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EFFECTS OF PLANT DENSITY AND NITROGEN FERTILIZATION RATE ON FORAGE YIELD AND QUALITY OF SECOND CROP MAIZE (*ZEA MAYS* L.)

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ABSTRACT

The aim of this experiment was to investigate the effects of plant density and nitrogen fertilization rate on dry matter yield and quality of maize planted as a second crop for silage. Field studies were conducted in Bursa with three densities of plants, namely 119,050, 142,850 and 178,570 plants ha^{-1}, and four rates of N fertilizer, 0, 250, 300 and 350 kg N ha^{-1}, that were applied in a split plot design with three replications. Based on a 2-year average, the effect of plant density was significant only for stem diameter. Thus, plant density had no significant effect on plant height, dry matter yield, LAI, SPAD value, crude protein content, and ADF and NDF contents. The maximum plant height, stem diameter, dry matter yield, SPAD value and crude protein content and the minimum ADF content were obtained at 250 kg N ha^{-1} and higher rates of N fertilization. Additionally, the highest LAI was at 300 kg N ha^{-1}. In conclusion, 119,050 plants ha^{-1} and 250 kg N ha^{-1} are recommended for cultivation of second crop silage maize in the southern Marmara Region.

Key words: maize, plant density, nitrogen fertilization rate, SPAD, forage yield, quality.

INTRODUCTION

Maize (*Zea mays* L.) is an important forage crop because the high dry matter yield with favourable quality characteristics is optimal for animal production (Roth et al., 1995). Maize forage yield and quality are influenced by many interacting environmental, cultural and genetic factors. Therefore, the producers of second crop silage maize require more information on the effects of nitrogen fertilization rate and plant density on dry matter yield and forage quality.

Each crop has an optimum plant density. The plant density is of particular importance in maize because it does not have the tillering capability to adjust to variations in the plant stand (Moosavi et al., 2012). Yılmaz et al. (2007) reported that the highest forage and dry matter yields are obtained with 114,000 and 143,000 plants ha^{-1}. Shirkhani et al. (2012) reported that an increase in plant density from 80,000 plants ha^{-1} to 90,000 plants ha^{-1} increases fresh and dry yields; however, with an increase to 100,000 plants ha^{-1}, fresh and dry yields decrease. Moosavi et al. (2012) reported that the leaf area index and total dry yield increase 3.39- and 1.84-fold with an increase in plant density from 50,000 to 140,000 plants ha^{-1}, respectively. One approach to increase the capture of solar radiation within the canopy and the accumulation of dry matter is to increase the LAI by increasing the plant density (Moderras et al., 1998). In a comparison of LAIs, Moosavi et al. (2012) reported that the density of 140,000 plants ha^{-1} has a 3.4-fold higher LAI than that of 50,000 plants ha^{-1}. However, Shirkhani et al. (2012) found that forage quality is reduced at higher plant densities because of increases in the indices of ADF and NDF in the second crop of maize.

Nitrogen fertilization is one of the most important agronomic practices, and many studies have examined the effects of nitrogen fertilizer in primary and second crop maize. The optimum rate of nitrogen fertilizer application for forage maize cultivation depends on many variable factors, such as environmental conditions, management

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systems and genotypes. Nitrogen fertilization of maize influences dry matter yields by affecting the leaf area index, leaf area duration and photosynthetic efficiency (Muchow, 1988; Muchow and Davis, 1988). Tajul et al. (2013) reported that the highest SPAD value was found in a primary maize crop treated with 220 kg N ha^{-1} followed by 180 kg N ha^{-1}. Many other researchers also find positive effects of nitrogen on dry matter yield and forage qualities in primary maize crops (Cox et al., 1993; Mullins et al., 1998; Kara et al., 1999; Cox and Cherney, 2001; Hamid and Nasab, 2001; Patricio Soto et al., 2002, 2004; Keskin et al., 2005; Sahar et al., 2005; Saruhan and Sireli, 2005). However, for the second crop of maize, Karasu et al. (2009) found the maximum dry matter yield at 300 kg N ha^{-1}.

The primary goal of this research was to determine the effects of different plant densities and nitrogen fertilization rates on dry matter yield and forage quality of second crop maize.

