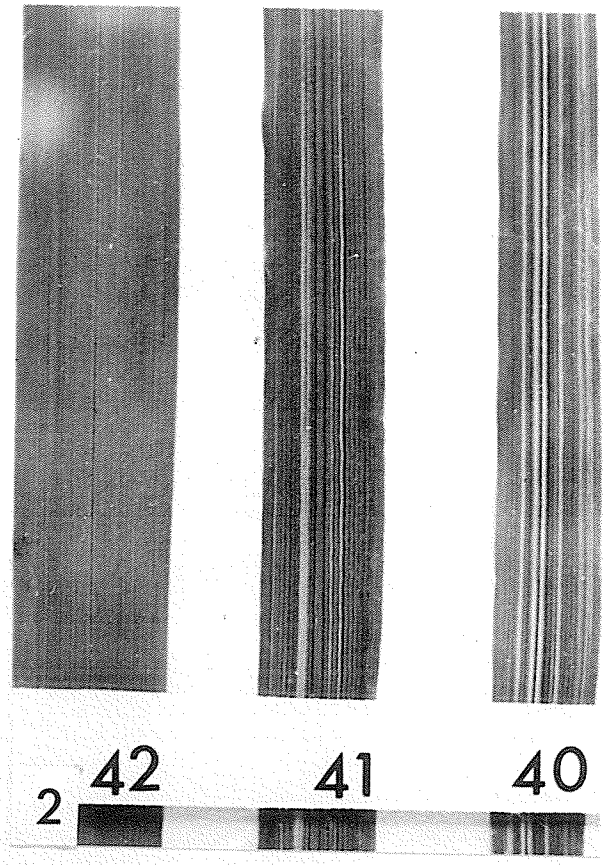

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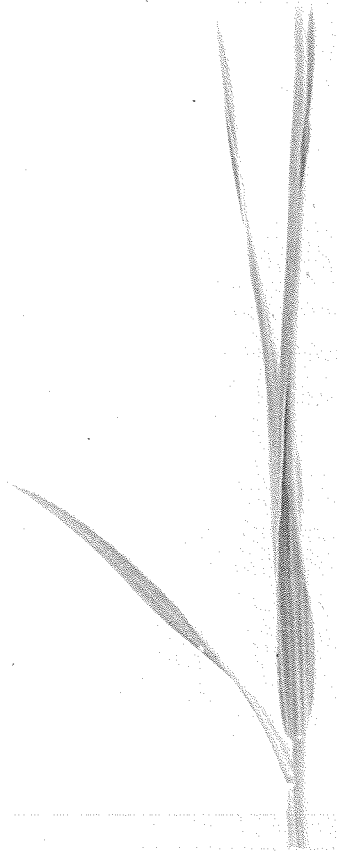

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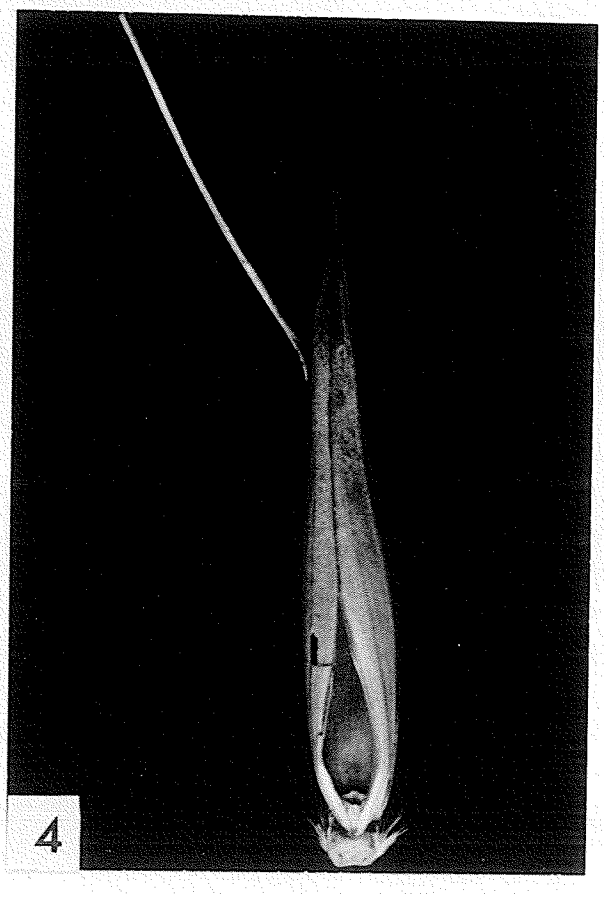

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prominent, especially under the field conditions (Plate I, Fig.2) so that they were readily recognized. In addition to this characteristic, the nullisomics displayed abaxial curling of leaves, that is they curled backward away from the stem (Plate I, Fig.3). This characteristic was observed by Hacker and Riley (7) on their Class II nullisomics, which differed from R 176-376 in its linkage with kinky neck of the panicle which has been reported to be associated with chromosome 20(5). Hacker and Riley suspected that a translocation between chromosome 14 and 20 has occurred. Chang and Sandanaga (3) also observed the abaxial curling of leaves in their nullisomic D. Whether a translocation is involved or not remains to be demonstrated by making a cross between Garry and Sun II. Nullisomics in line R 176-376 also had spikelets that were of the subintermediate fatuoid type which consisted of a pubescent sucker mouth and a slightly twisted awn (Plate I, Fig.4). Although it was not as exaggerated as the true fatuoid line, it was still easily recognized.

Monosomic plants were crossed to disomics and 32 seeds were produced. All were found to be monosomics and when grown in the greenhouse, these monosomics had striped leaves. In the F_2, four monosomics and four nullisomics were grown in the greenhouse. The nullisomics displayed a

pronounced striping on the leaves, and the monosomics exhibited white stripes as well. Thus it appears that two doses of the dominant gene are required for normal leaf color. In the hemizygous state, one dose of the dominant gene is ineffective in producing the normal leaf color. The gene or genes for both the abaxial curling of leaves and the subintermediate fatuoid chracteristic were completely recessive since these characters were only expressed in the nullisomic condition.

