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Evaluation of yield and some physiological traits of forage corn affected by chemical and biological nitrogen fertilizers intercropped with sweet basil

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Abstract

In order to evaluate yield and some physiological traits of forage corn under nitrogen fertilizers (biological, chemical and integrated) in additive intercropping with basil a field experiment was carried out in the Experimental Farm of Faculty of Agriculture, Lorestan University during 2014-2015 growing seasons. Treatments were arranged in a factorial experiment based on randomized complete blocks design with three replications. Experimental treatments were 100% chemical fertilizer (N), bio-fertilizer (nitroxin), integration of bio-fertilizer + 50% chemical fertilizer and control in different intercropping systems consisted of sole cropping corn and the additive intercropping of corn + 25% sweet basil, corn + 50% sweet basil, corn + 75% sweet basil and corn + 100% sweet basil. The results showed that integration of bio-fertilizer + 50% chemical fertilizer had the highest number of green leaves per plant (11.72) and leaf area index (LAI) (3.75) and there was no significant difference between this treatment and using 100% chemical fertilizer (N) in plant height, stem dry weight, chlorophyll a, chlorophyll b and carotenoids contents. Among different intercropping systems the highest plant height (179.25 cm), number of green leaves per plant (11.4), leaf dry weight (5.64 ton*ha^{-1}), ear dry weight (7.19 ton*ha^{-1}), stem dry weight (6.11 ton*ha^{-1}), total dry weight (19.22 ton*ha^{-1}), chlorophyll a (0.62 mg*g^{-1} FW), chlorophyll b (0.42 mg*g^{-1} FW), and total chlorophyll (1.04 mg*g^{-1} FW) were obtained from sole cropping pattern. However, sole cropping pattern in terms of mentioned traits except for number of green leaves per plant, ear dry weight and total dry weight had no significant difference with corn + 25% sweet basil and corn + 50% sweet basil treatments The results showed that integration of bio-fertilizer + 50% chemical fertilizer could be considered as an approach to reduce the consumption of chemical fertilizers for sustainable agriculture

Keywords: additive intercropping, chlorophyll, dry forage yield, forage corn, nitroxin

Introduction

Maize is an important cereal and forage crop and dedicated the third rank after wheat and rice. In developing countries, maize is a major source of income for many farmers (Tagne et al., 2008). Maize can be used as food for people, livestock feed, poultry feed and also in Industry (Bibi et al., 2010). With the growth population increase, the request for food has been increasing while land availability has been declining. Thus, the only way to increase production is to increase yield per unit area (Hirpa, 2014). Including ways to increase the yield per unit area can be mentioned to appropriate management in crops nutrition and intercropping systems plants.

Increasing nitrogen fertilizer applications has been a major management strategy to obtain high yield (Guo et al., 2010). However, nitrogen over plus combined with low nitrogen use efficiency have had negative subsequences to the environment such as soil acidification, environmental pollution and decreased soil microbial activity (Guo et al., 2010; Chen et al., 2014). Thus, the reduction of nitrogen fertilizer inputs and improved nitrogen use efficiency are crucial for sustainable production. One alternative to decrease the application of nitrogen fertilizer could be the use of free-living N-fixing bacteria in agricultural systems (Cocking, 2003), which could improve crop production, reduce the overuse of chemical fertilizers, and decrease greenhouse emissions (Kennedy et al., 2004). Inoculation of maize with free living N-fixing bacteria has been shown to raise crop yields through supplemental nitrogen input to the soil (Dobbelaere et al., 2002; Hungria et al., 2010). Bacteria from *Azotobacter* and *Azospirillum* groups have the ability to make and leak some biological material such as vitamin B, Nicotinic acid, biotin, oxins, gebrelins etc in plant's root environment which have an effective and beneficial role in increment of root's absorbance (Kader, 2002).

In conventional agriculture, one of the main procedures to increase livestock production is to use diet containing some cereal seeds like barley (*Hordeum vulgare* L.) and corn, or to use silage plants as well as many types of concentrates having food complementariness and chemical matters in line with stimulating animal's growth and taking precaution to catch diseases. The imprecise use of these compounds causes to break out many metabolic diseases like acidosis and liver abscesses in ruminants and ultimately leads to create many problems (Magkoric and Samard, 2005). In recent decades, these problems have directed increasingly researchers' attentions to produce organic crops and by-products free of any chemical residuals previously used (Koocheki and Khajeh Hosseini, 2008). In many criteria of organic agriculture used by some countries, it has been placed an emphasis on availability of some medicinal species like caraway (*Carum carvi* L.) and chicory (*Cichorium intybus* L.) on the pasture free grazed by livestock for meeting naturally their medicinal requirements (Smith and Brimer, 2005).