MATERIAL AND METHODS

Field studies were conducted during the 2014 and 2015 growing seasons on a clay loam soil at the Agricultural Research and Experiment Center of Uludag University, near Bursa (40°11′ N, 29°04′ E). The soil was a clay (average 58.6% clay content) textural class (Table 1). Soil samples were collected from the 0-0.3 m profile and analyzed. The concentrations of extractable phosphorus (P) and exchangeable potassium (K) were determined using the Olsen method, and the ammonium acetate method, respectively. The soil organic matter was determined using Walkley-Black method. Soil test levels of the experimental area in 2014 and 2015 were: pH 8.33 and 7.09, 1.422 and 1.280% organic matter, 8.44 and 7.18 mg kg^{-1} extractable P, 461.0 and 349.8 mg kg^{-1} exchangeable K and 0.770 and 0.612% $CaCO_3$, respectively (Table 1). The precipitation patterns and amounts differed markedly between the 2014 and 2015 growing seasons (Table 2). The total precipitation from July to October was 234.2, 223.4 and 139.3 mm in 2014 and 2015 and for the long-term (1975-2008), respectively. Of note, 89.3 mm of total precipitation fell after the harvest in October in 2015, and 2015 compared with 2014, was more arid historically. The mean temperatures of the experimental years and those of the long-term record were very similar.

Table 1. Soil characteristics of experimental field in 2014 and 2015

Year	Texture	pH	P (mg kg^{-1})	K (mg kg^{-1})	$CaCO_3$ (%)	Organic matter
2014	Clay	8.33	8.44	461.0	0.770	1.422
2015	Clay	7.09	7.18	349.8	0.612	1.280

Table 2. Precipitation and mean temperature in 2014 and 2015 and for the long-term (1975-2008) in Bursa

Months	Total precipitation (mm)			Mean temperature (°C)		
	2014	2015	Long-term	2014	2015	Long-term
July	4.6	0	18.9	25.55	26.10	24.60
Agust	45.4	6.8	13.8	25.73	26.70	24.30
September	115.6	108.5	39.1	20.61	23.60	20.10
October	68.6	108.1	67.5	16.35	16.08	15.30
Total/mean	234.2	223.4	139.3	22.06	23.12	21.08

The maize variety was Sincero. Three plant densities (119,050, 142,850 and 178,570 plants ha^{-1}) and four nitrogen fertilization rates (0, 250, 300 and 350 kg N ha^{-1}) were evaluated. The experimental design was a randomised complete block in a split plot arrangement with three replications. The main plots were planted to different densities, and the split plots were treated with different rates of nitrogen fertilizer. The split plots were 5 by

3.5 m with 5 rows. The split plots were planted with 0.70-m row spacing. Three-fold seeds for each plant density were sown in the split plots and then hand-thinned to the target plant densities. The maize was sown in July in 2014 and 2015 for the second crop. Half of the nitrogen at each rate with starter amounts of P and K each at 100 kg ha^{-1} was applied before planting. The rest of the nitrogen at each rate was side-dressed when the plants were 40-50 cm in height. Weeds were controlled by a post-emergence application of 2,4-D at a rate of 2.0 l ha^{-1} with mechanical hoeing when required. Immediately before forage harvest, 10 plants from selected rows of each split plot were cut to determine the morphological and physiological parameters of plant height, stem diameter, leaf area index and SPAD value. Leaf area was measured using a digital leaf area meter (LI-3000 Portable Area Meter; LI-COR, Lincoln, NE, USA) to calculate the leaf area index. Leaf chlorophyll content is used as an indirect indicator of crop N status, and chlorophyll meter values (SPAD) were recorded using a portable SPAD meter (Model SPAD-502; Minolta crop, Ramsey, NJ, USA). Of the 10 sampled plants from each split plot, five were separated into stem, leaf and ear fractions to determine the percentages of the whole-plant weight. After eliminating rows for border effects, the two centre rows of each split plot were harvested and the plant fresh-weights were recorded in situ to determine forage yields when the kernel was doughy (11 October 2014 and 18 October 2015). After harvest, 2 plants from the forage material of each split plot were dried at 78 ºC for 48 h, weighed and then ground in a mill to pass through a 1-mm screen. The data obtained from these procedures were used to calculate the dry matter yield of the split plots and to determine the total nitrogen and acid detergent fibre (ADF) and neutral detergent fibre (NDF) contents. A 1.0-g ground sample was used to determine total nitrogen, and 0.5 g was used for ADF and NDF contents. The ADF and NDF were analysed by the sequential detergent analysis method (Van Soest et al., 1991), and the total nitrogen was analysed by the Kjeldahl method. Crude protein content was calculated by multiplying total nitrogen by the constant 5.75. Before analysis of variance, the data for parameters of single years were averaged for both years. Then, the data obtained from measurements or analyses of morphological characters, dry matter yields and forage quality components were subjected to analyses of variance with MINITAB and MSTAT-C programs. To separate the means for plant density, nitrogen fertilization rate and the interaction effects when the F-tests were significant, the LSD test was used.