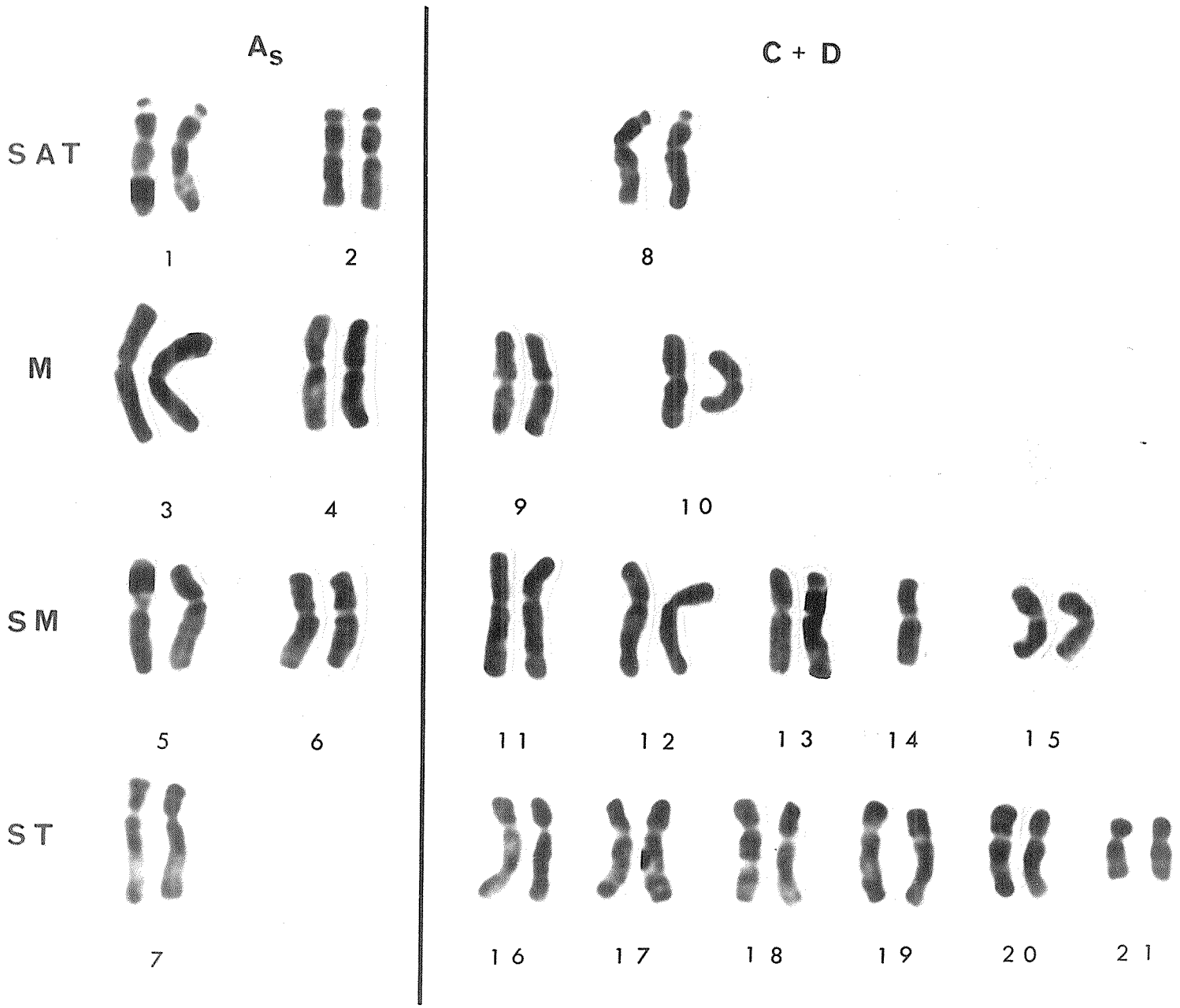
(b) Karyotype

From a critical analysis of 6 well-spread mitotic figures, it appeared that the missing chromosome is the second shortest one in the submedian group, namely chromosome 14 (Plate II, & Plate III, Fig.1). One plant monotelosomic for the short arm (Plate III, Fig.2) was identified among the 226 seedlings analysed. It also displayed the same phenotype as the nullisomics but had a higher fertility of 5%. This indicated that the long arm of chromosome 14 carries the genes for normal leaf color, abaxial curling of leaves, and subintermediate fatuoid characteristic.

(c) Univalent Transmission

A count of the micronuclei at the tetrad stage of meiosis provided the data for determining the theoretical

PLATE II. Karyotype of monosomic-14.


A_S
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21

univalent transmission rate in the male. The presence of no micronuclei in a tetrad indicates two gametes with 21 chromosomes, and two with 20 chromosomes, while the presence of one micronucleus indicates one gamete with 21 chromosomes, and three with 20 chromosomes. Where two or more micronuclei were observed in a tetrad it was concluded that all four gametes were deficient. These results are presented in Table 1.

TABLE 1. Number of micronuclei in tetrads of monosomic-14 plants and calculation of univalent transmission frequency.

Number of micronuclei per tetrad	Number of tetrads	Number of gametes	Total gametes with n=21	Percent gametes with n=21
0	105	420	210	50
1	393	1,572	393	25
2	405	1,620	0	-
3	19	76	0	-
	922	3,688	603	16.3%

To study the female transmission rate, 32 seedlings from crosses between monosomics and disomics were analyzed for chromosome number. All were found to be monosomics, indicating a very low univalent transmission rate through the female. This is again demonstrated in Table 2, in

which the data on the chromosome number of progeny of selfed monosomic are presented.

TABLE 2. Frequency of 42-, 41-, 40 + telocentric, and 40-chromosome oat seedlings from a selfed monosomic-14.

Chromosome number	Number of seedlings	Percentage of total
42	1	0.45%
41	86	38.10%
40 + telo	1	0.45%
40	138	61.00%
	226	100.00%

The disomics occurred in a frequency of only 0.45% which is in accordance with the observations by Philp (21), McGinnis and Taylor (15), McGinnis and Andrews (14), Chang and Sadanaga (3), Lin (12), Lafever and Patterson (11), and Gauthier and McGinnis (5).

R 252-452, -453, -456, -457

(a) Morphology

These sister lines were similar in certain features

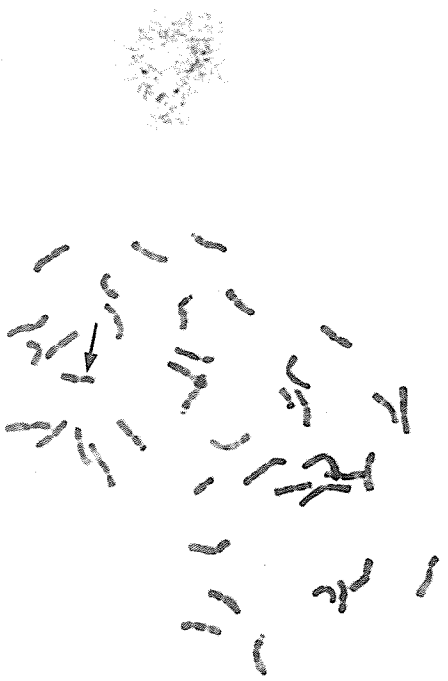
to the above aneuploid, which involved chromosome 14. For instance, the monosomics and nullisomics had striped leaves but the stripes were not as pronounced as for nulli-14, possibly because these nullisomics were too weak to express the character properly. Nullisomics were completely sterile while monosomics had a very low seed set.

The distinguishing phenotype which characterized these monosomic lines was the fatuoid expression which was intermediate, with a twisted awn on each spikelet and a hairy articulation surface on each grain in monosomic lines -452, -453, -456 (Plate IV, Figs.1 & 2). The monosomics in line -457 were identified as true fatuoids with a twisted awn and pubescent sucker mouth on each grain (Plate IV, Figs. 3 & 4). The nullisomics of all four sister lines had the true fatuoid expression. In the progeny of lines -452, -453, and -456, true fatuoid monosomics in addition to intermediate fatuoid monosomics were observed occasionally, and one monosomic plant with normal panicles (Plate V, Fig.1) was identified in the progeny of line -453. Line -457 bred true as far as the fatuoid character was concerned. A detailed description of the different types of plants observed in these four lines is presented in Table 3.

PLATE III. Mitotic metaphases of monosomic-14.

Figure 1. Mitotic metaphase of a monosomic-14 from which the karyotype is prepared (see arrow).

Figure 2. Mitotic metaphase of a monotelosomic for the short arm (see arrow).



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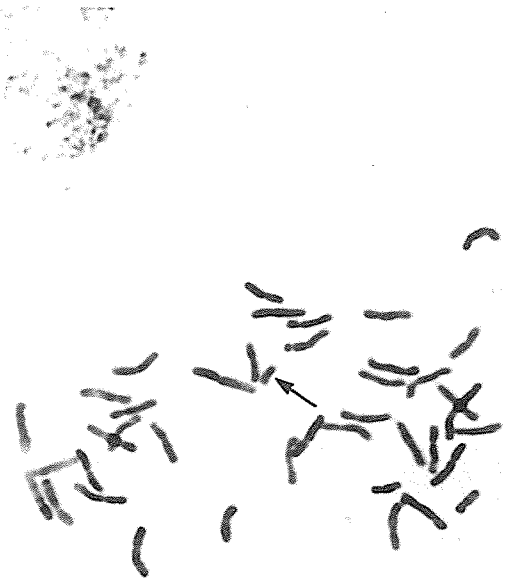

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PLATE IV. Characteristic morphology of monosomic line R 252.

Figure 1. Intermediate fatuoid seed with a long twisted awn and hairy articulation surface.

Figure 2. Panicle of a monosomic plant deficient for chromosome 14 and heterozygous for a deletion on chromosome 19 (Note an awn on each spikelet).