In this regard, the new idea so-called "medicinal forage" was introduced. From forage value's standpoint, although some annual or biennial medicinal plants solitarily have not attracted attentions to themselves, due to some problems such as forage unpalatability or possessing some anti-nourishment compounds (like prussic acid, coumarin, and phenolic compounds), intercropping these plants with conventional forage plants gives them high credit as medicinal forage through running an efficient and scientific management. Medicinal forage plants ensure many ecological and bioenvironmental advantages. Hence, one of the methods employed for producing medicine forage is to intercrop useful medicinal plants with predominant forage plants (Safikhani et al., 2014). Intercropping is an old and widespread practice used in many areas of the world (Lithourgidis et al., 2011) that may provide a balanced diet, reduce labor peaks, minimize adverse effects of biotic and abiotic factors, protect soil against erosion, improve the use of limited resources, increase stability of yield and provide higher outputs (Agegnehu et al., 2008). Girma (2015) reported that biological yield of corn was significantly affected by different intercropping systems and compared to sole maize decreased biological yield. Little information is available on the suitability of aromatic crops especially sweet basil as intercrops in maize in Iran, therefor this study was carried out in order to evaluate yield and some physiological traits of forage corn under nitrogen fertilizers (biological, chemical and integrated) in additive intercropping with basil.

Materials and methods

Location and plant materials

This study was conducted in the Experimental Farm Department of Agronomy, Faculty of Agriculture, Lorestan University, Iran (33°29′N, 48°22′E, and altitude 1,125 m), during 2014-2015 growing seasons, Physical soil analysis and chemical characteristics in soil depth 0-40 cm have been shown in Table 1. Different intercropping systems were including: corn sole cropping (10 plants m^{-2}) and the additive intercropping of sweet basil + corn (20 + 10, 50 + 10, 75 + 10 and 100 + 10 plants m^{-2}). Seed of crops were planted in plots with an area of 5 m^2 included five rows with 2 meters long and 50 cm apart, at the same day on early May, 2014 and 2015. The intra-row plant spacing for maize (*Zea mays* L. cv. S.C. 704) was 20 cm and for different ratio of sweet basil were 2.5, 3.3, 5 and10 cm, respectively All plots were irrigated immediately after sowing. Subsequent irrigations were carried out every 5 days. Hand weeding of the experimental area was performed as required.

Table 1. Physical and chemical analysis of soil before the experiment

	Soil texture	Clay (%)	Silt (%)	Sand (%)	pH	EC ($dS*m^{-1}$)	Total N (%)	P (ppm)	K (ppm)
Year 1	Clay loam	32.16	42	25.84	7.17	0.459	0.302	8	390
Year 2	Clay loam	31.52	41.5	26.98	7.36	0.536	0.285	6	356

Fertilizer and microbial inocula

Before cultivation, all the P and K fertilizers added to plots according to soil test. Nitrogen (as urea) in 100% chemical fertilizer treatments was applied on bases of 375 $Kg*ha^{-1}$. Half the amount of nitrogen in the tests added to 100% chemical fertilizer and integrated of biofertilizer + 50% chemical fertilizer plots with last plowing before planting. The rest of the nitrogen in two phases was distributed: an eight-leaf stage of corn and remain of that before the start of crown flower as head in the plots. Urea was used as source of inorganic (N) fertilizer. Corn seeds were inoculated with biofertilizer and kept for half an hour in shade. The biofertilizers were *Azospirillum* and *Azotobacter*. The both of *Azospirillum* and *Azotobacter* consist of 10^8 $CFU*ml^{-1}$ (colony forming $units*ml^{-1}$) inoculant. The inocula of the two rhizobacteria species, *Azotobacter sp*. *Azospirillum sp*, were purchased from Mehr Asia Biotechnology Company, Tehran, Iran.

Plot experiment

This experimental design was a factorial experiment based on Randomized Complete Blocks Design (RCBD) with three replications. Experimental treatments included; F1: (control); F2: 100% chemical fertilizer (N); F3: bio-fertilizer (nitroxin); F4: (integration of bio-fertilizer (nitroxin) + 50% chemical fertilizer) and different intercropping systems consisted of sole cropping corn (I1) and the additive intercropping of corn + 25% sweet basil (I2), corn + 50% sweet basil (I3), corn + 75% sweet basil (I4) and corn + 100% sweet basil (I5).

Traits measurement

The traits in this study included plant height , number of green leaves per plant, leaf dry weight, ear dry weight, stem dry weight, total dry weight, Chlorophyll a, chlorophyll b, total chlorophyll, carotenoids and LAI. Five plants from each plot were selected at random and traits including: plant height, number of green leaves per plant and LAI were measured and average was calculated from those five measured values. Maize harvested as forage on milking stage of grain. The samples were divided to stem, leaves and ear, then, the fresh weight was measured. The samples were oven dried at 75 ˚C for 72 h and weighed.

Measurement of chlorophyll A, chlorophyll B and carotenoids

Chlorophyll a, chlorophyll b and carotenoids were extracted with pure acetone from fresh leaf samples (0.1 g). Absorbance was measured at 663 nm, 645 nm and 470 nm for chlorophyll a, chlorophyll b and carotenoids, respectively, using a UV spectrophotometer (UNICO, Model SUV–S2100, USA). The mentioned traits were calculated according to the procedure described by Lichtenthaler (1987).