RESULTS AND DISCUSSION

Based on the analysis of variance, the year effect was significant for plant height, stem diameter, crude protein content, and ADF and NDF contents. For the data combined over the two years and those of the individual years, the plant density and nitrogen fertilization rates significantly affected most parameters. The plant density × nitrogen rate interaction was significant at a 5% level of probability for crude protein in 2015. Additionally, the Y × N interaction was significant for dry matter yield (Tables 3 and 4).

Plant density did not affect plant height, which ranged from 254 to 268 cm in 2014, 271 to 283 cm in 2015 and 263 to 276 for the combined years (Table 3). Others have found similar results for the primary maize crop (Bangarwa et al., 1993; Dogan et al., 1997; Turgut et al., 1997, 2005; Kara et al., 1999; Iptas and Acar, 2006; Azam et al., 2007; Yilmaz et al., 2007; Carpici et al., 2010). However, Tajul et al. (2013) found the maximum plant height at the lowest plant density. The stem diameter decreased significantly with the increase in plant density in 2014 and for the combined years, with the highest values obtained with 119,050 plants ha^{-1} (Table 3). These results are consistent with the findings in other studies of primary maize crops (Turgut et al. (2005), Yilmaz et al. (2007) and Carpici et al. (2010)). Dry matter yield was not affected by plant density in 2014, 2015 or for the combined years, with dry matter yields ranging from 15,010 to 15,870 kg ha^{-1} in 2014, 13,690 to 15,060 kg ha^{-1} in 2015 and 14,350 to 15,470 kg ha^{-1} for the combined years (Table 3). However, Yılmaz et al. (2007), Shirkhani et al. (2012), Moosavi et al. (2012) and Carpici et al. (2010) reported that plant density had a significant effect on dry matter yield in the primary maize crop.

LAI is positively correlated with the yield of a crop. Although plant density had no effect on the LAI in single years and ranged from 5.34 to 6.87 in 2014 and from 5.45 to 6.68 in 2015, for the combined years, the LAI increased significantly with increasing plant density, with the highest value at 178,570 plants ha^{-1} (Table 3). Tajul et al. (2013) reported that LAI decreased with increasing plant density, which is consistent with our results. By contrast, Amanullah et al. (2007) found that the plant density effect is not significant for the LAI, and Baron et al. (2006) reported that LAI increases with an increase in plant density. SPAD values were not affected by plant density in 2014, 2015 or for the combined years (Table 4). Our results are in contrast to those of Tajul et al. (2013), who reported that the SPAD values decrease with an increase in plant density. The effect of plant density on the crude protein content of the second maize crop was not significant in 2014, 2015 or for the combined years (Table 4). Cuomo et al. (1998), Patricio Soto et al. (2002) and Baron et al. (2006) reported results that are similar, but Alexander et al. (1963), Cusicanqui and Lauer (1999) and Widdicombe and Thelen (2002) observed that crude protein contents of forage maize decreased with an increase in plant density in the primary crop of maize. However, Jiwang et al. (2004) reported that crude protein contents increased with an increase in plant density in the primary crop of maize. Plant density had no effect on the ADF content of the second maize crop. Our findings are consistent with those of Iptas and Acar (2006), who reported that plant density had no effect on the ADF content of the primary crop of maize. Plant density had no significant effect on NDF content in our study (Table 4), which is consistent with the findings of Cox and Cherney (2001), Widdicombe and Thelen (2002), and Carpici et al. (2010) for the primary maize crop In Shirkhani et al. (2012), the NDF content increased in the second maize crop with increases in plant density. Similarly, Stanton et al. (2007) observed that forage quality increased with increases in plant density. In Baron et al. (2006), plant density affected NDF content but not ADF content, and the NDF content was significantly lower at 75,000 than 125,000 plants ha^{-1}, with values intermediate at 100,000 plants ha^{-1}.