Figure 3. Comparison of morphology of a true fatuoid seed and normal one.

Figure 4. Panicle of a monosomic plant deficient for chromosome 14 and homozygous for a deletion on chromosome 19. (Note two or three awns on each spikelet).

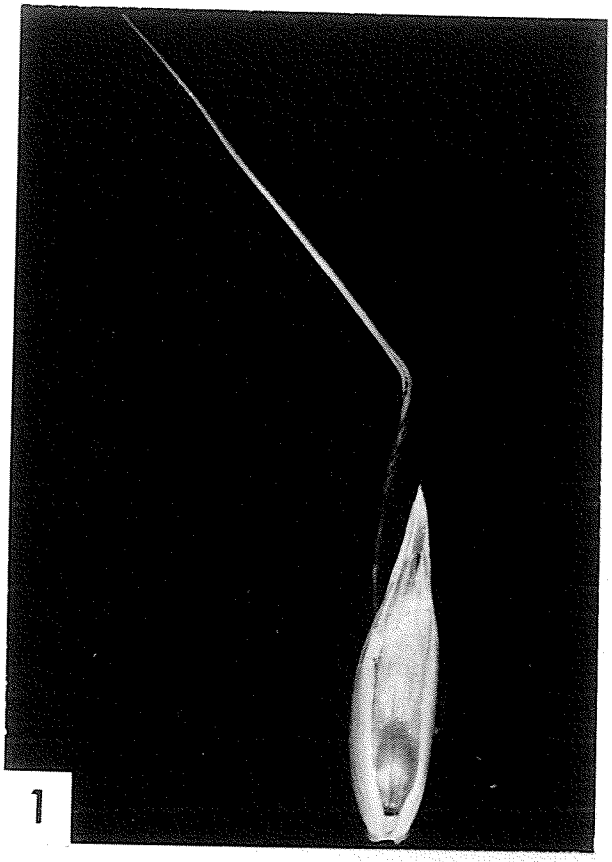

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PLATE V. Panicle morphology and mitotic metaphase of line R 252.

Figure 1. Normal panicle of a monosomic-14 plant obtained from R 252-453.

Figure 2. Panicle of a nullisomic-14 plant homozygous for a deletion on chromosome 19.

Figure 3. Mitotic metaphase of a monosomic-14 plant heterozygous for a deletion on chromosome-19 (see arrow).

Figure 4. Mitotic metaphase of a monosomic-14 homozygous for a deletion on chromosome-19 (see arrows).

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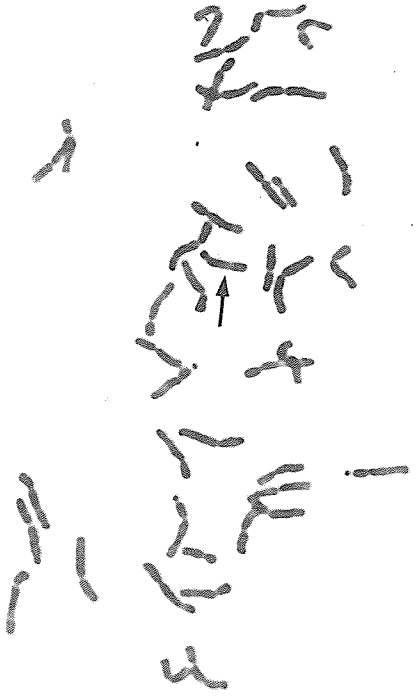

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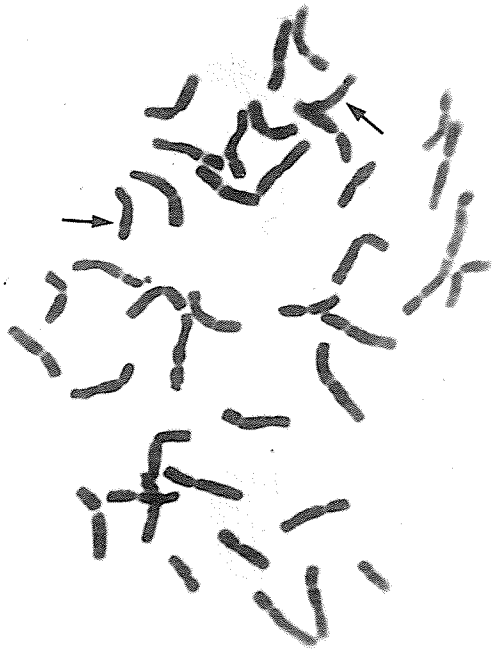

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TABLE 3. A description of different kinds of plants from lines R 252-452, -453, -456, and - 457.

	R 252-452 and R 252-456 (Heterozygous for deletion on chromosome-19)	R 252-453 (Heterozygous for deletion on chromosome-19)	R 252-457 (Homozygous for deletion on chromosome-19)
Nullisomics	weak, dwarf plant, narrow leaves, true fatuoid, striping leaves not pronounced, completely sterile	similar to -452	similar to the other nullisomics except the third awn of a spikelet was more obvious. (Plate V, Fig.2)
Monosomics	rather healthy plant, striped leaves, intermediate fatuoid, low seed fertility of about 10%. (Plate IV, Figs. 1 & 2)	similar to -452	rather healthy plant, striped leaves, true fatuoid, the third awn of a spikelet not so obvious, seed fertility of about 10%. (Plate IV, Figs.3 & 4)
Monosomics with normal panicle	nil	Normal plant without fatuoid expression, seed fertility higher, (about 25%). (Plate V, Fig.1 & Plate VI, Fig. 1)	nil
Disomics	normal, healthy plant, with good seed fertility of about 40%	similar to -452	similar to -452 (Plate VI, Figs.2 & 3)

(b) Karyotype

Careful cytological studies on 15 well-spread mitotic figures of these lines revealed the probable reason for the different morphology among them. All were deficient for chromosome 14 but in addition the upper part of the short arm of a subterminal chromosome was deleted in the lines which exhibited the intermediate fatuoid appearance, i.e. -452, -453, and -456 (Plate V, Fig.3). The subterminal chromosome was critically studied and appeared to be chromosome 19. Line -457 was found to be homozygous for the deletion on the chromosome 19 pair (Plate V, Fig.4). One normal monosomic plant from line -453 displaying no fatuoid characteristics and good fertility was analysed cytologically. The mitotic configurations showed that both chromosome-19 were normal and the fatuoid phenotype was not detected in its progeny (Plate VI, Fig.1). Disomics screened out from lines -452, -453, -456 and -457 were all normal (Plate VI, Figs. 2 & 3).

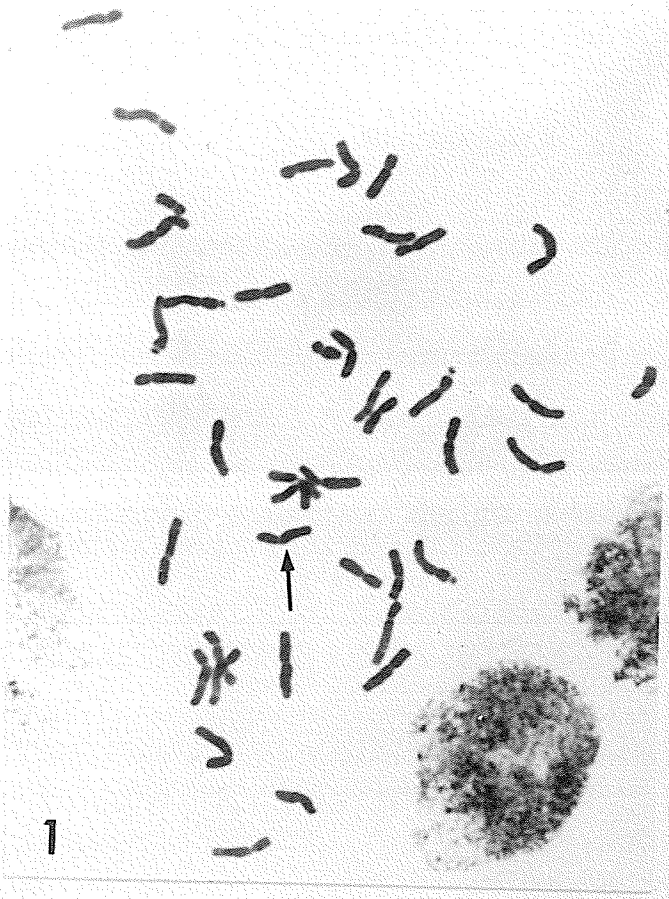
It was noteworthy that the deletion of the upper part of the short arm of chromosome-19 resulted in fatuoid expression in monosomic-14. Thus the gene or genes on the deleted part appear to function as a weak inhibitor of the fatuoid character. When chromosomes-14 are lost, the fatuoid character is expressed. The fact that chromosome-14 is more important in fatuoid expression is indicated by the fact that

PLATE VI. Mitotic metaphases of line R 252, and a panicle of a 42-chromosome plant homozygous for a deletion on chromosome-19.

