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PJBS

ISSN 1028-8880

# Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Pakistan Journal of Biological Sciences 7 (7): 1229-1235, 2004
ISSN 1028-8880


# Some Agronomical and Cytological Properties of Wild Sheep's Fescue Ecotypes (*Festuca ovina* ssp.)

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**Abstract:** In this study, the number of chromosome was determined in the wild ecotypes of *Festuca ovina* ssp., collected from five different natural pasture locations in Erzurum (Eastern Turkey). As a result of examinations, it was determined that the plants collected from Ispir town were diploid (2n=14), the plants from Tuzcu village, Koprukoy town and Uzundere town were tetraploid (2n=28) and those from the pasture of Ataturk University in Erzurum city were hexaploid (2n=42). There were no aneuploid plants in the collected ecotypes. The ecotypes (Ispir, Uzundere and University) of 3 various locations with different ploidy types were cultivated in the field for 2 years. During the cultivation period, the plant height, hay yield and plant crude protein content and yield were determined during various cutting times for each of the ecotypes. Evaluations were made according to the means of two years. Plant height (15.67-55.05 cm), fresh hay yield (22.71-65.52 g plant$^{-1}$) and dry hay yield (9.12-31.54 g plant$^{-1}$) and protein yield (1.49-3.47 g plant$^{-1}$) significantly increased until the cutting time of 16 June and the subsequent cutting reduced yield after this date. The crude protein content (7.74-16.42%) reduced as the cutting time was delayed. The maximum plant yield for the hexaploid plants was reached at the end of May while the maximum plant yield for diploid and tetraploid plants was at the end of June. The effect of ploidy type was significant on plant height and crude protein content. The diploid ecotypes had longest plant height (40.37 cm) and the highest crude protein content (12.99%).

**Key words:** Wild *Festuca ovina* ssp, ploidy, cutting time, hay yield, crude protein

## INTRODUCTION

Pastures and meadows comprises of 28% of agricultural land of Turkey. A large proportion was pastures (about 97%) while the proportion of meadows was only 3%. Eastern Anatolia region comprises of 58.4% of Turkey's total pasture and meadow lands[1]. However, the management and improvement of these lands are necessary for the productive and efficient use of these lands since 90% of the plant ecology of the pastures and meadows were destroyed due to heavy grazing[2]. The fact that the botanical compositions of Erzurum's pastures were dominated with *Festuca ovina* L. Gökkus and Koç[1] indicated that this plant is well adapted to the heavy grazing and extreme ecological conditions of the region.

All the species of *Festuca ovina* have been lately developed from the wild ecotypes. Thus, the differences between commercial varieties and the wild ecotypes are not so great[3]. *Festuca ovina* grass forage is scattered throughout Alpine and subalpine zones and resistant to the extreme conditions of cold and hot weather[3,4].

The basic chromosome number of *Festuca ovina* L. which is cross-pollinated is n=7. But, there are various subspecies with a chromosome number of 2n= 14, 28, 42 and 70[5]. According to the records of O.E.C.D there have been 33 varieties of *Festuca ovina* ssp. A large proportion of these varieties belong to the subspecies of *Festuca ovina* ssp. *duriuscula* which is a hexaploid. Other varieties are classified within the subspecies of *Festuca ovina* ssp. *capillata, Festuca ovina* var. *tenuifolia, Festuca ovina* ssp. *vulgaris* and *Festuca ovina* ssp. *vallesiaca*[6].

Cultivation and improvement of naturally grown plants will provide an opportunity for new varieties to be developed suitable for the climate conditions of the region Erzurum. In addition, identifying the regional varieties and collecting the species which can be annihilated under the heavy grazing circumstances could strengthen scientific efforts to preserve the natural genes sources of these species and would provide important materials for the improvement studies. In order for these plants to be used in the improvement studies, some important cytological and agronomic properties of these species must be given priority.

