*Turkish Journal of Field Crops, 2011, 16(1): 23-28*

# DEVELOPMENT OF AUTOTETRAPLOID PERENNIAL RYE (*Secale montanum Guss.*) AND SELECTION FOR SEED SET

İlknur AKGÜN[1] Metin TOSUN[2] Kamil HALİLOĞLU[3*] Murat AYDIN[2]

[1] *Süleyman Demirel University, Faculty of Agriculture, Department of Field Crops, Turkey*
[2] *Atatürk University, Faculty of Agriculture, Department of Field Crops, Turkey*
[3]*Atatürk University, Faculty of Agriculture, Department of Agricultural Biotechnology, Turkey*
**Corresponding author: kamilh@atauni.edu.tr*

Received: 13.12.2010

**ABSTRACT**

**Different concentrations of colchicine solution (0.1% and 0.2%) and temperatures (24 and 27°C) were applied to wild diploid rye and autotetraploid forms were produced. Five hundred seedlings were placed into Petri dishes for each treatment (total 2000). Cytological research was carried out on root tips of plants in $C_0$ and $C_1$ generations. Live seedling rate and tetraploid plant rate were changed by 6.4 – 37.4 % and 8.75 – 37.04%, respectively based on treatments. Plants forming the $C_1$ generation were determined as 68.12% tetraploid, 14.71% diploid, 16.89% aneuploid (13.35% hypoploid, 3.57% hyperploid) and 0.27% triploid. Chromosome numbers of aneuploid plants ranged between 2n=19 - 29 and it was determined that plants having a chromosome number of 2n=27 were the most common. Other aneuploid plants (2n=19 - 26) were rare. Prior to chromosome counting in the $C_1$ generation, the seed lots were divided into five seed size groups (9-11 g, 11-13 g, 13-19 g, 15-17g, 17-19 g). Chromosome numbers were determined by a modification of the conventional root tip squash technique. Chromosome numbers varied with seed size. The rate of tetraploids, diploids and aneuploids were varied between 17.86-87.25%, 2.04-42.86%, 10.71- 39.29 %, respectively. As the size of seed was increased, the rate of diploid and aneuploids decreased. Seed set rate, spike length and the number of spikelets in plants belonging to different generations ($C_1$, $C_2$, $C_3$ and $C_4$) were investigated. Seed set rate was increased from 30.51% to 61.45% with the selection of plants passing over general average in respect to seed set in each generation, however, it was determined that spike length and number of spikelets decreased. The differences were generally highly significant.**

**Key Words: *Secale montanum Guss,* colchicine, autotetraploid, aneuploid, seed set, selection.**

## INTRODUCTION

The wild perennial rye (*Secale montanum* Guss.) is believed to be the ancestral species of the cultivated rye, *S. cereale* L. (Stutz, 1972). *Secale montanum* is commonly cultivated in Mediterranean countries, Anatolia, Southern Caucasus and Iran. There are many perennial forms of rye in Turkey and *Secale montanum* Guss *var. anatolicum* Boiss. and Secale *montanum* Guss *var. vavilovi* Grossh. are important varieties among them (Kün, 1988). Compared to cultivated rye, this perennial species has a large stature, frost resistance, strong tillering ability, a slightly more prostrate habit and tolerance to poor soil and poor water supply. Their seeds are easily germinated (Reimann-Philipp 1986; Richard & Wang 1987).

Perennial diploid species might be favorable breeding material for induced autoploidy breeding. *Secale montanum* Guss has a low chromosome number (2n=14) and is naturally cross-pollinated (Akgün et al. 1996). These characters are ideal for autoploidy. Generally, autotetraploid plants are more vigorous than their diploid parents. In autotetraploids, stems are typically thicker and stouter; leaves thicker, larger, and deeper green in color; roots, flowers and seeds are larger. On the other hand, auotopolyloid plants in general have a reduced number of seeds per plant. (Poehlman, 1995). This reduction is caused partly by a reduction in the number of flowers per plant, and partly by a reduction in the percentage of fertile flowers (Aasveit 1968). Meiotic irregularities also appear to be responsible for low fertility through the production of genetically imbalanced products following aberrant chromosome pairing and /disjunction at meiosis. The effects of doubling the chromosome number have been studied in many autotetraploid crops (Hazerika and Ree,s 1967; Hossain and Moore, 1975; Narasinga and Pantulu, 1982; Klinga, 1986 a b).