Figure 1. Mitotic metaphase of monosomic 14 with a normal univalent (see arrow).

Figure 2. A panicle of a 42-chromosome plant homozygous for a deletion on chromosome-19.

Figure 3. Mitotic metaphase of a 42-chromosome plant homozygous for a deletion on chromosome-19 (see arrows).



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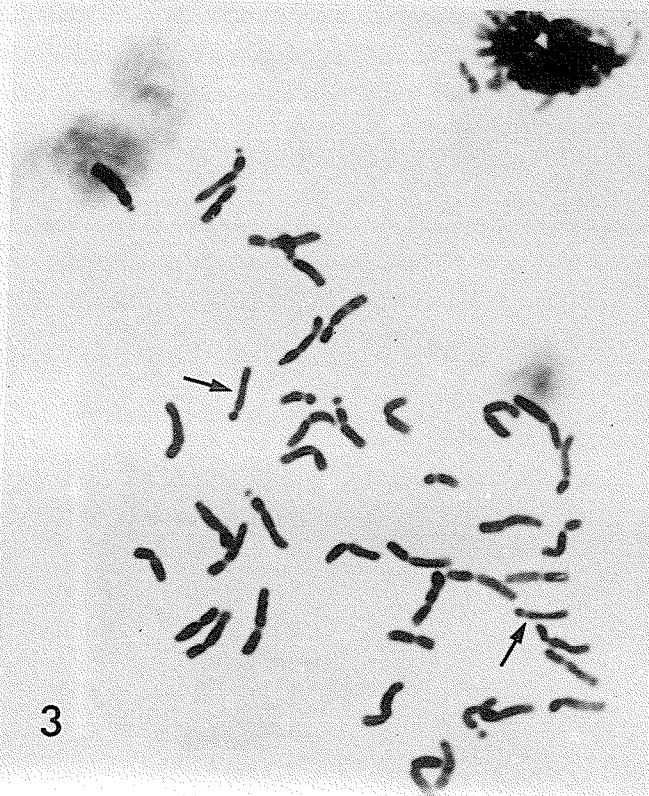


3

disomics homozygous for the deletion of the upper part of the short arm of chromosome-19 were normal. Whether chromosome-19 was the same one associated with fatuoidy by Huskins (8), Nishiyama (17,18), and Chang and Sadanaga (3) is unknown. If so, a strong fatuoid inhibitor might possibly be present on the lower part of the short arm.

Crosses between R 176-376 and R 252 were attempted but unfortunately no seeds were obtained from more than 50 pollinations. Nevertheless, 7 monosomics from R 252 back-crossed with disomics all displayed the phenotype of striped leaves again indicating that R 252 is deficient for chromosome 14.

R 524-575

(a) Morphology

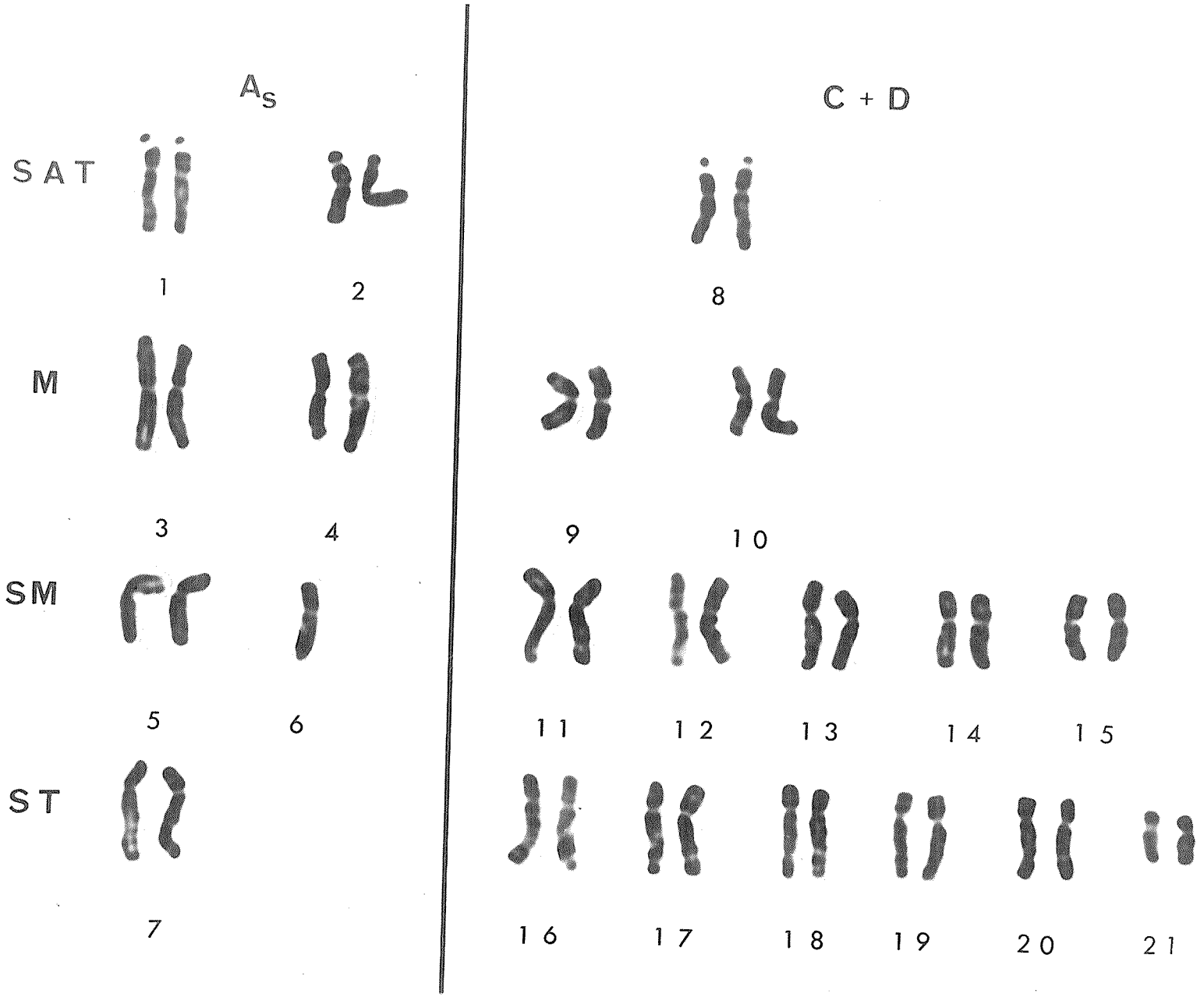
There was relatively little difference between the morphology of the nullisomics and monosomics of this line, and the disomics. Attempts to distinguish them phenotypically were in vain. The seed fertility of monosomics and nullisomics in the greenhouse was very low, but quite high under field conditions (above 30%).