## MATERIALS AND METHODS

In the study, *Festuca ovina* ssp. plants were used which were grown naturally in Erzurum region. Plant

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Table 1: Soil compositions and geographical features of the collection points of *Festuca ovina* ssp. in Erzurum

| Locations | Attitude | Soil composition |
|---|---|---|
| Ispir town | 1150 | Pasture, pH = 7.32, Salt = 0.6%, $CaCO_3$ = 1.75%, $P_2O_5$ kg $da^{-1}$ = 1.12, $K_2O$ kg $da^{-1}$ = 58.50, Organic matter = 3.0% |
| Tuzcu village | 2350 | Pasture, pH = 8.16, Salt = 0.250%, $CaCO_3$ = 2.30%, $P_2O_5$ kg $da^{-1}$ = 1.29, $K_2O$ kg $da^{-1}$ = 61.20, organic matter = 3.32% |
| Koprukoy town | 1610 | Pasture, pH = 7.95, Salt = 0.3%, $CaCO_3$ = 0.5%, $P_2O_5$ kg $da^{-1}$ = 1.25, $K_2O$ kg $da^{-1}$ = 175.30, Organic matter = 2.93% |
| Uzundere town | 950 | Pasture, pH = 8.03, Salt = 0.028, $CaCO_3$ = 3.27%, $P_2O_5$ kg $da^{-1}$ = 0.92, $K_2O$ kg $da^{-1}$ = 96.15, Organic matter = 0.96% |
| Ataturk University | 1850 | Pasture, pH = 7.86, Salt = 0.03%, $CaCO_3$ = 0.95%, $P_2O_5$ kg $da^{-1}$ = 2.98, $K_2O$ kg $da^{-1}$ = 52.7, Organic matter = 0.51% |

materials were collected with their roots from 5 different locations of Erzurum (Ataturk University, Ispir, Tuzcu, Koprukoy and Uzundere) between April and May. Soil samples from the above collection points were taken from a 20 cm depth and analyzed. Soil compositions and geographical features of the collection points were presented in Table1.

The climate of Erzurum placed in East Anatolia region was cold during the winter and dry during the summer. Average annual rainfall was <500 mm and average annual temperature was around 6°C. The rain falled in winter and spring months. Generally from the beginning of June until the middle of September the whole period was dry.

**Glasshouse experiment:** The collected plants from different locations were transferred into the pots (each location 15 plants, total 75 plants) and kept in the glasshouse conditions. The plants, which were kept in the glasshouse conditions for about 3 to 4 months, the numbers of chromosomes were determined. In the study of somatic chromosomes squashing root-tip method was used. Four or five samples of root points were taken from each plant sample placed in the α-monobromnaphthalene tubes and kept at room temperature for 8 h .The samples then were transferred to Carnoy's solution (6:3:1) and kept at room temperature for 2 h. This was followed by placing the samples in the tubes containing 1N HCl at 60°C for 20 min and then the samples were hydrolyzed. The staining was done by Feulgen solution. The prepared samples were then examined under the microscope to count the chromosome number.

**Field trial:** At the end of chromosome determination, only the plants of one location from the same ploidy group (Uzundere) were selected and field trial included 3 different ecotypes on the whole (Ispir, Uzundere and University). The plants to be under the glasshouse condition grown were transferred to the field condition in April of 1998. The plants belonging every locations were planted in a field with a 30 cm spaced plants The samples were started to be taken 5th April 1999 and continued till the middle of July in which the vegetation was entirely dry. The growth period was until the mid-July 1999. During the whole growth period, there were 7 sampling times in an interval of 2-weeks period (15th April, 1st May, 16th May, 1st June, 16th June, 1st July and 16th July). From each location, 5 samples were taken during each of cutting/sampling period. The field experiment lasted for 2 years and the evaluations were pooled for both years. In the field trial, plant height from the soil surface to the plant top, green hay yield (weight of fresh vegetative parts), dried hay yield (weight of dried vegetative parts), crude protein content (nitrogen content x 6.25) and crude protein yield (crude protein content multiply by dried plant yield) were determined for all the samples. The data was analyzed in SPPS program under the Windows operating system by multivariate analyses of variance. The differences between the means were separated by Duncan's multiple comparison tests.