Autotetraploid populations are also generally characterized by the occurrence of aneuploids. The frequency of aneuploids varies among species and among populations within species. The average seed set of a tetraploid rye population (*S. cereale L.*) is correlated with the frequency of aneuploids present in the population (Hossain, 1978).

The aims of this research were (1) to produce perennial autotetraploid rye and determine the effects of different colchicine concentrations and temperature durations on chromosome duplication, (2) to determine the rates of

aneuploids and euploids in $C_0$ and $C_1$ generations, (3) to study the correlation between seed size and aneuploid frequency in the $C_1$ generation and (4) to increase seed set.

In order to increase seed set rate, which is important problem in tetraploid ryes, the selection of plants having greater rates than general average was applied in generations of $C_2$, $C_3$ ve $C_4$ and values of genetic advancements were determined in this study.

## MATERIALS AND METHODS

Seeds of perennial rye *(Secale montanum* Guss*)* were collected from the plants naturally grown int Erzurum, Turkey. Plants were selected according to morphological characteristics such as morphological vigor and high tillering capacity.

### *Production of autotetraploids*

Two different colchicine solutions (0.1% and 0.2%) and two temperature (24 and 27$^o$C) were employed in this study. Seeds of diploid plants were germinated under greenhouse conditions. Two thousand seedlings with root lengths of 2 – 3 mm were selected. Five hundred seedlings were plated into Petri dishes for each application and colchicine solutions of 0.1% or 0.2% were added until it fully covered the germinated seeds. Each sample was kept for 3 hours at 24 and 27$^o$C. At the end of this period, seedlings were washed thoroughly with tap water and separated into different boxes based on treatments. In addition, control plants (total 50 seedlings) were planted in one row in each wooden box. Seedlings that reached the soil surface were assessed in comparisons with controls and selected ones were transplanted into pots and their chromosomes were counted under microscope.

### *Cytological Investigations*

Cytological research was carried out on root tips of plants in $C_0$ and $C_1$ generations. The percentages of euploid and aneuploid plants were determined in $C_0$ and $C_1$ generations. Chromosome numbers were determined by a modification of the conventional root tip squash method, the root tips were pretreated in a saturated solution of α-monobromnaphthalene for 3 hours at room temperature. After this, the root tips were fixed in 3:1 solution of ethyl alcohol and acetic acid (Farmer's fixative) for 2 hours. This was followed by placing the samples in the tubes containing 1N HCl at 60 °C for 20 minutes to hydrolyzed the tissue. After staining by Feulgen solution, root tips were squashed in 45% acetic acid and examined under the microscope. To determine the chromosome number, it was usually necessary to examine at least five good metaphase plates from each plant.

1000 – kernel weights were determined in seeds of tetraploid plants in the $C_1$ generation. Five groups (9-11 g, 11-13 g, 13-15 g, 15-17 g, 17-19 g) were established based on 1000-kernel weight. The $C_1$ plants were grown in plastic pots containing a mixture of soil: farm manure: sand (1:1:1) under greenhouse condition.

### *Field Research*

After about 2 months (4 May 1995), 1060 $C_1$ plants were transplanted into plots that spaced 50 x 50 cm on and between rows in the field. Each plant was numbered. Assessments of the $C_1$ generation were made by average of years 1996 and 1997.

Eight hundred and fifty two plants with greater than the general mean based on seed set rate formed the $C_2$ generation. Each plant was planted randomly into a 2 m wide row in 1997 fall. The distance between rows was 50 cm. The total 852 plant rows were formed. Assessments belonging to the $C_2$ generation were made based on the average results of years 1998 and 1999. Five hundred and eight plants with values greater than the general mean in respect to seed set rate formed the $C_3$ generation, and 247 plants formed the $C_4$ generation. The same experimental design as $C_2$ was employed for $C_3$ and $C_4$. Assessments of the $C_3$ generation were made based on the average results of years 2000 and 2001, assessments of the $C_4$ generation were made based on the average results of years 2002 and 2003.