(b) Karyotype

Critical examination of 11 enlarged photographs of well-spread mitotic configurations disclosed that the critical

chromosome involved was chromosome-6 (Plate VII, & Plate VIII, Fig.1). In some of the photographs the difference between chromosome-6 and chromosome-14 could not be always observed. Therefore a cross between monosomics of these two lines was made as a further step to verify that they were different. Five seeds were obtained from 45 pollinations, two of which failed to germinate. The rest grown in greenhouse gave rise to very dwarf plants which tillered profusely but had no white stripes on their leaves. The meiotic configurations of these F_1 hybrids were very interesting. Nineteen pairs and two univalents were observed in all 100 pollen mother cells analysed at diakinesis and metaphase I. In nearly all cells analysed at diakinesis, however, the univalents were not distributed randomly but were oriented in close proximity to one another; T-shaped configurations were most often present (Plate VIII, Figs. 3 & 4). In only one case were the two univalents arranged side by side (Plate VIII, Fig.2). Wide separation of the two univalents occurred in an extremely low frequency, and no configuration of 20 bivalents was recorded. There was little doubt that the two chromosomes were non-homologous, but their behaviour strongly suggested homoeology with a tendency to attract each other but not sufficient homology to pair. At metaphase I, the two univalents tended to be separated (Plate

PLATE VII. Karyotype of monosomic-6.


A_S
C + D
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SM
ST
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21

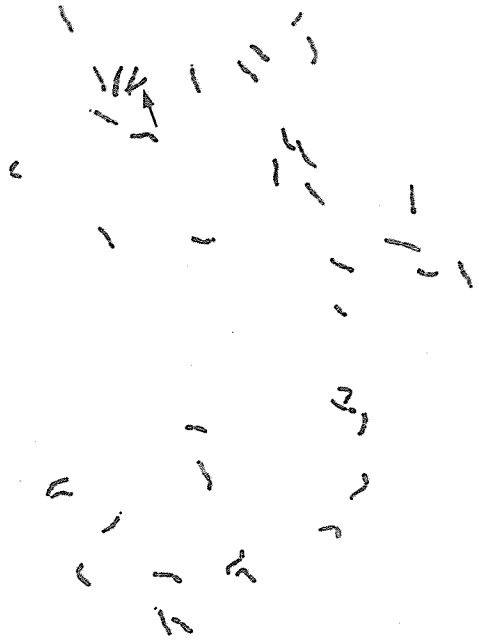
PLATE VIII. A mitotic metaphase of a mono-6 and meiotic configurations of F_1 hybrid between monosomic-14 and monosomic-6

Figure 1. Mitotic metaphase of monosomic-6 from which the karyotype is prepared (see arrow).

Figure 2. 19 bivalents and two univalents parallel side by side in diakinesis (see arrow).

Figure 3. The two univalents in a T-shaped configuration in diakinesis (see arrow).

Figure 4. The two univalents in a T-shaped configuration in late diakinesis (see arrow)



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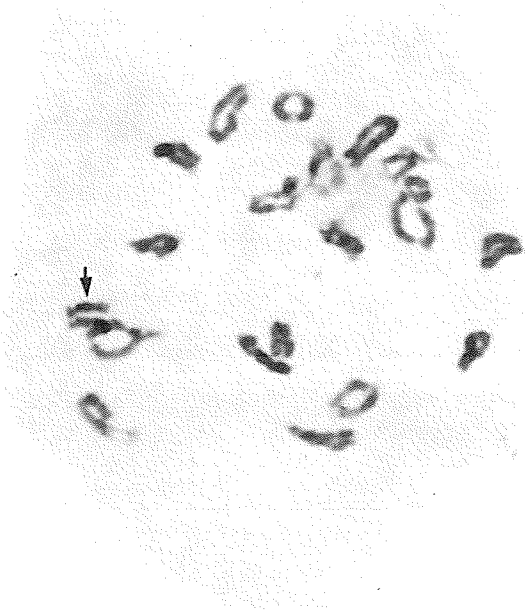

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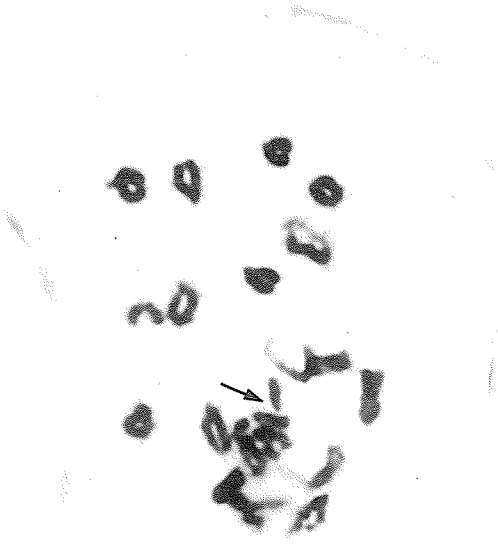

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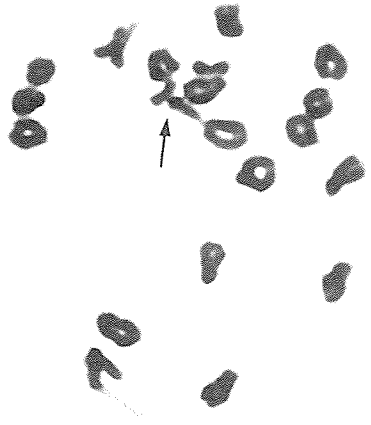
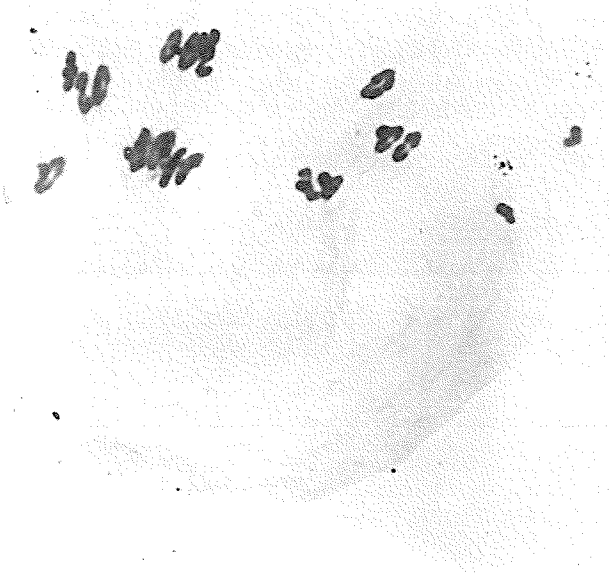


4

PLATE IX. Meiotic configurations of F_1 hybrid between monosomic-14 and -6.

Figure 1. 19 bivalents plus two univalents in metaphase stage.

Figure 2. The two univalents lagged in metaphase plate in anaphase stage.



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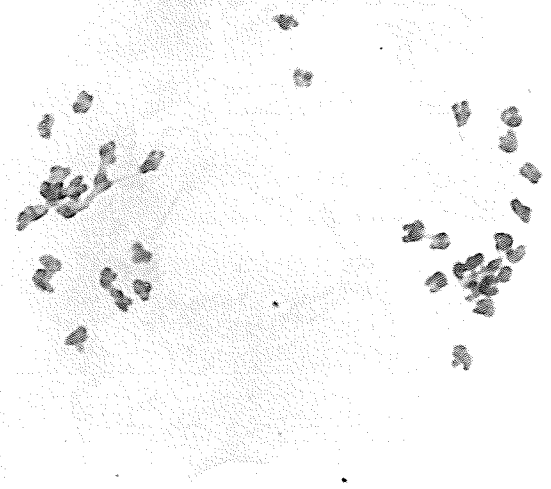


2

IX, Fig.1) and appeared to be distributed in the metaphase plate at anaphase I (Plate IX, Fig.2).

(c) Univalent Transmission

The theoretical univalent transmission rate calculated from the presence of micronuclei in tetrads is presented in Table 4.

TABLE 4. Number of micronuclei in tetrads of monosomic-6 plants and calculation of univalent transmission frequency.

