



# The effect of plant density and nitrogen fertilizer on leaf area, chlorophyll, and grain protein of *Zea mays* in competition with *Amaranthus retroflexus*

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## Abstract

In order to investigate the effect of plant density and different amounts of nitrogen fertilizer on yield components, leaf area, chlorophyll and grain protein content of *Zea mays* cultivars and *Amaranthus retroflexus* biomass, an experiment was conducted in two crop years (2012-2013) in the form of a split plot based on a randomized complete block design with four replications. The main factors included maize cultivars (single cross hybrids 370 and 704) and the sub-factors included different levels of maize densities (60000, 70000, 80000, and 90000 plant ha<sup>-1</sup>) and different levels of nitrogen fertilizer including 200, 300, 400, and 500 kg ha<sup>-1</sup>. The effect of density on all traits was significant while the effect of cultivar on chlorophyll and leaf area index, and also the effect of nitrogen on the number of ears per square meter were not significant. Maize cultivar 704 had significantly higher number of grain per ear, weight of one thousand grains, and grain protein compared to cultivar 370 but the number of ears was significantly lower than that of cultivar 370 and there was no significant difference in chlorophyll content and leaf area index between the two cultivars. The highest grain protein was obtained at 80000 plant ha<sup>-1</sup> density and application of 400 kg ha<sup>-1</sup> N. Also, cultivar 704 significantly decreased redroot biomass compared to cultivar 370. According to the findings, cultivation of cultivar 704 is recommended with a density of 80,000 plant ha<sup>-1</sup> along with application of 400 kg N ha<sup>-1</sup>.

**Keywords:** fertilizer, leaf, maize, protein, weed

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## Introduction

Corn (*Zea mays* L.) is a valuable crop with high compatibility and nutritional value making it one of the most important crops in the world (Cox et

al., 2006). In fact, based on the area under cultivation and production, it is the third most important crop after wheat and rice in the world (Oktem et al., 2004).

Yield components and physiological characteristics such as leaf area index are affected by intra-plant competition for photosynthetic

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material distribution and extra-plant competition for the use of environmental growth factors (Abuzar et al., 2011). Corn is very sensitive to plant density and the desired yield is not achieved under low density. On the other hand, excessive plant density causes sterilization of flowers and reduced number of seeds (Hashemi et al., 2005). Adjustments for the optimum density of corn are recommended to make plants produce tiller. Proper spacing between plants reduces competition between plants, thus increasing the absorption of water, light, and nutrients, which increases photosynthesis and ultimately grain performance. Raja (2001) reported that the decrease in grain weight due to the increase in density is related to the reduction of grain dry matter accumulation during the three weeks after flowering to the physiological maturity stage. Purcell et al. (2002) reported that with increasing soybean density from 49300 to 59500 plant ha<sup>-1</sup>, protein yield increased from 1.02 to 1.9 tons ha<sup>-1</sup>. With increasing density, competition between plants increases, so the leaf area of the plant decreases, but due to the increase in the number of plants per unit area with the development of leaf area, the amount of light absorption and photosynthetic material increases (Shapiro and Wortman, 2006).

Nitrogen plays an essential role in plant growth and is highly related to the amount of water and how water is distributed (Azeez and Adetunji, 2007). Proper fertilizer management leads to optimal use of nitrogen by plants and reduces nitrogen fertilizer leaching and thus prevents ecosystem degradation (Sedlar et al., 2011). Leaf area index and protein yield of corn, like other crops, are affected by chemical fertilizers (Di paolo and Rinaldo, 2008). Factors such as climate, crop rotation, drainage, irrigation, fallow, weed species, and weed density are effective in crops response to chemical fertilizers (Sedlar et al., 2011). Shafi et al. (2011) treated barley (*Hordeum vulgare*) with 40, 60, and 80 kg ha<sup>-1</sup> nitrogen and recorded the highest protein yield under application of 80 kg ha<sup>-1</sup> nitrogen. Skarson et al. (1999) showed that increasing nitrogen in the form of urea fertilizer up to 180 kg ha<sup>-1</sup> caused a significant increase in leaf area, chlorophyll content, number of spikes, and

number of grains per spike. Zaman and Khan (2016) showed that grain yield in corn at different levels of nitrogen with the presence of weeds was much lower compared to non-weed conditions. Excessive application of nitrogen fertilizers causes nitrate contamination of water sources (Evans et al., 2003; Abouzienna et al., 2007). Regarding the effect of nitrogen fertilizer on the relative ability of wheat to compete with weeds, Blackshaw et al. (2004) reported that at high levels of nitrogen, wild mustard is more competitive against wheat, but under the same conditions, bermuda grass weed was less competitive than wheat. Lindquist et al. (2010) reported that the presence of nitrogen fertilizer significantly increases the competitiveness of maize against weeds.