## RESULTS

**Cytological studies:** Totally seventy-five plants species, 15 plants from each group, were examined for the determination chromosome number. As result of the microscopic examination, the species of Ispir location was identified as diploid (2n=14), the species from Tuzcu, Uzundere and Koprukoy as tetraploid (2n=28) and those from University location as hexaploid (2n=42) whereas no species were identified as to be aneuploid.

### Field trial

**Plant height:** According to two-year averages, the plant height significantly differed between the locations depending on the cutting time and ploidy types ($P<0.01$). The average of plant height with regard to cutting time ranged from 15.67 to 55.05 cm (Table 2). The highest plant height was observed in 16th of June, the plants of this group significantly differed from the plants of other cutting groups.

When examined according to the ecotypes or ploidy types, the plant heights was 40.37 cm in the diploid plants (Ispir), 39.88 cm in the tetraploid plants (Uzundere) and 34.47 cm in the hexaploid plants (University) (Table 3). The plants of hexaploid was the shortest plants and significantly differed from the other groups. The interaction between the cutting time and ploidy type was also significant ($P<0.01$) (Fig. 1).

Table 2: Mean values of studied parameters of the different ecotyped of *Festuca ovina* ssp. by the cutting times

| | Cutting time | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Parameters | 15 April | 1 May | 16 May | 1 June | 16 June | 1 July | 16 July | Std Error |
| Plant height (cm) | 15.67d | 17.27d | 33.33c | 48.33b | 55.05a | 49.83b | 48.20b | 1.825 |
| Total fresh hay yield (g plant$^{-1}$) | 22.71c | 25.43c | 60.73a | 60.59a | 65.52a | 42.33b | 31.67bc | 4.384 |
| Fresh leaf yield (g plant$^{-1}$) | 22.71ab | 24.55a | 25.66a | 18.47ab | 24.85a | 15.87b | 8.91c | 2.390 |
| Fresh stem yield (g/plant) | 0.00d | 0.88d | 35.08ab | 42.13a | 40.67a | 26.45bc | 22.76c | 4.068 |
| Total dry hay yield (g plant$^{-1}$) | 9.12c | 12.73c | 22.30b | 23.00b | 31.54a | 22.38b | 19.52b | 2.312 |
| Dry leaf yield (g plant$^{-1}$) | 9.12ab | 12.37a | 10.10ab | 7.47b | 10.62ab | 8.67ab | 7.55b | 1.247 |
| Dry stem yield (g plant$^{-1}$) | 0.00c | 0.35c | 12.20b | 15.53ab | 20.92a | 13.71b | 11.97b | 2.138 |
| Crude protein (%) | 16.42a | 16.37a | 14.75b | 12.30c | 10.92d | 10.12e | 7.74f | 0.263 |
| Crude protein yield (g plant$^{-1}$) | 1.51d | 2.12bc | 3.30a | 2.82ab | 3.47a | 2.25bc | 1.49d | 0.296 |

Table 3: Mean values of studied parameters of the different ecotypes of *Festuca ovina* ssp. by the ploidy types