Number of micronuclei per tetrad	Number of tetrads	Number of gametes	Total gametes with n=21	Percent gametes with n=21
0	56	224	112	50
1	183	732	183	25
2	475	1,900	0	-
3	0	0	0	-
	714	2,856	295	10.3%

The low percentage of 21-chromosome gametes (Table 4) makes it difficult to explain the occurrence of a high frequency of monosomics and low frequency of nullisomics from the selfed monosomic plants as shown in Table 5. A certation effect favoring the 21-chromosome gametes through the male might account for part of this discrepancy and zygotic lethality of nullisomics for the rest.

TABLE 5. Frequency of 42-, 41-, 40-chromosome oat seedlings from a selfed monosomic-6.

Chromosome number	Number of seedlings	Percentage of total
42	6	4.91%
41	104	85.25%
40	12	9.84%
	122	100.00%

R 364-535

(a) Morphology

Two nullisomic plants derived from the selfed monosomics were screened out for the purpose of morphological examination. One died after 3 weeks, the other one was obviously dwarf, attaining only half the height of the normal disomics (Plate X, Fig.1) with leaves about half the width of the normal ones. The plant grown in the greenhouse was quite healthy, producing a good number of tillers. Panicles were smaller and shorter with only a few spikelets on each. No seed was obtained from this plant. This nullisomic plant also exhibited the kinky neck in every tiller very similar to that of Mono-20 (Plate X, Fig.2); in addition all the spikelets appeared to be compact and upright.

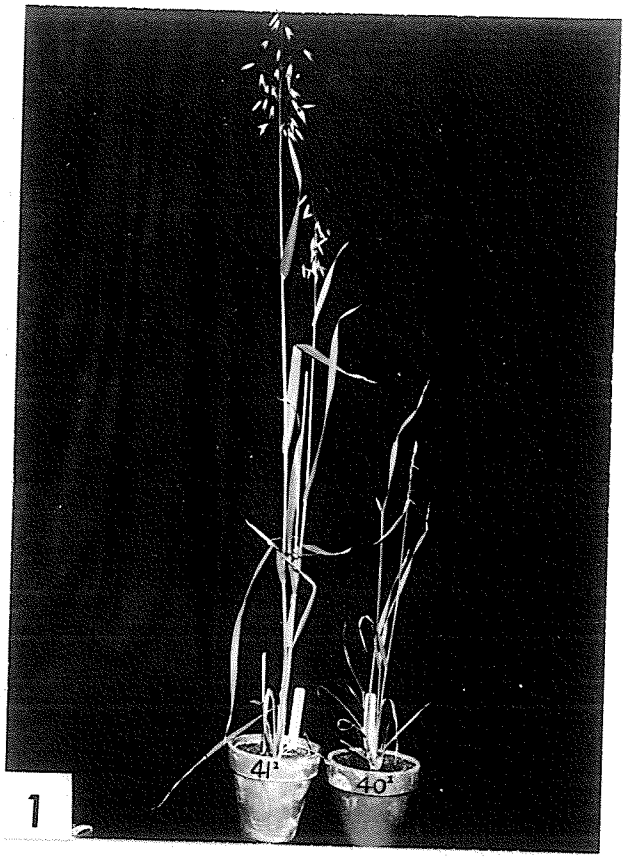
PLATE X. Plant morphology and mitotic metaphases of line R 364-535.

Figure 1. Comparison in height of a monosomic plant and nullisomic plant.

Figure 2. Kinky necks on 40-chromosome and 41-chromosome plants.

Figure 3. Mitotic metaphase of a monosomic-18 (see arrow).

Figure 4. Mitotic metaphase of a monotelosomic for the short arm of chromosome-18 (see arrow).



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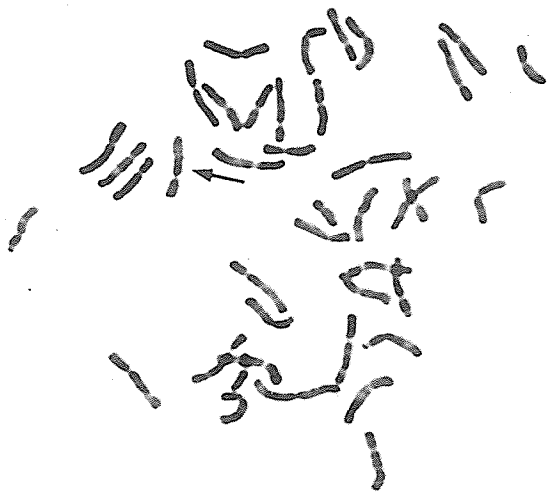

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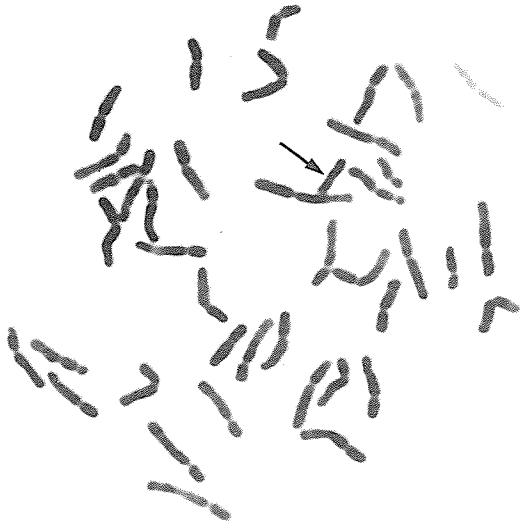