Weeds are an integral part of agricultural and non-agricultural ecosystems and one of the most important factors reducing crops (Kolb et al., 2012). In general, the term interference is used to describe the effect of a plant on the environment of other plants (Olsen et al., 2012). Competition is one of the forms of negative interference, i.e., it is a negative interaction amongst plants that use a limited environmental factor (Barker et al., 2006). Abdollahian and Froud (1997) reported that Lamb (*Chenopodium album*) weed competition with sugar beet significantly reduced sugar beet yield. One of the most important weeds in corn fields is redroot (*Amaranthus retroflexus*), which is a broadleaf annual weed of the Amaranthaceae family (Makarjian et al., 2003). Redroot stays in the field for a long time due to the high capacity of a plant to produce seeds in one year and its ability to disperse by the wind. Lindquist et al. (2010) reported that competition between *Amaranthus retroflexus* and crop reduced crop yield by 50%. Vazin (2012) showed that redroot in competition with corn reduced the rate of leaf area development, leaf area index, and number of leaves in corn. The purpose of this study was to determine the best density and amount of nitrogen fertilizer in increasing the yield components, protein yield, leaf area index, and chlorophyll content of corn and increasing the competition of corn against redroot (*Amaranthus retroflexus*).

## Materials and Methods

This research was conducted in the Agricultural and Natural Resources Research Center of Miandoab with a longitude of 48 degrees and 52 minutes east, latitude of 35 degrees and 20 minutes north, and 1300 meters above sea level in Miandoab, East Azarbaijan, Iran. The experiment was conducted in two crop years (2012-2013) in the form of a split plot based on a randomized complete block design with four replications. The main factors included maize cultivars (Single Cross hybrids 370 and 704) and sub-factors included different levels of maize densities (60000, 70000, 80000, and 90000 plant ha<sup>-1</sup>) and different levels of nitrogen fertilizer including 200, 300, 400, and 500 kg ha<sup>-1</sup> from the source of urea fertilizer. The texture of soil was sandy clay loam and pH was 7.8. Nitrogen fertilizer in the form of urea was applied based on experimental treatments in two stages of planting and 50 cm height of corn. Corn hybrids were planted in the ridges. Each plot had four planting rows of five meters in length and with a distance between the planting rows of 75 cm.

The predominant weed in the area was redroot, which was collected from farms around the area and prepared from the Agricultural Research Center and planted in a corn field at a distance of 10 to 15 cm of corn and at depth of two cm before all the other weeds of the field were weeded, except the redroot. The first irrigation was done immediately after planting corn. At the end of the growing season and with physiological maturation of corn, sampling was performed by removing two rows of side plantings and randomly selecting 7 plants to measure study traits (number of ears per plant, number of grains per ear, and weight of one thousand grains). Corn leaf area was measured with an area meter (Model 3100-Li) at the emergence stage of tassel and the following equation was used to determine the leaf area index (Soleymani et al., 2003):

$$LAI = LA/A$$

where LAI, A, LA are leaf area index, sampling area, and leaf area, respectively.

At the stage of tassel emergence, the average chlorophyll index was measured with SPAD-502 at three leaf points (tip, middle and base

of the leaf) including flag leaf and leaf of pre-flag leaf (Costa et al., 2011). Grain protein yield was determined by method of Abedi et al. (2011). The dry weight of redroot was weighed after transfer to the laboratory. MSTATC and EXCE software were used to analyze the treatments and draw the graphs, respectively. Duncan test (5%) was used to compare the means.

## Results

### Number of ears per square meter

Analysis of variance showed that the effect of cultivar ( $p \leq 0.01$ ), the effect of density ( $p \leq 0.05$ ), the interaction effect of fertilizer and cultivar ( $p \leq 0.05$ ), and the interaction effect of fertilizer and density ( $p \leq 0.01$ ) were significant on the number of ears per square meter (Table 1). Among the two cultivars studied, cultivar 370 with an average of 47.69 ears per square meter had a higher number of ears than cultivar 704 with an average of 40.11 ears per square meter (Table 2). Comparison of the mean effects of different levels of density on the number of ears per square meter showed that with increasing plant density, the number of ears per square meter also increased. The density level of 90000 plants ha<sup>-1</sup> with an average of 42.41 ears per square meter had the highest number of ears and compared to the density levels of 60000, 70000, and 80000 plant ha<sup>-1</sup>, the number of ears increased by 34.50%, 15.36%, and 2.46%, respectively.