Analysis of variance and Duncan's Multiple Range Test were used to compare examined characters in the different generations. All computation were done by using SAS® 9.0 software for Windows®.

The following parameters were investigated in plants belonging to $C_1$, $C_2$, $C_3$ and $C_4$ generations.

*Spike length:* Spike length was found by measuring the length between the lowest node and the toppest node (Akgün 1994). Five spikes from each plant in the $C_1$ generation and 20 spikes from each line of the other generations were measured and the mean was taken.

*Spikelet number:* Spikelet number was determined as an average spikelet number per spike by counting all spikelets on the same spike. The top and bottom spikelets in all cases were excluded (Aastveit, 1968) .

*Seed set ratio:* Seed set was determined by counting the number of flowers and grains on the spikes (Aastveit 1968). Spikelets at the top and bottom of each spike were not taken into consideration (Fedak, 1975).

*Genetic advance:* Genetic advance for seed set was calculated as follows (Wricke, 1972).

Genetic Advance = $h^2$ x Sf x i

$h^2$ = realized heritability, Sf = standard deviation, i = selection intensity

$h^2$=R/S

R = $\overline{X}_{offspring} - \overline{X}_{population}$ (the mean of the offspring of the selected plants - mean of the base population).

S= $\overline{X}_{selected} - \overline{X}_{population}$ (the mean of the selected plants from the population - mean of the base population).

Values of selection intensity (*i*) in respect to population size was described by Becker (1984).

## RESULTS

*Cytological Results:* Seedlings treated with different temperatures and colchicine applications were transferred to growth box and compared with controls. The appearance of unaffected seedlings from colchicine treatment was similar to the control and showed characteristics of fast development. The coleoptile was thicker in affected seedlings and the first leaf was typically bigger than normal and darker green in color. Table 1 summarizes survival rates of the seedlings after one month.

**Table 1.** The Survival Number and Rates of Perennial Rye Seedlings at Different Temperatures and Colchicine Treatments.

| Colchicine Concentrations (%) | Temperatures (°C) | Number of treated seeds | Survival number and rates | | | |
|---|---|---|---|---|---|---|
| | | | Colchicine Treatment | | Control | |
| | | | Number | % | Number | % |
| 0.1 | 24 | 500 | 187 | 37.4 | 43 | 86 |
| | 27 | 500 | 165 | 33.0 | 46 | 92 |
| 0.2 | 24 | 500 | 56 | 11.2 | 47 | 94 |
| | 27 | 500 | 32 | 6.4 | 45 | 90 |
| Total/Avarage | | 2000 | 440 | 22.0 | 181 | 90.5 |

The highest survival rate was determined at 24°C and 0.1% treatment (37.4%) and the lowest rate was determined at 27°C and 0.2% treatment. The average survival rate in untreated seedlings was 90.5%. Seedlings similar to control in appearance were removed from growth boxes and a total affected 207 plants were transferred to pots. Somatic chromosome numbers were determined in these plants (Table 2).

**Table 2.** The somatic chromosome numbers of investigated plants at different temperatures and Colchicine Treatments in Perennial Rye.

| Colchicine Con. (%) | Temperatures (°C) | Numbers of plants | 2n=28 | | 2n=14 | | Mixoploid | |
|---|---|---|---|---|---|---|---|---|
| | | | n | % | n | % | n | % |
| 0.1 | 24 | 80 | 7 | 8.75 | 39 | 48.75 | 34 | 42.5 |
| | 27 | 83 | 13 | 15.66 | 21 | 25.30 | 49 | 59.04 |
| 0.2 | 24 | 27 | 10 | 37.04 | 6 | 22.22 | 11 | 40.74 |
| | 27 | 17 | 5 | 29.41 | 4 | 23.53 | 6 | 35.29 |
| Total | | 207 | 35 | 16.91 | 70 | 33.82 | 100 | 48.31 |