| | Ploidy levels | | | |
|---|---|---|---|---|
| Parameters | 2n=14 (İspir) | 2n=28 (Uzundere) | 2n=42 (Atatürk Üniv.) | Std Error |
| Plant height (cm) | 40.37a | 39.88a | 34.47b | 1.194 |
| Total fresh yield (g plant$^{-1}$) | 42.80 | 44.63 | 44.99 | 2.870 |
| Fresh leaf yield (g plant$^{-1}$) | 19.66 | 20.39 | 20.38 | 1.565 |
| Fresh stem yield (g plant$^{-1}$) | 23.14 | 24.24 | 24.61 | 2.663 |
| Total hay yield (g plant$^{-1}$) | 19.94 | 20.24 | 20.07 | 1.514 |
| Dry leaf yield (g plant$^{-1}$) | 9.53 | 9.77 | 8.93 | 0.816 |
| Dry stem yield (g plant$^{-1}$) | 10.40 | 10.47 | 11.14 | 1.400 |
| Crude protein (%) | 12.99a | 12.33b | 12.66ab | 0.172 |
| Crude protein yield (g plant$^{-1}$) | 2.48 | 2.35 | 2.44 | 0.194 |

**Green hay yield:** Total fresh hay yield and the proportion of stem and leafs within the total yield significantly ($P<0.01$) differed according to the cutting times. No difference was found considering the ploidy types in terms of three characteristics. Depending on the cutting times, total green hay yield ranged from 22.71 to 65.52 g plant$^{-1}$ and the yield of plants between 16th May and 16 June was the highest and significantly differed from the plants of other groups (Table 2). Fresh hay yields were 42.80 g plant$^{-1}$ for diploid group, 44.63 g plant$^{-1}$ for tetraploid group and 44.99 g plant$^{-1}$ for hexaploid group (Table 3). Significant interaction ($P>0.01$) was observed between the cutting time and ploidy type (Fig. 1).

Statistically the percentages of leaf and stem yield were significant for cutting times but non-significant for ploidy types (Table 2 and 3). The highest leaf yield was obtained from 16th May (25.66 g plant$^{-1}$). However, this value did not significantly differ from the leaf yield of plants obtained from the cutting periods until 16th June. So it was in the same group. The lowest leaf yield (8.91 g plant$^{-1}$) was obtained from the last cutting time. Stem yield ranged from 0.00 to 42.13 g plant$^{-1}$ as overall. The highest stem yield was at 16th May (35.08 g plant$^{-1}$) and 16th June (40.67 g plant$^{-1}$) and there were no significant differences between the cutting times. The lowest stem yield was at the first two cutting times. According to the ploidy type, the leaf yield ranged from 19.66 to 20.39 g plant$^{-1}$ and stem yield ranged from 23.14 to 24.61 g plant$^{-1}$. The interaction between cutting time and ploidy type was significant ($P<0.05$) for leaf yield and non-significant for stem yield (Table 3 and Fig. 1).

**Dry hay yield:** Dry hay yield, leaf and stem yield were statistically ($P<0.01$) differed by cutting time (Fig. 2). Overall mean of dry hay yield was between 9.12 to 31.54 g plant$^{-1}$ and reached the highest yield at 16th June. Dry hay yield at 16th June significantly differed from the yields of other cutting times. Dry leaf yield ranged from 7.47 to 12.37 g plant$^{-1}$ and dry stem yield ranged from 0.00 to 20.92 g plant$^{-1}$. The highest leaf yield (12.37 g plant$^{-1}$) was obtained from 1st May. However, no significant differences were observed between the other cutting times (9.12 g plant$^{-1}$ at 15th April, 10.10 g plant$^{-1}$ at 16th May, 10.62 g plant$^{-1}$ at 16th June and 8.67 g plant$^{-1}$ at 1st July). Stem yield reached the highest yield (20.92 g plant$^{-1}$) at 16th June and did not differ from the yield at 1st June (15.53 g plant$^{-1}$) (Table 2).

Although no significant differences were observed according to the ploidy types or ecotypes, the highest dry hay yield (20.24 g plant$^{-1}$) and leaf yield (9.77 g plant$^{-1}$) were obtained from the group of tetraploidies (Uzundere) while the highest stem yield (11.14 g plant$^{-1}$) was obtained from the hexaploid (Table 3). No interaction effect was observed between cutting time and ploidy type on all three parameters.