The lowest number of ears per square meter with an average of 31.53 belonged to the density level of 60000 plants ha<sup>-1</sup>. However, no statistically significant difference was observed between the levels of 80000 and 90000 plants ha<sup>-1</sup> (Table 3). Comparison of the means of interaction between fertilizer and cultivar treatments showed that increasing the fertilizer level in cultivar 370 could not have a significant effect on the number of ears per square meter, but it caused a significant increase in cultivar 704. In addition, in all fertilizer levels, cultivar 370 had the highest number of ears per square meter compared to cultivar 704 (Fig. 1). Comparison of the means of interaction between density and nitrogen fertilizer showed that the combined density of 90000 plants and the fertilizer level of

Table 1  
Analysis of variance of the study traits

S.O.V	df	Number of ears	Number of grains per ear	Weight of one thousand grains	Leaf area index	protein yield	Chlorophyll content	Biomass of redroot (g m <sup>-2</sup> )
Year	1	0.3	1505.5	0.74	21.9	296.4	52.7	22276.5
Repeat×Year	6	857.8	2442.2	165.8	87.2	1843.3	993.89	34386.9
Cultivar	1	14816.7**	19160.9*	11420.7**	116.9	6732.7*	315.2	85933.5**
Cultivar×Year	1	16.5	9759.3	44.7	93.4	8400.3	7.45	10661.1
Error <sub>a</sub>	6	1071.7	4332.2	802.6	10.5	5276.8	121.68	6132.7
Density	3	350.1*	24609.9*	13396.5**	535.8**	5972.6*	1428.2**	78268.6**
Density×Year	3	47.5	6699.5	248.8	75.6	442.2	17.88	2021.3
Density×Cultivar	3	227.2	7767.3	2142.8	88.7	9037.5**	423.3	46825.6*
Fertilizer	3	207.5	28798.4*	15867.3**	346.1*	8965.8**	1970.5**	63821.9**
Fertilizer×Year	3	17.1	16095.3	480.5	12.7	451.2	21.1	2865.3
Fertilizer×Cultivar	3	451.6*	25537.4*	6567.5	44.2	470.4	239.2	3953.7
Fertilizer×Density	9	523.2**	10071.2	6935.6*	119.5	4319.96*	475.5	89841.5**
Fertilizer×Year×Cultivar	3	29.4	7405.5	225.2	101.4	224.9	15.4	302.8
Fertilizer×Year×Density	9	25.5	6251.1	262.5	19.7	244.7	13.1	7641.1
Fertilizer×Cultivar×Density	9	19.5	7195.4	237.5	36.2	419.1	393.5	34435.4
Fertilizer×Cultivar×Density×Year	12	27.2	6083.2	343.1	58.3	501.9	17.2	3721.5
Error <sub>b</sub>	180	160.3	10077.3	3167.1	125.8	2171.7	342.16	15873.1

\* and \*\* are significant at 5% and 1% probability levels, respectively.

Table 2  
Comparison of the mean effects of cultivar on the studied traits

Treatments	Number of ears (m <sup>2</sup> )	Number of grains per ear	Weight of one thousand grains (g)	Leaf area index	Protein yield (g m <sup>-2</sup> )	Chlorophyll content (Spad)	Biomass of redroot (g m <sup>-2</sup> )
Cultivar SC 370	47.69 a	398.36 b	363.18 b	3.72 a	120.45 b	72.54 a	670.86 a
Cultivar SC 704	40.11 b	453.71 a	382.79 a	3.84 a	130.72 a	68.67 a	591.67 b

Means with different letters indicate a significant difference based on Duncan's test at 5% probability level.

500 kg ha<sup>-1</sup> and also the density of 60000 plants ha<sup>-1</sup> with a fertilizer level of 200 kg ha<sup>-1</sup> had the highest and lowest number of ears per square meter, by 44.32 and 30.11 ears in square meter, respectively (Table 5).

### Number of grains per ear

The results of analysis of variance of data on the number of grains per ear showed that the main effects of cultivar, density, fertilizer, and the interaction of fertilizer with density on the number of grains per ear were statistically significant at 5%

probability level (Table 1). Among the two cultivars studied, cultivar 704 had significantly higher number of grains per ear than cultivar 370 (Table 2). Comparison of the mean effects of different levels of density on the number of grains per ear showed that the number of grains per ear were significantly reduced with increasing plant density from 60000 to 90000 plants.