All parameters increased significantly with the increase in nitrogen fertilization rate, with the exception of ADF and NDF contents (Tables 3 and 4). The maximum plant height and stem diameter were obtained at rates of 250-350 kg N ha^{-1} (Table 3). The plants were taller in 2015 than in 2014, whereas the diameter of the stems was wider in 2014 than 2015. Our findings are consistent with those of Keskin et al. (2005), Sahar et al. (2005) and Safdarian et al. (2014), who reported that the height of plants increased with increases in nitrogen fertilization rates in primary maize crops. However, Karasu et al. (2009) reported that plant height and stem diameter of the second maize crop were not affected by the rate of nitrogen fertilizer. Nitrogen fertilization had a significant effect on dry matter yield; as nitrogen fertilization rates increased, the dry matter yield increased and reached a peak value at 250 kg N ha^{-1}, with no further increase at the higher N fertilization rates in 2014 and for the combined years. However, in 2015, the maximum dry matter yield was obtained at 350 kg N ha^{-1}. In Karasu et al. (2009), the maximum dry matter yield for the second crop of maize was obtained at 300 kg N ha^{-1}. Both Valadabadi and Farahani (2010) and Safdarian et al. (2014) reported that nitrogenous fertilizers increased the total dry matter yield of maize. The LAI response to nitrogen fertilization was statistically significant, and LAI increased up to 300 kg N ha^{-1} and then decreased at 350 kg N ha^{-1} in 2014 and for the combined years. However, in 2015, the LAI increased up to 250 kg N ha^{-1}, with no further change at 300 kg N ha^{-1} (Table 3). Similar results were reported by Valadabadi and Farahani (2010), Hammad et al. (2011) and Safdarian et al. (2014); however, Amanullah et al. (2007) reported results that are in contrast to those of this study. As the rate of nitrogen fertilization increased, the SPAD values also increased and reached a peak value at 250 kg N ha^{-1}; the SPAD values did not change with further nitrogen additions in 2014 and 2015 and for the combined years (Table 4). Tajul et al. (2013) reported that SPAD values increased with increases in the nitrogen fertilization rate, with the highest SPAD value in the plants treated with 220 kg N ha^{-1}. Nitrogen fertilization had a significant effect on crude protein content, with the highest crude protein contents obtained at 250, 300 and 350 kg N ha^{-1} in 2014 and 2015 and for the combined years (Table 3). These results are consistent with those of some other workers (Patricio Soto et al., 2002; Patricio Soto et al., 2004, Carpici et al., 2010; Safdarian et al., 2014). The rates of nitrogen fertilization had no effect on the ADF content in 2014 but had a significant effect in 2015 and for the combined years. Nitrogen fertilization decreased the ADF concentration in the second maize crop (Table 4), which is a finding that is consistent with that of Keskin et al. (2005) and Safdarian et al. (2014), who reported that the ADF contents decreased at higher nitrogen fertilization rates. The rate of nitrogen fertilization had no effect on the NDF content in 2014 and 2015 and for the combined years. However, others have reported results that are in contrast to ours (Carpici et al., 2010; Safdarian et al., 2014).

Table 3. Result of variance analysis and mean effects of plant density (PD) and nitrogen fertilization rate (N) on plant height, stem diameter, dry matter yield and leaf area index (LAI) of second crop maize in 2014, 2015 and for the combined years