**Crude protein content:** The effects of cutting time and ploidy type were significant ($P<0.05$) on crude protein content. The average crude protein content ranged from

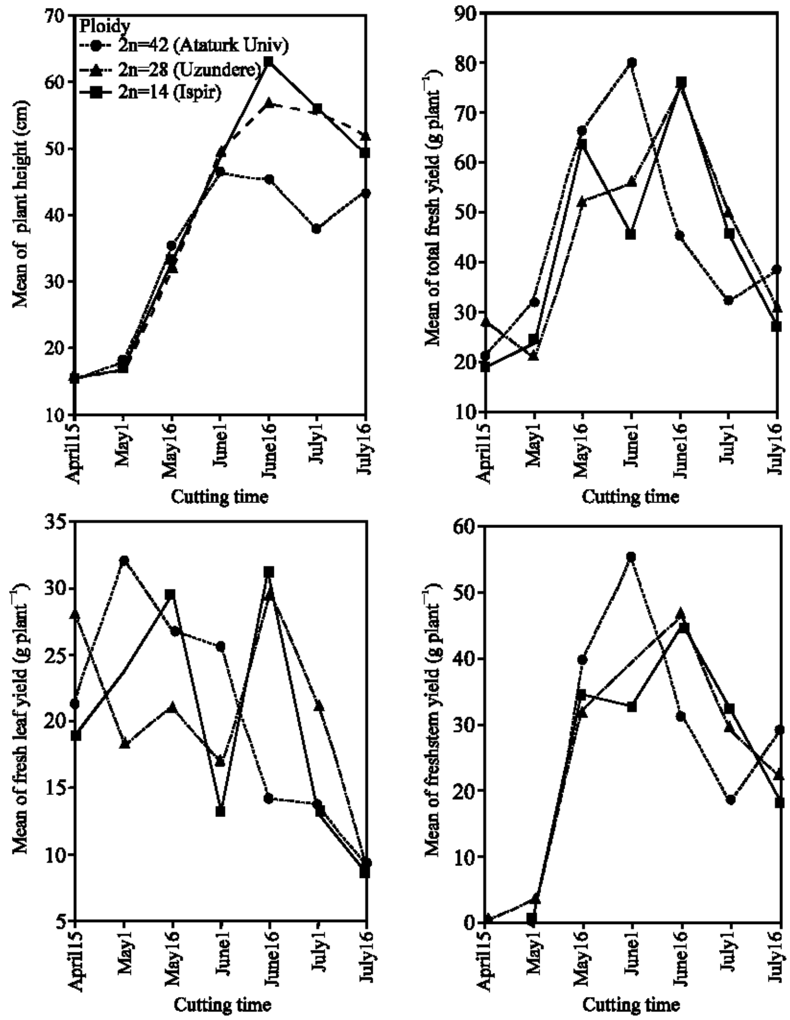


Fig. 1: Changes in plant height, total fresh hay yield, fresh leaf yield and fresh stem yield of *Festuca ovina* ssp. of different diploid types by cutting time

7.74 to 16.42% between the cutting times. Although the first two cutting times were in the same group according to the Duncan test, others differed significantly and crude protein content reduced as the cutting times increased (Table 2 and Fig. 2).

The crude protein content was 12.99% for diploid group (Ispir), 12.33% for tetraploid group (Uzundere) and 12.66% for hexaploid group (University). The difference between Ispir and Uzundere groups was found to be significant (Table 3) while the interaction effect of cutting time x ploidy type was not significant on the crude protein level.

**Crude protein yield:** The crude protein yield, which was calculated by multiplying hay yield with crude protein content (Table 2 and 3). Total protein yield per plant ranged from 1.49-3.47 g and there seemed to be a significant effect of cutting times ($P<0.01$) on total protein yield. Total protein yield insignificantly differed between the cutting periods of 16th May, 1st and 16th June. The effect of ploidy type was not significant on crude protein yield the corresponding yields were 2.48, 2.35 and 2.44 g plants$^{-1}$ for diploid, tetraploid and hexaploid, respectively.