The highest tetraploid plant formation (37.04) was observed following 0.2% of colchicine treatment at 24°C. Based on number of treated plants (500 plants), however, the highest tetraploid plants percentage (2.6%) was determined at 27°C and %0.2 of colchicine treatment, whereas, the lowest tetraploid plant rate (%0.1) was observed at 27°C and %0.2 of colchicine treatment. The highest diploid plant formation was determined at 24°C and 0.1% of colchicine treatment, while mixoploids were observed at 27°C and %0.1 of colchicine treatment. The increase of colchicine concentration reduced the tetraploid and mixoploid plants rate (Table 2).

**Table 3.** Diploid, tetraploid and aneuploid plant rates of different seed weight groups in $C_1$ generation.

| | | Chromosome Number | | | | | | | | Diploid | Tetraploid | Aneuploid |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Weight of fraction (g) | N | **14** | **19** | **21** | **25** | **26** | **27** | **28** | **29** | (%) | (%) | (%) |
| 9.0-11.0 | 28 | 12 | - | - | 3 | - | 8 | 5 | - | 42.86 | 17.86 | 39.29 |
| 11.0-13.0 | 33 | 13 | - | - | - | 1 | 7 | 12 | - | 39.39 | 36.36 | 24.24 |
| 13.0-15.0 | 16 | 3 | 1 | - | - | - | 3 | 9 | - | 18.75 | 56.25 | 25.00 |
| 15.0-17.0 | 94 | 22 | - | 1 | - | 1 | 16 | 53 | 1 | 23.40 | 56.38 | 19.15 |
| 17.0-19.0 | 196 | 4 | - | - | - | - | 9 | 171 | 12 | 2.04 | 87.25 | 10.71 |
| Total | 367 | 54 | 1 | 1 | 3 | 2 | 43 | 250 | 13 | 14.71 | 68.12 | 16.89 |

In mixoploid ears, the distribution of diploid and tetraploid pollen mother cells showed marked variations. In some cases, whole flowers were diploid or tetraploid, a flower had two anthers which were diploid and one that was tetraploid, or there were anthers having a mosaic of diploid and tetraploid cells. Therefore, seeds of mixoploid plants were not used to form the $C_1$ generation. The mature ears of all of 35 tetraploid plants were harvested. 1000-kernel weight of these plants ranged between 9.00 – 19.00 g and 5 different groups were established (9-11 g, 11-13 g, 13-15 g, 15-17 g, 17-19 g). The seeds from each group were sown separately in small pots. A total 367 seedlings were studied cytologically by means of squash preparation of root tips. The results are given in Table 3.

It was determined that plants in the $C_1$ generation were generally 14.71% diploid, 68.12% tetraploid, 16.89% anueuploid and 1.06% triploid. Chromosome numbers of aneuploid plants ranged between 2n=19-29 and the highest chromosome number was 2n=29, followed by 2n=27 chromosome number plants. When 1000-kernel weight was higher than 15 gr, tetraploid plant rate increased enormously.

Also, as 1000-kernel weight increased, the rate of hyperploids increased.

After cytological investigation, total 1060 plants were transferred to the field. After chromosome counting, plants were labeled and seed set rate was assessed separately. Most aneuploid perennial rye individuals show a low degree of fertility and also poor vegetative development. The seed set rate of hypoploid (2n=19-27) plants ranged 11.13 – 21.79%, whilst hyperploids (2n=29) varied between 21.37 – 38.87%.

In order to reduce the rate of diploid and aneuploids in other generations, plants morphologically similar to diploids were removed from the field before flowering (athesis) and plants with a seed set rate above the average were selected and formed to form the 3rd generation.

*2. Selection for improving fertility in autotetraploid perennial rye*

*a. Spike length:* Between the $C_1$ and $C_3$ generations, average spike length reduced with the advancement of each generation (P= 0.003). However, there were no significant differences between spike lengths of plants of the $C_3$ and $C_4$ generation. The longest spikes were determined in $C_1$ generation. The reason might be that plants of the $C_1$ generation were widely spaced at planting (50 x 50 cm). Seeds from spikes in $C_2$, $C_3$ and $C_4$ generations were sown in row with narrow spacing compared to plants in the $C_1$ generation (Table 4).