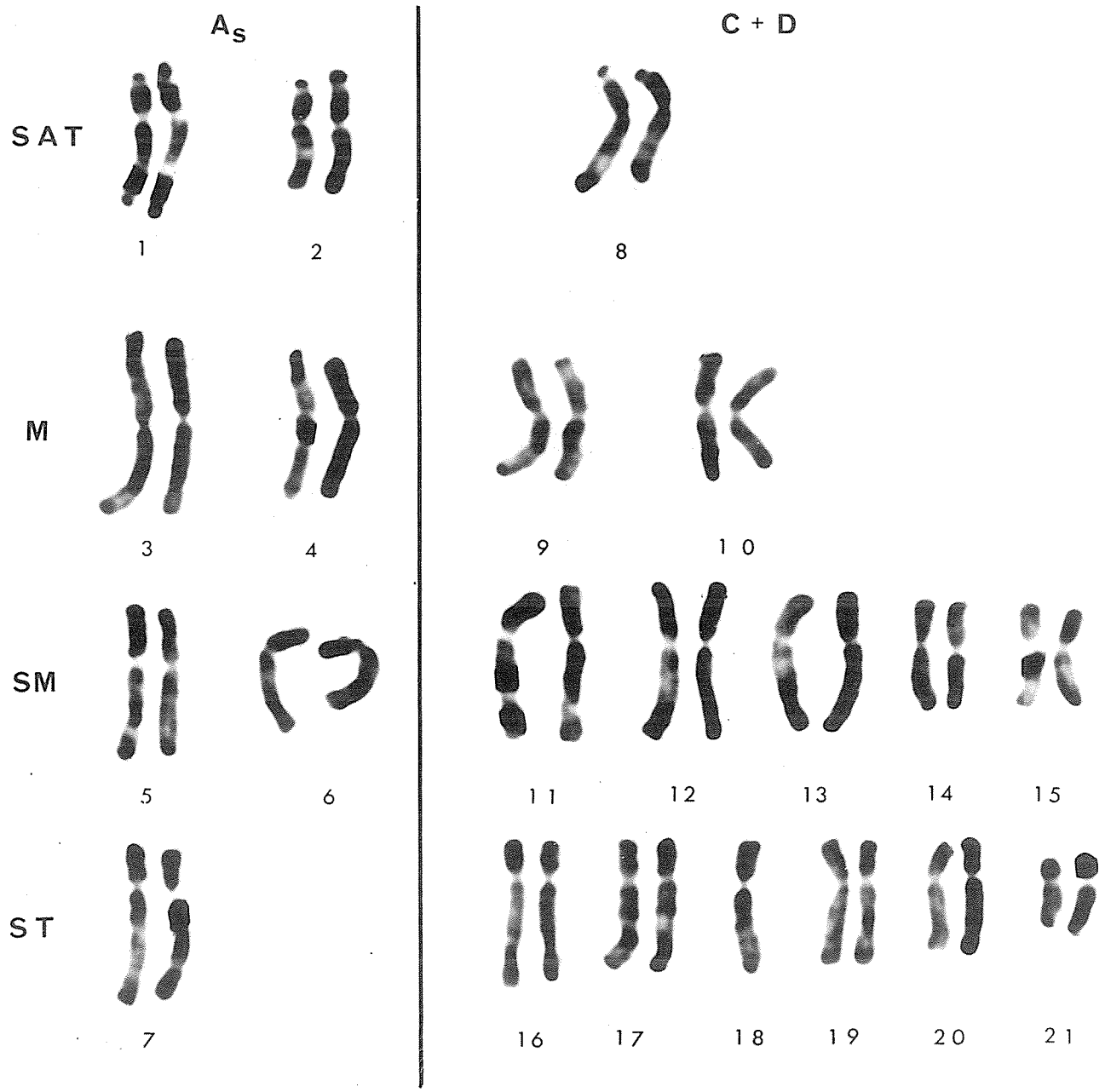
4

(b) Karyotype

It was quite difficult to determine which chromosome was involved in the deficiency. A preliminary examination of well-spread mitotic figures placed the missing chromosome in the group, 17, 18 and 19. Several good enlarged photographs showed a difference among the three chromosomes (Plate X, Fig.3, & Plate XI). In some of the photographs, chromosome-18 was similar to chromosome-17 in length but much longer than chromosome-19, in other cases, chromosome-18 was similar to chromosome-19, but much shorter than chromosome-17. Thus, from a study of 15 enlarged photographs, it was concluded that the critical chromosome was number 18.

The frequent occurrence of monotelosomics for the long arm (16.7%) in the progeny of selfed monosomics indicated that the univalent had a tendency to misdivide in meiosis (Plate X, Fig.4). The short arm apparently is easily lost since it was observed only once in the 180 cells examined. Both the monosomics and monotelosomics for the long arm were normal in morphology, suggesting that genes for the normal plant growth and normal neck-shape are probably located on the long arm of the chromosome. The kinky neck phenotype was first observed by Gauthier and McGinnis (5) on a spontaneous monosomic plant in the variety Garry and associated with chromosome-20, the long arm of which carries a gene for normal neck development. Later, Hacker and

PLATE XI. Karyotype of monosomic-18.


A_S
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Riley (7) observed another monosomic line in the variety Sun II exhibiting the kinky neck. In this monosomic, abaxial curling of leaves was also observed leading the authors to conclude that the critical chromosome resulted from a translocation between chromosome-14 and -20.

Since common oats is comprised of three genomes, the kinky neck phenotype could be conditioned by three pairs of genes and the results of this study support such a hypothesis.

(c) Univalent Transmission

The theoretical frequency of 21-chromosome gametes in the male was calculated from micronuclei counts in the tetrads as shown in Table 6.

TABLE 6. Number of micronuclei in tetrads of monosomic-18 plants and calculation of univalent transmission frequency.

Number of micronuclei per tetrad	Number of tetrads	Number of gametes	Total gametes with n=21	Percent gametes with n=21
0	206	824	412	50
1	333	1,332	333	25
2	176	704	0	-
3	10	40	0	-
	725	2,900	745	25.7%

A very high frequency of monosomics was present in the progeny of selfed monosomic plants (Table 7).

TABLE 7. Frequency of 42-, 41-, 40-chromosome oat seedlings from a selfed monosomic-18.

Chromosome number	Number of seedlings	Percentage of total
42	6	3.23%
41	142	78.88%
40+telo	30	16.67%
40	2	1.12%
	180	100.00%

A very effective certation favored the 21-chromosome gametes and a zygotic lethality of nullisomics appears to have occurred.

R 355-508

(a) Morphology

The nullisomic plants were very weak, had leaves 1/3 the width of the normal, and were characterized by their extremely slow growth as compared with the monosomic plants. A comparison of monosomics and nullisomics in one-month and two-month stages is shown in Plate XII, Figs 1 & 2. After two and a half months, the panicles of

nullisomics emerged.

(b) Karyotype

From the analysis of 11 mitotic figures of different monosomic plants, the missing chromosome was determined to be the second one in the median group, namely chromosome-4 (Plate XII, Fig.3 & Plate XIII). A configuration of metaphase I of a monosomic plant is also shown in Plate XII, Fig. 4.

(c) Univalent Transmission

The results of micronuclei counts in the PMCs at the tetrad stage are presented in Table 8 and the distribution of aneuploids and disomics in the progeny of selfed monosomics in Table 9.

Since the nullisomics were always found to be from the late-germinating seeds (three to four days later); one might suspect most of the ungerminated seeds were nullisomics which would raise the frequency of nullisomics in the progeny of selfed monosomics higher than the observed.

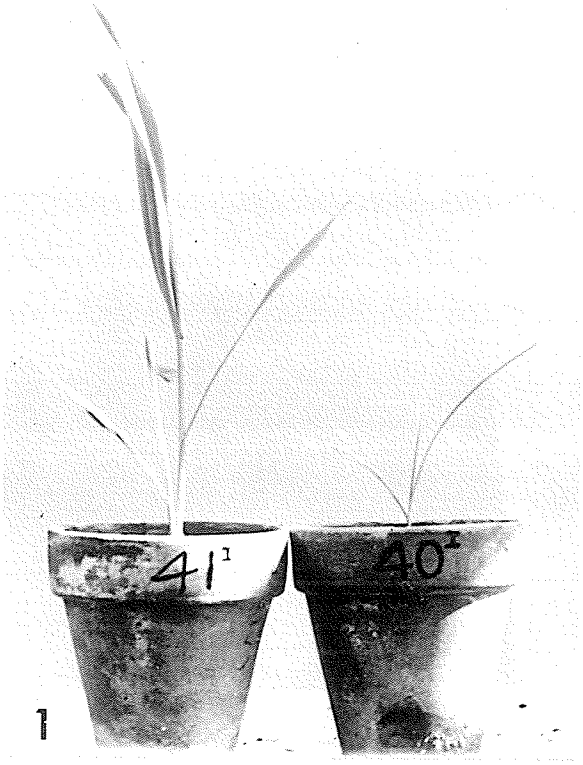
PLATE XII. Plant morphology and metaphase of monosomic-4.

Figure 1. Comparison in height of a monosomic and nullisomic plant at a one-month seedling stage.

Figure 2. Comparison in height of a monosomic and nullisomic plant at a two-month stage.

Figure 3. Mitotic metaphase of a monosomic-4 (see arrow).

Figure 4. Meiotic metaphase of a monosomic-4 (see arrow).



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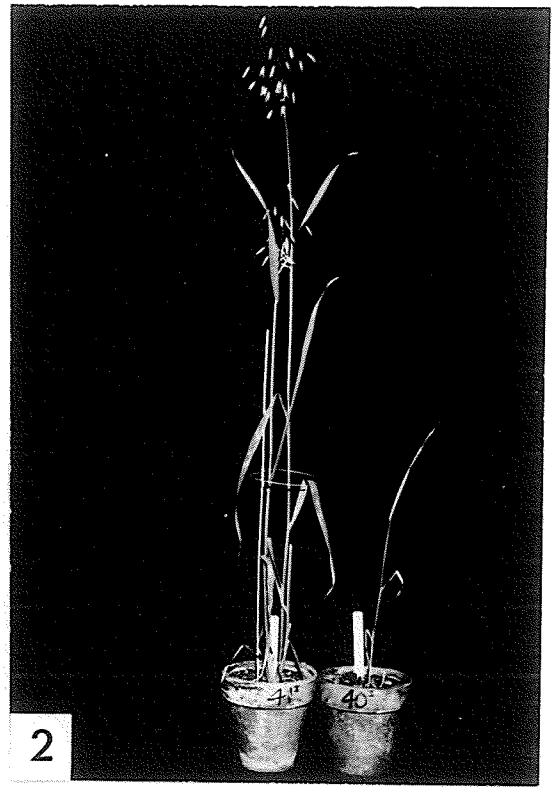