Treatment	Plant height			Stem diameter			Dry matter yield			Leaf area index		
	2014	2015	Combined years	2014	2015	Combined years	2014	2015	Combined years	2014	2015	Combined years
Plant density (plants ha^{-1})												
119.050	268	283	276	19.6a	17.9	18.8a	1518	1420	1469	5.34	5.66	5.51b
142.850	262	273	268	18.3b	17.1	17.7b	1501	1369	1435	5.66	5.45	5.55b
178.570	254	271	263	16.7c^{x}	16.4	16.6c	1587	1506	1547	6.87	6.68	6.81a
Nitrogen rate (kg ha^{-1})												
0	217b	257b	237b	16.1b	16.4b	16.3b	817b	859c	838b	4.49c	4.60b	4.60c
250	274a	282a	278a	18.6a	17.2ab	17.9a	1759a	1514b	1637a	6.21b	6.27a	6.24b
300	278a	286a	282a	19.0a	17.3ab	18.1a	1828a	1621ab	1725a	6.92a	6.60a	6.76a
350	276a	278a	277a	19.1a	17.6a	18.4a	1738a	1732a	1735a	6.20b	6.25a	6.23b
Mean (year)	262B	276A		18.2A	17.1B		1536	1432		5.95	5.96	
F value significance												
Year (Y)	-	-	**	-	-	**	-	-	ns	-	-	ns
Blocks (Year) (B)	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
PD	ns	ns	ns	**	ns	**	ns	ns	ns	ns	ns	*
PDxY	-	-	ns	-	-	*	-	-	ns	-	-	ns
N	**	**	ns	**	*	**	**	**	**	**	**	**
PDxN	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
YxN	-	-	**	-	-	**	-	-	*	-	-	ns
YxPDx N	-	-	ns	-	-	ns	-	-	ns	-	-	ns

Notes. x – means in the same location followed by the same letter were not significantly different at 0.05 level using LSD test.
*, ** – *F*-test significant at p>0.05, and p>0.01, respectively, ns – not significant.

Table 4. Result of variance analysis and mean effects of plant density (PD) and nitrogen fertilization rate (N) on SPAD value, crude protein, acid detergent fibre (ADF) and neutral detergent fibre (NDF) of second crop maize in 2014, 2015 and fort he combined years

Treatment	SPAD value			Crude protein			Acid detergent fibre			Neutral detergent fibre		
	2014	2015	Combined years	2014	2015	Combined years	2014	2015	Combined years	2014	2015	Combined years
Plant density (plant ha^{-1})												
119.050	48.8	49.0	48.9	6.1	7.6	6.9	25.0	21.7	23.3	48.1	411	44.6
142.850	43.8	46.6	45.1	6.3	7.6	6.9	25.1	20.5	22.8	48.1	40.9	44.5
178.570	45.5	44.9	45.2	6.5	7.0	6.7	26.8	22.9	24.9	49.6	41.0	45.3
Nitrogen rate (kg ha^{-1})												
0	37.5b	36.7b	37.1b	4.7b	6.7b	5.7b	27.7	25.1a	26.4a	50.6	42.2	46.4
250	47.9a	49.1a	48.5a	6.8a	7.4ab	7.1a	25.0	20.8b	22.9b	47.4	41.4	44.4
300	49.3a	49.8a	49.6a	6.7a	7.5ab	7.1a	25.0	20.6b	22.8b	47.9	39.6	43.7
350	49.5a	51.6a	50.5a	7.0a	7.9a	7.4a	24.7	20.2b	22.5b	48.5	40.7	44.6
Mean (year)	46.0	46.8		6.3B	7.4A		25.6A	21.7B		48.6A	41.0B	
F value significance												
Year (Y)	-	-	ns	-	-	**	-	-	**	-	-	**
Blocks (Year) (B)	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
PD	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
PDxY	-	-	ns	-	-	ns	-	-	ns	-	-	ns
N	**	**	**	**	*	**	ns	**	**	ns	ns	ns
PDxN	ns	ns	ns	ns	*	ns	ns	ns	ns	ns	ns	ns
YxN	-	-	ns	-	-	ns	-	-	ns	-	-	ns
YxPDx N	-	-	ns	-	-	ns	-	-	ns	-	-	ns

Notes. x – means in the same location followed by the same letter were not significantly different at 0.05 level using LSD test. *, ** – *F*-test significant at p>0.05, and p>0.01, respectively, ns – not significant.

CONCLUSION

Forage maize is a crop that is highly responsive to plant density and the rate of nitrogen fertilization. Based on our findings, plant density had no significant effect on plant height, dry matter yield, LAI, SPAD value, crude protein content, and ADF and NDF contents. The maximum values for plant height, stem diameter, dry matter yield, SPAD, and crude protein content and the minimum value for ADF content were obtained at 250 kg N ha^{-1} and the higher N fertilization rates. The maximum LAI was obtained at 300 kg N ha^{-1}. Therefore, 119,050 plants ha^{-1} and 250 kg N ha^{-1} are recommended for cultivation of second crop silage maize.

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