*b. Spikelet number***:** Average spikelet number changed significantly between generations (P=0.001). It was 43.61 in the $C_1$ generation and 35.40 in the $C_4$ generation. The lowest spikelet number was observed in $C_3$ plants (Table 4).

*c. Seed Set:* Seed set rate was significantly increased in subsequent generations (P=0.002). The highest seed set rate was determined in the $C_4$ generation (61.45%) followed by the $C_3$ (48.71%),$C_2$ (40.0%) and $C_1$ (30.51%) generations.

**Table 4.** Spike length, spikelet number and seed set rate of perennial tetraploid rye (*Secale montanum* Guss) of different generations.

| Characters | Generation | N | Min. | Max. | Mean | Std Deviation | CV (%) |
|---|---|---|---|---|---|---|---|
| **Spike length (cm)** | $C_1$ | 1060 | 8.83 | 17.23 | 14.04 **a**[1/] | 1.272 | 9.06 |
| | $C_2$ | 852 | 6.00 | 18.78 | 12.63 **b** | 1.9825 | 15.70 |
| | $C_3$ | 508 | 8.75 | 15.25 | 11.85 **c** | 1.1298 | 9.53 |
| | $C_4$ | 247 | 9.75 | 14.65 | 12.07 **c** | 0.9820 | 8.14 |
| **Spikelet number** | $C_1$ | 1060 | 29.50 | 55.50 | 43.61 **a** | 3.843 | 8.81 |
| | $C_2$ | 852 | 17.00 | 57.00 | 40.21 **b** | 6.1210 | 15.22 |
| | $C_3$ | 508 | 16.60 | 44.30 | 30.37 **d** | 5.6805 | 18.70 |
| | $C_4$ | 247 | 19.50 | 47.50 | 35.40 **c** | 4.2722 | 12.07 |
| **Seed Set (%)** | $C_1$ | 1060 | 14.84 | 58.69 | 30.51 **d** | 5.582 | 18.30 |
| | $C_2$ | 852 | 14.84 | 59.00 | 40.50 **c** | 7.6114 | 18.79 |
| | $C_3$ | 508 | 30.34 | 76.00 | 48.71 **b** | 10.0595 | 20.65 |
| | $C_4$ | 247 | 35.64 | 89.25 | 61.45 **a** | 7.3446 | 11.95 |

[1/] Values with same letter in a character are not significantly different according to Duncan test at P<0.01

*d. Genetic Advance:* Using selection based on seed set in different generations, 852 out of 1060 (80%) were selected in the $C_2$ generation, 508 out of 852 (60%) in the $C_3$ generation and 247 out of 508 (49%) in the $C_4$ generation. Thus, the genetic advancement value (*i*) with respect to generations was calculated based on the percentage of selection intensity. Genetic advancement values were determined as 1.829 in $C_2$, 5.891 in $C_3$, and 11.856 in $C_4$ through the selection of plants above the overall mean. An approximately 6% increment in the $C_2$ generation [(1.829/30.51).100 = %5.99], 15% increment in the $C_3$ generation [(5.891/40.4974).100 =%14.55], and 24% increment in the $C_4$ generation [(11.856/48.71).100 =%24.34] were observed in respect to general mean of previous generation.

## DISCUSSION

Seedlings of perennial rye were increasingly damaged from colchicine treatments depending on the increase of temperature. Therefore, colchicine affected seedling rate was found 7.8% and 4.2% at 24 and 27°C in treatment of 0.1% colchicine concentration, respectively. On the other hand, in 0.2% colchicine concentration colchicine affected seedling rate were 1.2% and 0.8%, respectively. According to studies in different plant species, survived plant rate were changed based on density of colchicine, temperature and species (Hassan et al. 1989). Increment in temperature increased the colchicine affected seedling rate. Chromosome counting was done in all plants which were appeared to be affected. Existence of diploid plants in seedling was interesting. This could be explained either by existence of mutation or chimera structure is transformed into diploid form by totally extinction of polyploid cells during development which were few at the beginning.