In this study, the density of 60,000 plant ha<sup>-1</sup> with an average of 512.85 grains per ear had the highest number of grains per ear. However, densities of 70000 and 80000 plants ha<sup>-1</sup> were not

Table 3  
Comparison of the mean effects of density on the study traits

Treatments	Number of ears (m <sup>2</sup> )	Number of grains per ear	Weight of one thousand grains (g)	Leaf area index	protein yield (g m <sup>-2</sup> )	Chlorophyll content (Spad)	Biomass of redroot (g m <sup>-2</sup> )
Density (Plant ha <sup>-1</sup> )	60000	31.53 c	512.85 a	405.5 a	2.46 d	106.31 c	74.95 a
	70000	36.76 b	474.05 b	396.12 ab	3.09 c	120.88 b	624.27 b
	80000	41.39 a	452.37 b	387.24 b	3.55 b	133.49 a	550.82 c
	90000	42.41 a	395.42 c	342.35 c	4.27 a	130.43 a	517.95 c

Means with different letters indicate a significant difference based on Duncan's test at 5% probability level.

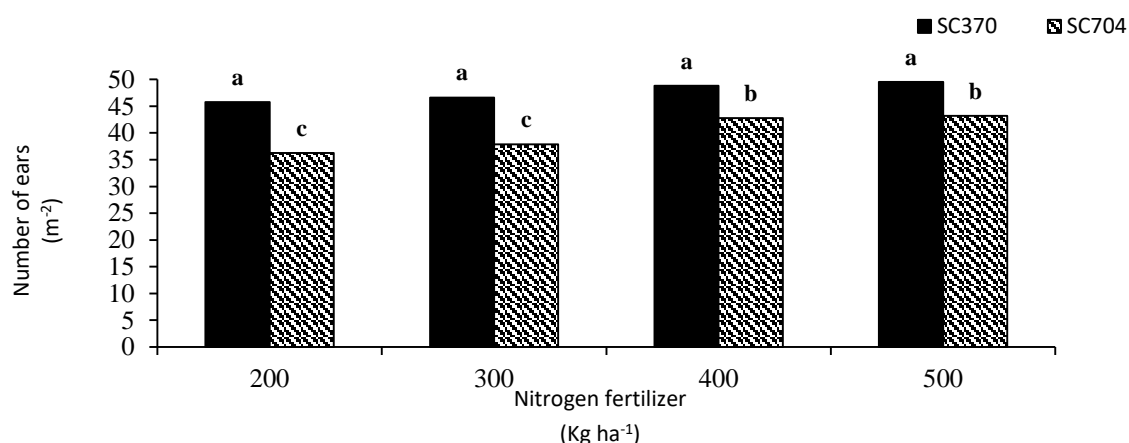


Fig. 1. Interaction effect of cultivar and nitrogen fertilizer on the number of ears; the means with different letters indicate a significant difference based on Duncan's test at 5% probability level.

significantly different (Table 3). Comparison of the mean effect of different levels of fertilizer on the number of grains per ear revealed that 200 kg N ha<sup>-1</sup> with an average of 366.2 grains had the lowest number of grains per ear. However, there was no statistically significant difference between 200 and 300 ha<sup>-1</sup> kg nitrogen fertilizer. The highest number of grains per ear with an average of 513.55 grains was related to 500 kg ha<sup>-1</sup>. However, there were no statistically significant difference between 500 and 400 kg ha<sup>-1</sup>. nitrogen fertilizer (Table 4). Densities of 60000 and 70000 plants ha<sup>-1</sup> in combination with a fertilizer level of 400 kg ha<sup>-1</sup> had the highest number of grains with an average of 609.36 and 595.76 grains per ear, respectively. Finally, the density of 90000 plant ha<sup>-1</sup> with a level of 200 kg N ha<sup>-1</sup> had the lowest number of grains per ear with an average of 312.56 grains per ear (Table 5).

#### Weight of one thousand grains

The results of analysis of variance showed that the main effects of cultivar, density and fertilizer at 1% level and the interaction effect of fertilizer with density at 5% level were statistically significant on weight of one thousand grains (Table 1). Cultivar 704 with 382.79 g had a higher weight of one thousand grains compared to cultivar 370 with 363.18 g (Table 2). With increasing the level of density, the weight of one thousand grains decreased, although this decrease was not statistically significant from the levels of 70000 and 80000 plant ha<sup>-1</sup> (Table 3).