## DISCUSSION

The population of sheep's Fescue collected from various ecotypes significantly differed in the number of chromosomes: The plants of Ispir was diploid, the plants of Uzundere, Tuzcu and Korukoy were tetraploid and those from University was hexaploid. Aiken and Fedak[7] reported that the plants of Eurasia and North America were predominantly tetraploid and hexaploid species. No

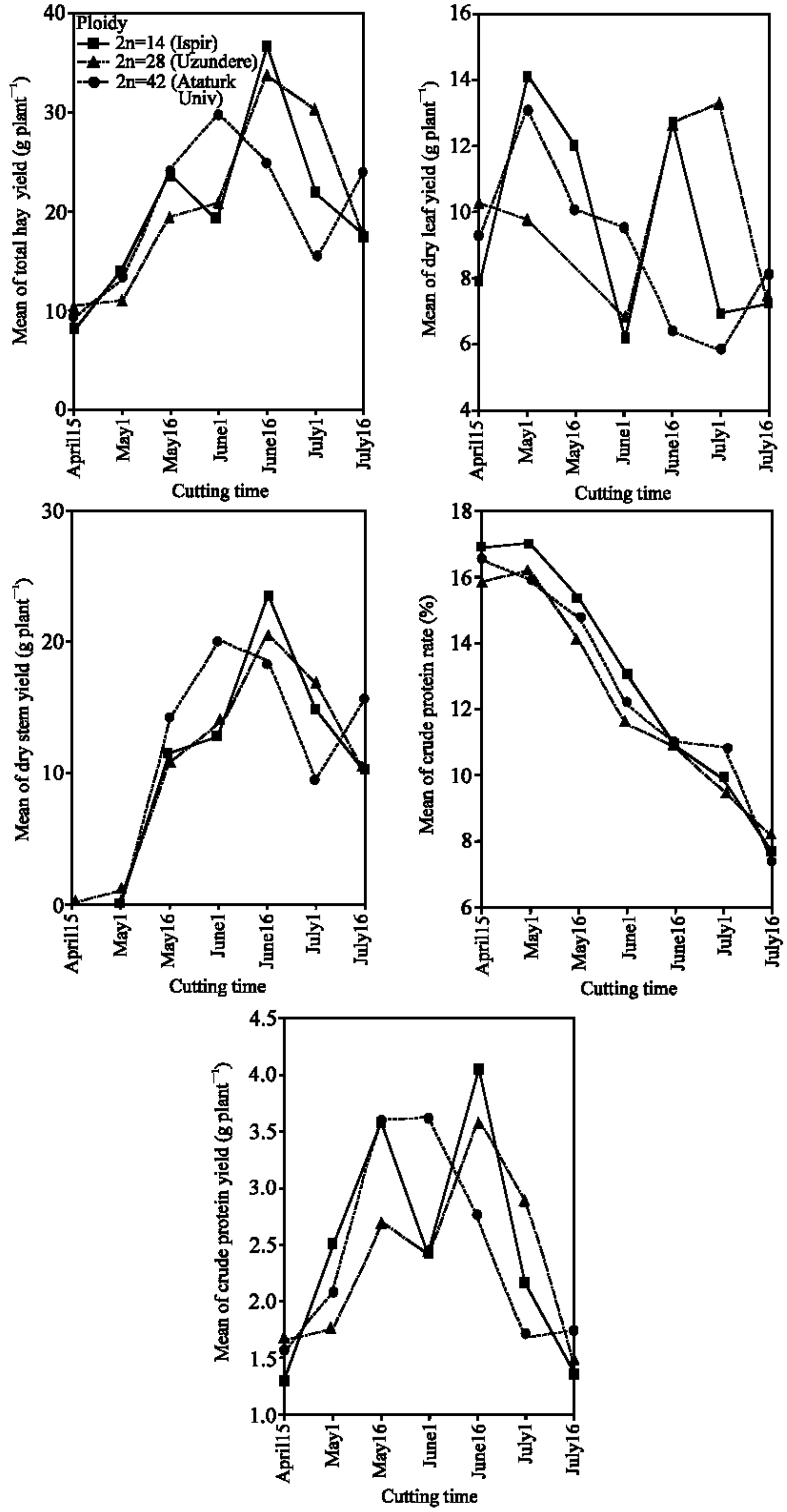


Fig. 2: Changes in dry hay yield, dry leaf yield, dry stem yield and crude protein contents of *Festuca ovina* ssp. of different location by cutting times

aneuploidy plants were determined in the studied location during the course of investigation. This could be due to the fact that the plants of aneuploidy had low rate of shooting or weak growth rates. So they are easily eliminated from the natural population. In addition, this can indicated that the wild species have been growing in nature for along time and thus they could have reached to a cytological balance.

This study allowed determining the protein content of plants obtained from three populations of sheep's Fescue and analyzing the biomass of surface soil formed by wild plants for long periods. Evaluations were made according to the data of 2 years and the results revealed significant differences between various cutting times. During vegetation period including all the cutting periods, the plants appear to grow slowly at the beginning of vegetation period, then the growth rate gradually increased and finally the growth of plant reduced drastically at the end of vegetation period[8]. The plant height and plant yield are the indicators of plant growth during the whole session. In the present study, these two parameters reached to the highest value at 16th June and thereafter the plant height and yield reduced. The reductions in plant height and plant yield were seen to be parallel to the reductions in leaf and stem. The flowering started at the beginning of June in sheep's fescue and continued until 16th June. The highest fresh and dry hay yield was obtained from the diploid and tetraploid types at 16th June whereas the corresponding highest values were obtained for hexaploid types at 1st June. In all three ploidy types, the crude protein content drastically reduced by increasing cutting time.