Generally, when colchicine concentration is lower, mixoploid plant rate was higher. However, mixoploid plant rate also increased in low concentration of colchicine when temperature was high. On the other hand, in the high temperature, cell division has become low, thus few numbers of cells were formed, and then the most of the new formed cells were affected by colchicine. Number of divided cells in perennial rye increased at 24°C, but number of cells affected from colchicine was low. Whereas cell division was lower at high temperature, and the most of the new formed cell were affected by colchicine.

Tetraploid plant rate changed depending upon type of the

treatment and the highest tetraploid plant rate was obtained at 24°C from 0.2% colchicine application. However, when it was assessed based on number of seedlings at the beginning of the application (500 seedlings), the highest tetraploid plant rate was observed at 27°C in 0.1% colchcine concentration (2.6%). A number of researchers (Sağsöz 1982; Hague & Jones 1987; Luclett 1989) reported that number of tetraploid plants was changed based on species and application according to studies carried out different species.

Aneuploid plant rate changed between 10.47-39.29% based on 1000-kernel weight. The increment of 1000-kernel weight increased the tetraploid plant rate, while decreased diploid and aneuploid plant rate. When 1000-kernel weight was higher than 17 g, most of plants (87.25%) were determined as tetraploid.

Close relationship was determined between 1000-kernel weight and aneuploid plant rate in this study. Similar investigations in other crops commonly display a close relationship between seed size and aneuploid frequency; particularly the frequencies of hypoploids vary with seed size, since it seems that a loss of one or two chromosomes has more severe effect on the seed size and quality than when the seed is hyperploid. In addition, it was determined that seed weight could be a selection criterion for removing aneuploids (Weirmarch, 1975; Gradskova and Panina, 1981; Klinga, 1986 a b; Tosun et al. 2003).

Most of the aneuploid plants had 2n=27 and 29 chromosomes, other aneuploid plants having 2n=19-26 chromosomes were rarely seen. As seed weight increased, frequency of hypoploid plants decreased. It was determined that survival rate of gametes having 1 chromosome missing in perennial rye plants is higher than other hypoploid gametes. Hossain (1978) reported that hyperaneuploid gametes were less functional than hypoaneuploid gametes. The tolerance of aberrant gametes and zygotes varies among species and in some crops, the viability is generally high. Klinga (1986 b) determined the aneuploid frequency to about 50% in two different autotetraploid populations of *Lolium multiflorum*. On the other hand Ahloowalia (1971) determined the aneuploid frequency in nine populations of *Lolium perenne* and found frequencies ranging from 6 to 50%.

In this study, it was determined that frequency of triploid plants was 1.06%. Jain (1960) stated that seeds in triploid rye did not develop well due to mitotic instability in endosperm tissue, even resulted in death of embryos. On the other hand, as seed size increase, frequency of diploid plants decreases. Diploids can be formed either by transforming tendency of tetraploid plants to diploid (diploidization) or partogenetic development of reduced eggs in tetraploids.

Autotetraploid populations are generally characterized by a decreased fertility, and by an occurrence of aneuploids to various degrees. Aneuploidy frequency varies depending upon species, generations or applied selection method. Several researchers (Ahloowalia, 1971; Simonsen 1973; Sağsöz et al. 1998, 2002) reported that aneuploidy frequency was high in first generations, and then decreased in further generations. Cytological instabilities in beginning generations result in gametes having deficient number of chromosomes and increase frequency of aneuploids. In addition, this can reduce seed set rate. Usually, euploids are superior to aneuploids, hyperploids are superior to hypoploids, and the effects are stronger on the generative characters than on the vegetative ones. (Klinga 1986b). Hossain (1978) found that the frequency of aneuploids was significantly reduced in the rye population selected for high seed set with regular meiosis. It can be said that selection of over passing general average in respect to seed set rate is effective for protection cytological stability. Seed set rate increased up to 61.45% by selection, which was indeed 30.51% in $C_1$ generation. Similar results were also reported by Akgun and Tosun (2007). In addition, it was reported that fertility gets closed to diploids by selection through 7 generations in artificial tetraploid *Trifolium hibridum* (Elgersma, 1993). Beside this, Aastveit (1968) determined that seed set of autotetraploid rye is a continuously varying character which is under genetic control, but is also highly influenced by environmental factors. Because of low heritability the response to selection is relatively slow. Therefore, even we tried to reduce the effect of environment by assessments based on data of two years in our study, genetic advancement was found to be low.