Among the fertilizer levels, two levels of 400 and 500 kg N ha<sup>-1</sup> with 407.43 and 414.47 g, respectively, had the highest and the level of 200 kg N ha<sup>-1</sup> with 345.59 g had the lowest weight of one thousand grains (Table 4). The treatment combination with a density of 60000 plant ha<sup>-1</sup> and 500 kg of nitrogen fertilizer had the highest weight of one thousand grains (435.07 g) and the treatment combination with a density of 90000 plant ha<sup>-1</sup> and 200 kg of nitrogen fertilizer had the

lowest weight of one thousand grains (317.04 g) (Table 5).

statistically significant on corn protein yield (Table 1). As Table (2) suggests, protein yield in cultivar

Table 4  
Comparison of the mean effects of nitrogen fertilizer on the study traits

Treatments	Number of ears (m <sup>2</sup> )	Number of grains per ear	Weight of one thousand grains (g)	Leaf area index	Protein yield (g m <sup>-2</sup> )	Chlorophyll content (Spad)	Biomass of redroot (g m <sup>-2</sup> )
Nitrogen fertilizer (Kg ha <sup>-1</sup> )	200	35.78 a	366.2 b	345.59 b	2.19 d	114.77 b	63.81 c
	300	37.49 a	381.76 b	364.66 b	2.87 c	117.42 b	69.42 b
	400	38.70 a	573.17 a	407.43 a	3.64 b	130.01 a	74.06 a
	500	39.65 a	513.55 a	414.47 a	4.03 a	128.90 a	76.14 a

Means with different letters indicate a significant difference based on Duncan's test at 5% probability level.

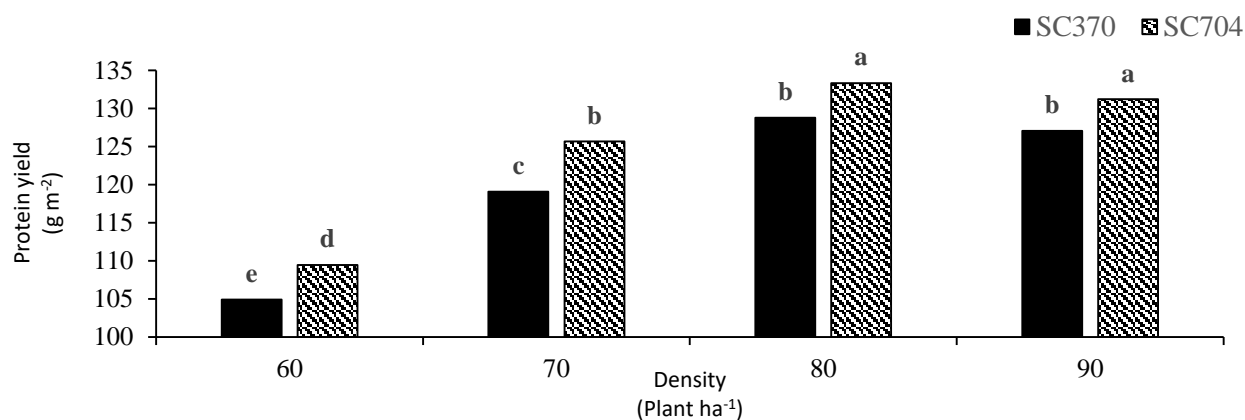


Fig. II. Interaction of cultivar and density on grain yield; means with different letters indicate a significant difference based on Duncan's test ( $p \leq 0.05$ ).

### Leaf area index

Leaf area index showed a significant difference (Table 1) under the effect of density ( $p \leq 0.01$ ) and application of nitrogen fertilizer ( $p \leq 0.05$ ). At densities of 60000, 70000, 80000, and 90000 plant ha<sup>-1</sup>, the leaf area index values were 2.46, 3.09, 3.55, and 4.27, respectively (Table 3). The leaf area indexes under treatments with 200, 300, 400, and 500 kg nitrogen were 2.19, 2.87, 3.64, and 4.03, respectively (Table 4).

### Protein yield

The effects of cultivar and density ( $p \leq 0.5$ ), fertilizer ( $p \leq 0.01$ ), combined treatments of cultivar and density ( $p \leq 0.01$ ), and combined treatments of fertilizer and density ( $p \leq 0.01$ ) were

704 (130.72 g m<sup>-2</sup>) was significantly higher than that in cultivar 370 (120.45 g m<sup>-2</sup>).

Comparison of the mean effect of different levels of density on grain yield (Table 3) showed that increasing plant density from 60000 to 80000 plants ha<sup>-1</sup> increased protein yield, but with increasing plant density from 80000 to 90000 plant ha<sup>-1</sup> decreased this variable although the reduction in protein yield was not statistically significant. In this study, the density of 80,000 plants ha<sup>-1</sup> increased the protein yield compared to 60000, 70000, and 90000 plant ha<sup>-1</sup> densities by 25.56, 10.43, and 2.24%, respectively. Comparison of the mean effect of nitrogen fertilizer levels on corn protein yield revealed that increasing the level of nitrogen fertilizer increased protein yield. The highest grain yield in corn with 130.01 g m<sup>-2</sup> was observed at 400 kg nitrogen treatment although there was no statistically significant

Table 5

Comparison of the mean interaction effects of density with nitrogen fertilizer on the number of ears, number of grains per ear, weight of one thousand grains, grain yield, and biomass of redroot