All the vegetative parts of plants were leaf at 1st cutting time whereas the stem was just obtained at 2nd cutting period. The water content of plants at 1st cutting period was over 60% while the water content reduced to 30% and less toward to the last cutting period. At the beginning of vegetation period, the plants seemed to have low dry matter content, perhaps due to the carbohydrate metabolism of the plant. The fact that a large amount of carbohydrate stored in the stem leads to increase in the mass of stem[9]. Dry matter yields can be usually affected by a number of genetic, cultural and environmental factors[10]. Quality changes with maturity are related to reduced stem quality and an increased proportion of stem in the forage. The leaves comprises of digestible nutrients than do stems[11].

The plants are highly active at the start of growth period during which there is a great importance of endogenous enzymes and increased amount of nitrogen, thereby increased crude protein content, in their activities[12]. During subsequent periods of growth, the crude protein content of the plants reduces while the cellulose content decreases due to the increased content of carbohydrates[13]. However, forage quality generally affected by genetic, environmental management and physical factors[10].

The effect of ploidy type on plant height and crude protein level was significant while there were no significant effects of ploidy types on other studied parameters. The plants of diploid and tetraploid were significantly taller than the plants of hexaploid. Pavlick[14] reported that the plants height of wild sheep's fescue ranged from 6.5 to 50 cm. In the present study, the plants with highest protein content were the diploid plants of Ispir location whereas there were significant differences among the groups in crude protein yield.

In conclusion, significant differences were observed among the plants groups in chromosome number. On the other hand, there were no significant differences among the groups in other studied parameters. The results revealed that the plants of hexaploid were seen to grow up until the beginning of June in which they had to be harvested while the plants of tetraploid and diploid could grow up to the middle of June and could lately be harvested. It was also observed that the more delay in cutting time the more reduction in hay yield.

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