Applied selection affected spike length and spikelet number in negative way. However, higher spike length and spikelet number in plants of $C_1$ generation compared to plants of other generations could be resulted from growing plants of $C_1$ generation in spaced row (50x50 cm). Because seeds were sown in row in other generations, soil space for plant utilization was low in $C_1$ generation. Elgersma (1990) stated that environmental factors along with genetic factors were effective for seed yield and yield components. On the other hand, spike length and spikelet number were higher in $C_4$ generation than $C_2$ and $C_3$ generation. This increase could be due to environment and/or genetic factors. It would be possible to determine whether this increase originated either from genetic structure or environment conditions when further generations were investigated.

## CONCLUSION

According to results obtained from this research, 0,2% colchicine treatment should be applied at 24°C temperature to produce autotetraploid perennial rye. Chromosome number at $C_1$ generation indicated variation in respect to seed size. While seed size got larger, rate of diploids and aneuploids decreased. Low seed set rate is the main problem in artificial autotetraploids. Seed set ratio increased from 30,51% at $C_1$ to 61,45% at $C_4$ as a result of selection based on higher seed set rate through four generations ($C_1$ – C4).

## LITERATURE CITED


Aastveit, K., 1968. Variation and selection for seed set in tetraploid rye. Hereditas, 60: 294 - 316.

Ahloowalia, B.S., 1971. Performance of diploid varieties and their tetraploid progenies in perennial ryegrass. Ir.J.Agric.Res. 10:333-340.

Akgün, İ., 1994. Autotetraploid Çok Yıllık Çavdar (*Secale montanum* Guss.)'ın $C_1$ Generasyonunda Bulunan Aneuploidlerin Seçilmesi ve Farklı Seviyelerde Uygulanan Azotun Diploid ve Eutetraploid Bitkilerin Bazı Sitolojik ve

Morfolojik Özellikleri Üzerine Etkisi. Atatürk Univ., (Ph. Dissertation), Erzurum, Türkiye.

Akgun, I., M. Tosun, 2007. Seed set and some cytological characters in different generations of autotetraploid perennial rye (*Secale montanum* Guss). New Zealand Journal of Agricultural Research 50: 339-346

Akgün, İ., S. Sağsöz, M. Tosun, 1996. Alternatif bir yem bitkisi çok yıllık çavdar. Tarım - Çevre İlişkileri Sempozyumu. "Doğal kaynakların Sürdürülebilir Kullanımı", 13-15 Mayıs 1996, Mersin, 909-918

Becker, W., 1984. Manual of Quantitative Genetics. Academic Enterprises, Pullman

Elgersma, A., 1990. Seed yield related to crop development and to yield components in nine cultivars of perennial ryegrass (*Lolium perenne* L.). Euphytica 49:141-154.

Elgersma, A., 1993. Effects of aneuploidy on seed production in autopolyploid forage crops. Procceeding of the XVII. Inter Grassland Cong. Feb. 8, 1993, New Zealand pp.1694-1695

Evans, G.M., M.M. Rahman, 1990. The basis of low grain yield and interfertility in autotetraploid barley (*Hordeum vulgare* L.). Heredity 64:305-313.

Fedak, G. 1975. Fertility and meiotic behaviour in tetraploid barley. Can. J. Genet. Cytol. 17:: 121-123.

Gradskova, L.A., E.B. Panina, 1981. A study of relationship between breeding and cytogenetic traits in tetraploid rye populations. Plant Breed. Abst. 52: No:2.

Hague, L.M., R.N. Jones, 1987. Cytogenetics of *Lolium perenne*. 4. Colchicine induced variation in diploids. Theo. Apl. Genet. 74: 233-241.