Density (Plant ha <sup>-1</sup> )	Nitrogen fertilizer (Kg ha <sup>-1</sup> )	Number of ears (m <sup>2</sup> )	Number of grains per ear	Weight of one thousand grains (g)	Protein yield (g m <sup>-2</sup> )	Biomass of redroot (g m <sup>-2</sup> )
60	200	30.11 e	421.72 d	365.82 cd	93.97 e	644.25 c
	300	30.25 e	452.75 d	391.36 bc	97.63 e	701.63 bc
	400	32.6 de	609.36 a	429.75 a	116.76 d	712.88 bc
	500	33.18 d	567.59 ab	435.07 a	117.51 d	903.63 a
70	200	35.5 cd	377.24 e	358.25 cd	116.48 d	515.01 e
	300	36.43 cd	386.98 e	372.17 c	119.30 cd	576.03 de
	400	37.51 c	595.76 a	426.24 ab	124.23 cd	612.8 cd
	500	37.63 c	536.21 b	431.03 ab	123.50 cd	793.25 b
80	200	38.27 bc	353.29 f	341.25 de	126.39 bc	418.04 fg
	300	41.18 b	337.63 fg	365.6 c	127.88 bc	550.75 e
	400	42.6 ab	571.42 ab	419.17 ab	141.81 a	593.25 de
	500	43.52 ab	547.15 b	423.51 ab	138.16 ab	641.24 c
90	200	39.25 b	312.56 g	317.04 e	122.25 cd	371.88 g
	300	42.12 ab	349.71 f	329.53 de	124.38 c	487.37 f
	400	44.01 a	516.14 c	354.56 cd	139.07 ab	586.87 de
	500	44.32 a	403.28 d	368.29 c	135.52 ab	625.69 c

Means with different letters indicate a significant difference based on Duncan's test at 5% probability level.

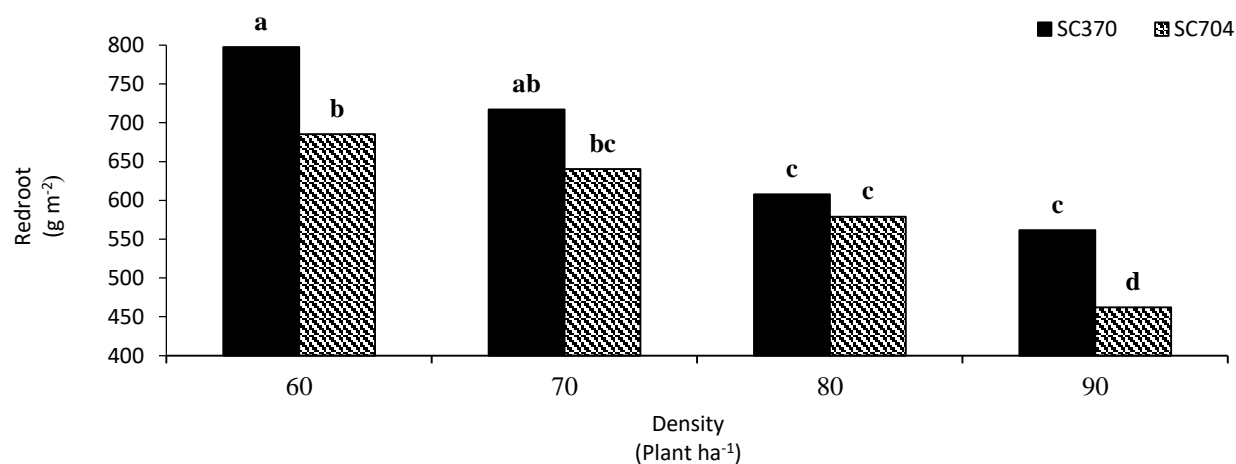


Fig. III. Interaction of cultivar with density on redroot biomass; means with different letters indicate a significant difference based on Duncan's test at 5% probability level.

difference between 400 and 500 kg nitrogen fertilizer. Also, the lowest protein yields, 114.77 and 117.42 g m<sup>-2</sup>, were obtained with application of 300 and 200 kg nitrogen fertilizer, respectively (Table 4). In both corn cultivars 370 and 407, the highest and lowest protein yields were obtained under 80000 and 60000 plant ha<sup>-1</sup> densities,

respectively. Cultivars 704 and 370 showed reduced protein yield under the density of 90,000 plants ha<sup>-1</sup> in comparison with 80,000 plants ha<sup>-1</sup> even though the reduction was not statistically significant (Fig. II). The highest protein yield was obtained under 80000 plant ha<sup>-1</sup> density and application of 400 kg ha<sup>-1</sup> N (Table 5).