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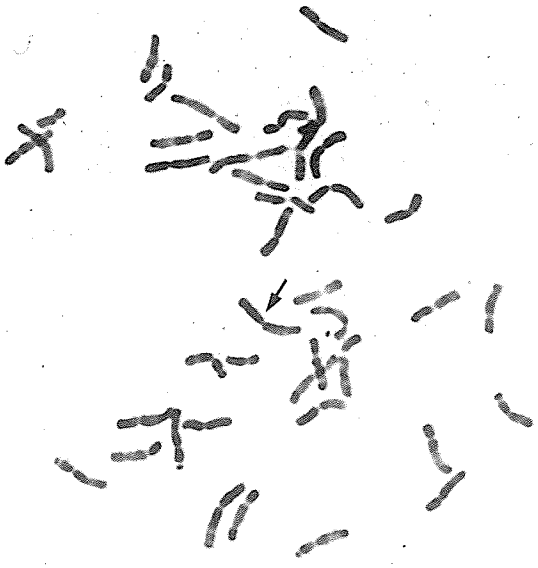

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PLATE XIII. Karyotype of monosomic-4.

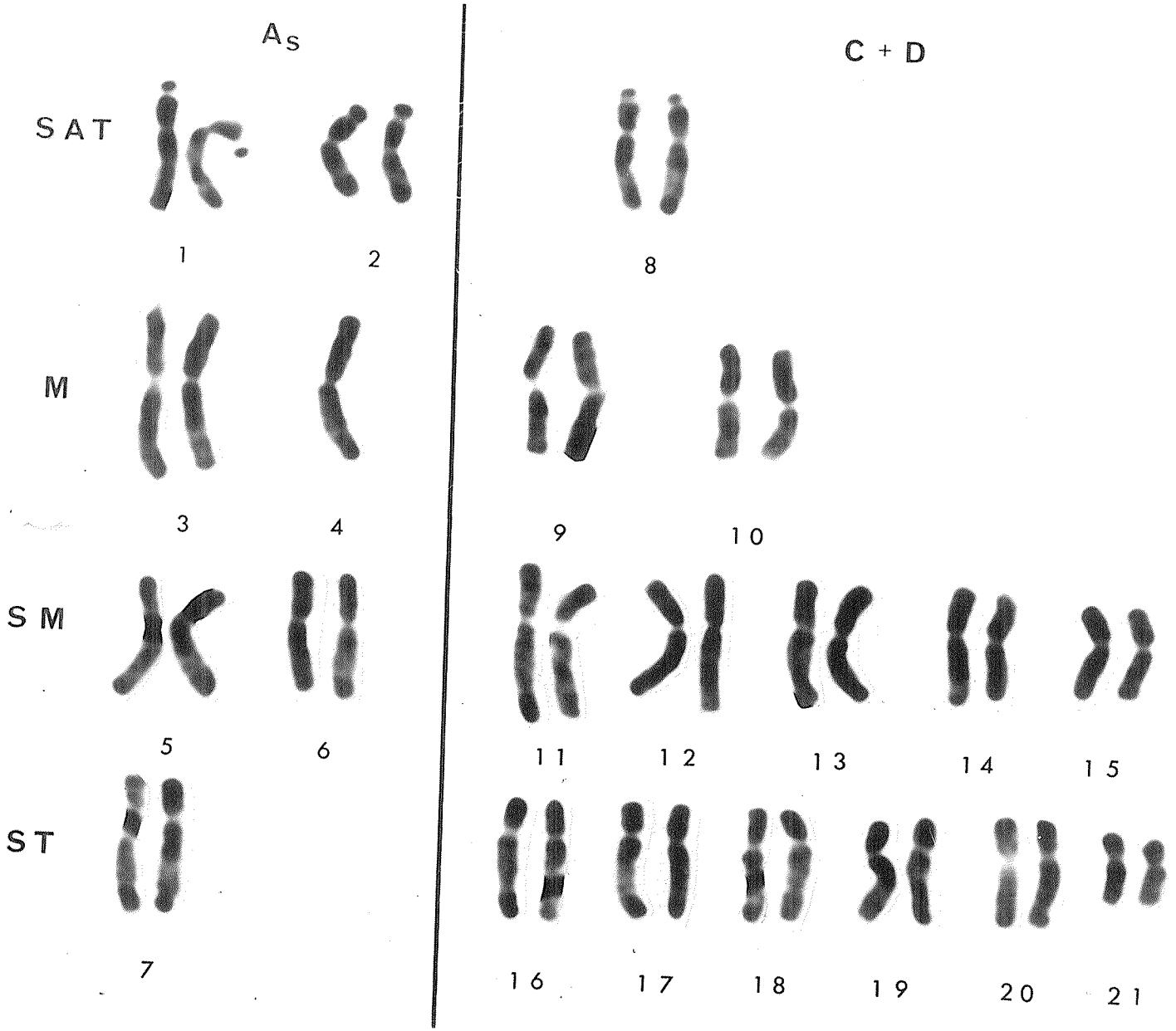
A S
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TABLE 8. Number of micronuclei in tetrads of monosomic-4 plants and calculation of univalent transmission frequency.

Number of micronuclei per tetrad	Number of tetrads	Number of gametes	Gametes with n=21	Percent gametes with n=21
0	24	96	48	50
1	144	576	144	25
2	500	2,000	0	-
3	80	320	0	-
4	24	96	0	-
	772	3,088	192	6.23%

TABLE 9. The distribution of aneuploids and disomics in the progeny of selfed monosomics.

Chromosome number	Number of seedlings	Percentage of total
42	1	1.7%
41	13	22.8%
40	43	75.5%
	57	100.0%

R 222-413, R 263-477, 21-139-156

All three lines were monosomic for chromosome 15 as determined by critical karyotype analysis. In addition, R 222-413 was crossed with a known line of monosomic-15 and as expected 20 bivalents were formed regularly at metaphase I in the F_1 hybrids.

R 267-492

This line was found to be monosomic for chromosome 21, the shortest of the complement.

CONCLUSIONS

It seems probable that because of the considerable variation in chromosome size and position of the centromere in common oats, one can classify the monosomics into four major groups and separate them further into individual lines on the basis of the idiogram proposed by Rajhathy (22). However, the differences among intra-group chromosomes, especially submedian and subterminal groups, are not always so obvious that from a single enlarged photograph it is possible to tell them apart. As a consequence, many well-prepared enlarged photographs must be used which collectively permit a correct identification. The combination of careful examination of idiograms made from well-spread mitotic figures and intercrossing between lines deficient for chromosomes that are morphologically similar greatly reduces the probability of an incorrect identification. Any monosomic line associated with a distinct gene marker is also very useful in sorting out monosomics belonging to the same chromosome group. In the present study, for example, monosomic-6 and monosomic-14 were not easily distinguished in photographs but the distinct gene marker, white striping of the leaves of mono-14 only, left little doubt as to their being different.

Unfortunately, a gene marker is not necessarily specific for a monosomic line. For instance, chromosomes-15

and -21 are known to carry genes for chlorophyll production and could be homoeologous. They are also the shortest members of the karyotype. The kinky neck phenotype was not specific to only mono-20 but also was observed in mono-18 which is similar morphologically to mono-20. Thus gene markers alone cannot sort out various monosomics. All evidence, morphological, karyotypic, and the cytology of F_1 hybrids of similar lines should be combined in identifying new monosomics.

Similar univalent transmission rates in both sexes was not observed in all the four monosomics studied. The male univalent transmission rate was calculated to be 16.3% for mono-14, 10.3% for mono-6, 25.7% for mono-18, and 6.23% for mono-4, but the frequency of 40, 41, 42-chromosome plants in monosomic progeny, indicated that relatively few deficient male gametes were successful in fertilization and/or nullisomic zygotes were generally lethal.

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