Hassan L, R.N. Jones, U.K. Posselt, 1989. A novel source of genetic variation in rye grasses *(Lolium multiflorum, L. perenne*). Heredity 63:339-342.

Hazarika, M.H., H. Rees, 1967. Genotypic control of chromosome behavior in rye. X. Chromosome pairing and fertility in autotetraploids. Heredity 22: 317-332.

Hossain, M.G., K. Moore, 1975. Selection in tetraploid rye. I. Effects of selection on the relationships between seed set, meiotic regularity and plant vigor. Hereditas 81:141-152.

Hossain, M.G., 1978. Effect of external environmental factors on chromosome pairing in autotetraploid rye. Cytologia 43: 21-34.

Jain, S.K., 1960. Cytogenetic of rye. Bibl. Genetica 29: 86

Klinga, K., 1986a. Aneuploidy in induced Autotetraploid populations of *Festuca pratensis, Lolium multiflorum* and *Lolium perenne*, I. The Frequency of aneuploids and performence of spaced plants in two autotetraploid populations of *Festuca pratensis.* Hereditas 104:75-83.

Klinga, K., 1986b. Aneuploidy in induced Autotetraploid populations of *Festuca pratensis, Lolium multiflorum* and *Lolium perenne*, I. The Frequency of aneuploids and performence of spaced plants in *Lolium multiflorum* and *Lolium perenne.* Hereditas 104:121-130.

Kün, E. 1988. Serin İklim Tahılları. Ankara Üniv. Ziraat Fak. Yay. 875, 254-255.

Luckett, D.J., 1989. Colchicine mutagenesis is associated with substantial heritable variation in cotton. Euphytica 42: 177-182.

Narasinga, P.S.R.L., J.V. Pantulu, 1982. Fertility and meiotic chromosome behaviour in autotetraploid pear millet. Theor. Appl. Genet. 62: 345-351.

Poehlman, J.M., 1995. Breeding Field Crops. Iowa State University Press Ames, Iowa

Reiman- Philipp, R., 1986. Perennial spring rye as a crop alternative. J. Agron. and Crop Sci., 157: 281-285.

Richard, R., C. Wang, 1987. Diploid perennial intergeneric hybrids in the tribe *Triticeae*. III. Hybrids among *Secale montanum*, *Pseudoroegneria spicata* and *Agropyron mongolicum*. Genome 29: 80-84.

Sağsöz, S., 1982. Farklı İngiliz çimi çeşitlerinde poliploid bitki elde etme olanakları üzerinde bir araştırma. Atatürk Üni. Yay. No: 596, Zir. Fak. Yay. No: 277, 1-14.

Sağsöz, S, M. Tosun, İ. Akgün (1998). Frequency of aneuploids in $C_1$ and $C_2$ generations of tetraploid ryegrass (*Lolium perenne* L.). Tukish Journal of Field Crops 3 (1): 1-4.

Sağsöz, S., M. Tosun, İ. Akgün, 2002. Use of anaphase I separations and the number of micronuclei in tetrads for selection of fertility in ryegrass. Israel J. Plant Sci.,50 (3), 181-188.

Simonsen, O., 1973. Cytogenetic investigations in diploid and autotetraploid populations of *L. perenne* Huds. Hereditas 79: 73-108.

Stutz, H.C., 1972. On the origin of cultivated rye. Am. J. Bot. 59: 59-70.

Tosun, M, K. Haliloğlu, M.S. Taşpınar, S. Sağsöz, 2003. Test weight, kernel shrivelling and aneuploidy frequency in triticale. New Zealand Journal of Agricultural Research 46:27-30.

Tutluer, M.I., 1993. Possibilities of obtaining forage rye from perennial rye by gamma radiation and variations found during meiosis and mitosis. PhD Dissertation, Institute of Applied Science, Ankara University

Weirmarch, A., 1975. Kernel size and frequency of euploid in Octoploid Triticale. Hereditas 80, 69-72.

Wricke, G., 1972. Populations Genetic. Walter de Gruyter, Berlin-NewYork