### Chlorophyll content

Analysis of variance showed that only the main effects of density and fertilizer were statistically significant on chlorophyll content at 1% probability level (Table 1). In this study, the lowest chlorophyll content belonged to the density level of 90,000 plant ha<sup>-1</sup> (Table 3). Among fertilizer levels, comparison of the mean effects of treatments showed that the highest (76.14 spad) and lowest (63.81 spad) chlorophyll contents belonged to 500 and 200 kg nitrogen fertilizer, respectively (Table 4).

### Redroot biomass

Analysis of variance showed the significant effects of cultivar, density, and fertilizer ( $p \leq 0.01$ ) and the interaction effects of fertilizer and density ( $p \leq 0.05$ ) and also cultivar and density ( $p \leq 0.05$ ) on redroot biomass (Table 1). Moreover, analysis of the effect of corn cultivars on redroot biomass revealed that redroot weeds had higher biomass in cultivation with corn cultivar 370 compared with corn cultivar 704. Cultivar 704 decreased weed biomass by 11.8% compared with cultivar 370 (Table 2).

In this study, redroot biomass decreased with increasing corn density. The lowest redroot biomass (517.95 g m<sup>-2</sup>t) was recorded at the corn plantation density of 90,000 plants ha<sup>-1</sup>. Also, the density of 90,000 plant ha<sup>-1</sup> reduced redroot biomass compared with 60,000, 70,000, and 80,000 plant ha<sup>-1</sup> densities by 30.06, 17.03, and 5.96%, respectively (Table 3). Analyzing the reaction of redroot biomass to nitrogen fertilizer levels was showed that the treatment with 500 kg ha<sup>-1</sup> nitrogen resulted in the highest level of biomass (741.94 g m<sup>-2</sup>) while no significant difference was observed between 400 and 500 kg ha<sup>-1</sup> nitrogen fertilizer (Table 4). Investigation of the interaction effects of plant densities and cultivars on redroot biomass showed that both maize cultivars had the lowest and highest redroot weight at the densities of 90,000 and 60,000 plant ha<sup>-1</sup>, respectively (Fig. III). In all four levels of corn densities, redroot biomass increased by increasing the level of nitrogen fertilizer. The lowest redroot biomass was obtained under the combined treatment of 90,000 corn plants ha<sup>-1</sup> and 200 kg

nitrogen fertilizer while the highest redroot biomass was recorded under 60,000 corn plants ha<sup>-1</sup> in combination with 500 kg ha<sup>-1</sup> N treatment (Table 5).

### Discussion

The number of grains per ear is one of the important components of grain yield in corn, which is affected by plant density (Shi et al., 2016). Quevedo et al. (2015) reported a significant decrease in the number of grains per ear of maize with increasing density. This decrease can be attributed to the decrease in photosynthesis per unit area of leaf (Chen et al., 2017), the decrease in plant growth rate (Monneveux et al., 2005), and the decrease in the penetration of active light in photosynthesis into the canopy (Andrade et al., 2002). Saberali (2008) and Sangoi et al. (2002) stated that increasing the density reduces the space for each plant with the result of less soil volume available to the plant roots and reduced water, nutrients, and growing space for each plant. Also, reducing the space due to increasing density reduces the solar energy absorbed by the plant leaves and ultimately reduces yield. With increasing density and improper distribution of plants, the emergence of silk is delayed compared to the emergence of tassel and as a result, the number of sterile florets increases while the number of grains per ear decreases (Shi et al., 2016).

Balem et al. (2014) reported that the highest number of grains per row of ear was obtained at a density of 55,000 plants per hectare and increasing the density of maize significantly reduced the number of grains per ear. Izadi and Emam (2009) reported that the effect of planting density on the number of grains per ear was significant and the highest number of grains per ear was obtained at a density of 80,000 plant ha<sup>-1</sup>. Nitrogen fertilizers encourage the plant to grow vegetatively, thus increasing ear length and number of grains in ear (Raja, 2001). Prasad and Singh (1999) and Azeez and Adetunji (2007) reported that the increase in nutrients, especially nitrogen, during the emergence of tassel and grain formation, which is the most sensitive stage in nitrogen uptake and photosynthetic material formation, increases the number of grains per ear.



Izadi and Emam (2009) reported that the interaction effect of nitrogen fertilizer with plant density on the number of grains per ear was significant and the highest number of grains per ear (586 grains) was obtained by applying 400 kg ha<sup>-1</sup> in the form of urea and a density of 70,000 plants ha<sup>-1</sup>. Zaman and Khan (2016) stated that nitrogen fertilizer treatments affect the number of ears in maize so that in the nitrogen fertilizer treatment of 400 kg ha<sup>-1</sup> there were two ears per maize, but in the nitrogen fertilizer treatment of 200 kg ha<sup>-1</sup>, there were not two ears per maize. Shi et al. (2016) reported that the effect of plant density on the weight of one thousand maize grains was significant and observed the highest weight of one thousand grains (381.4 grams per square meter) at 55,000 plant ha<sup>-1</sup>. Izadi and Emam (2009) reported that the interaction of density and nitrogen on weight of one thousand maize grains was significant and the highest weight of one thousand grains (414.3 g) was obtained from a combination of 80000 plants ha<sup>-1</sup> and 400 kg nitrogen fertilizer in the form of urea. Increasing plant density for maize can be beneficial to some extent, but higher densities are stressful for maize (Andrade et al., 2002).

Shapiro and Wertman (2006) in a study on the effect of density and planting pattern on leaf area index of maize concluded that increasing the density increased leaf area index and the highest leaf area index was obtained from 75000 plant ha<sup>-1</sup> density. The increase in protein yield due to the application of nitrogen fertilizer in the present study can be attributed to the positive effect of nitrogen on the number of grains per ear and weight of one thousand grains, which is consistent with the research reported by Lindquist et al. (2007). Di paolo and Rinaldi (2008) found that protein yield increased significantly with increasing nitrogen and maximum protein yield of maize was obtained by applying 350 kg ha<sup>-1</sup> N in the form of urea. Positive role of nitrogen fertilizer in increasing protein yield of maize has been confirmed by other researchers (Bruns and Abbas, 2005; Shapiro and Wortman, 2006). Quevedo et al. (2015) reported the highest protein yield of maize under 75,000 plant ha<sup>-1</sup> density.

In the present study, protein yield increased with the application of nitrogen fertilizer, which was attributed to the positive effect of nitrogen on the number of grains per ear and weight of one thousand grains, which is consistent with Di Paolo and Rinaldi (2008). Shapiro and Wortman, (2006) reported that protein yield increased significantly with increasing nitrogen and the maximum corn protein yield was obtained under 225 kg ha<sup>-1</sup> N. Also, the positive role of nitrogen fertilizer in increasing corn protein yield has been confirmed by other researchers (Bruns and Abbas, 2005; Abouzienna et al., 2007). Monneveux et al. (2005) obtained the highest chlorophyll content at combination treatment of 180 kg ha<sup>-1</sup> pure nitrogen and a density level of 60,000 plant ha<sup>-1</sup>. Makarian et al. (2003) and Balm et al. (2014) concluded that by increasing corn density from 1.7 to 5.9 plant m<sup>-2</sup>, weed biomass decreased significantly. The results of studies by Evans et al. (2003) showed that wheat biomass and seed production under corn canopy with a density of 73000 plant ha<sup>-1</sup> decreased significantly. Fertilizers affect both crop growth and weeds (Abouzienna et al., 2007).

The amount of fertilizer, especially nitrogen, affects the number of weed seeds germinated in the soil and weed biomass. As the amount of nitrogen increases, more seedlings will be present and will have a higher dry weight (Barker et al., 2006). Lindquist et al. (2010) stated that weeds use more nutrients than needed and therefore use more fertilizers than crops. Numerous studies have shown that the addition of fertilizer has been more beneficial to weeds. Obviously, crops and weeds show different responses to soil nutrient levels (Blackshaw et al., 2004).

## Conclusion

The highest protein yield was obtained at a density of 80,000 plant ha<sup>-1</sup> and the application of 400 kg ha<sup>-1</sup> N. Also, yield components were significantly higher at 80,000 plant ha<sup>-1</sup> and the application of 400 kg ha<sup>-1</sup> N. The highest leaf area index and chlorophyll content were obtained by application of 500 kg ha<sup>-1</sup> N. The highest leaf area index and chlorophyll content were obtained at 90,000 and

60,000 plant ha<sup>-1</sup>, respectively. In this study, by increasing nitrogen fertilizer at densities of 60,000 and 70,000 plant ha<sup>-1</sup>, weed was able to use fertilizer more efficiently and increase its biomass, but with increasing corn density due to shading and nutrients needed for corn plant growth as a result, the weeds were deprived of light and nitrogen sources for growth and its biomass was reduced significantly. The lowest redroot biomass

was obtained with a corn density of 90,000 plant ha<sup>-1</sup> and application of 200 kg nitrogen. Also, cultivar 704 was more competitive than cultivar 370 and significantly reduced redroot biomass. According to the results of this study, farmers in Miandoab region are recommended to use cultivar 704 with a density of 80,000 plant ha<sup>-1</sup> and application of 400 kg ha<sup>-1</sup> N.

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