Chlorophyll a = 11.24 * A 662 - 2.04 * A 645

Chlorophyll b = 20.13 * A 645 - 4.19 * A 662

Total chlorophyll = 7.05 * A 662 + 18.09 * A 645

Carotenoids = (1000 * A 470 – 1.90 chlorophyll a – 63.14 chlorophyll b) /214

Data analysis

SAS (version 9.1) and MSTATC programs were used to conduct an analysis of variance (ANOVA) and means comparison, respectively. Treatment mean differences were separated by Duncan test at the 5% level of probability

Results and discussion

Physiological traits of maize

The result of mean comparisons showed that the highest chlorophyll b, total chlorophyll, and carotenoids were achieved in second year (Table 2). The effects of different intercropping systems were significant on chlorophyll a, chlorophyll b, total chlorophyll and LAI. The data presented in Table 2 revealed that among the different intercropping systems the lowest chlorophyll a, chlorophyll b, Total chlorophyll and LAI were obtained from corn + 100% sweet basil pattern. Sole cropping pattern in terms of mentioned traits had no significant difference with corn + 25% sweet basil, corn + 50% sweet basil and corn + 75% sweet basil treatments.

Increases in density of sweet basil plants from 60 to 80 in intercropping with corn was caused a significant decrease in chlorophyll a, chlorophyll b, total chlorophyll and LAI of maize. The significant reduction in this traits in corn + 100% sweet basil pattern could be associated with higher competition for soil resources or allelopathic effect as basil population increased in intercrop with maize (Olowe and Adeyemo, 2009). Shobeiri et al.

(2015) reported that the amount of chlorophyll a and b in grass pea (*Lathyrus sativus* L.) intercropped with barely was significantly lower than that of in its monoculture. Similarly, the reductions of maize LAI under intercropping systems have been also reported by other researchers (Zegada-Lizarazu et al., 2006; Thobatsi, 2009). It seems that the main reason of decline in LAI in corn + 100% sweet basil pattern compared to other intercropping systems is associated with the interspecies competition mechanism by which the extent of leaf expansion and duration will be restricted.

Table 2. The effect of different source of nitrogen on some physiological traits of forage corn under different intercropping systems during 2014-2015 growing seasons

Treatment	Chlorophyll a ($mg*g^{-1}$ FW)	Chlorophyll b ($mg*g^{-1}$ FW)	Total chlorophyll ($mg*g^{-1}$ FW)	Carotenoids ($mg*g^{-1}$ FW)	Leaf area index (LAI)
Year 1	0.61[a]	0.4[b]	1.01[b]	0.11[b]	3.22[a]
Year 2	0.61[a]	0.42[a]	1.04[a]	0.15[a]	3.31[a]
I1	0.62[a]	0.42[a]	1.04[a]	0.12[a]	3.33[a]
I2	0.62[a]	0.42[a]	1.04[a]	0.13[a]	3.33[a]
I3	0.61[a]	0.42[a]	1.03[a]	0.13[a]	3.32[a]
I4	0.61[a]	0.41[a]	1.03[a]	0.13[a]	3.22[ab]
I5	0.6[b]	0.38[b]	0.98[b]	0.13[a]	3.12[b]
F1	0.57[b]	0.29[c]	0.86[c]	0.13[a]	2.69[c]
F2	0.63[a]	0.47[a]	1.1[a]	0.13[a]	3.68[a]
F3	0.62[a]	0.44[b]	1.06[b]	0.13[a]	2.94[b]
F4	0.63[a]	0.45[ab]	1.08[b]	0.13[a]	3.75[a]

I1, I2, I3, I4 and I5: sole cropping corn, the additive intercropping of corn + 25% sweet basil, corn + 50% sweet basil, corn + 75% sweet basil and corn + 100% sweet basil, respectively. F1: (control), F2: 100% chemical fertilizer (N), F3: bio-fertilizer (nitroxin), F4: integration of bio-fertilizer (nitroxin) + 50% chemical fertilizer. The means in each column having the same letter are not significantly different at 5% Level of probability based on Duncan test.

The effects of different fertilizer treatments were significant on chlorophyll a, chlorophyll b, total chlorophyll and LAI. The data presented in Table 2 showed that among the different fertilizer treatments the highest chlorophyll a, chlorophyll b and LAI values shown by treatments F2 and F4 treatments and there was no significant difference between integration of bio-fertilizer + 50% chemical treatment and 100% chemical fertilizer, that indicating the effective role of nitrogen-fixing bacteria on meeting the plants nutrient demands in term of nitrogen, and so nitroxin biofertilizer treatment had better effect on the majority of traits compared to control group.

Application of nitroxin biofertilizers (F3) resulted in 9, 51, 23 and 9% extra chlorophyll a, chlorophyll b, total chlorophyll and LAI of maize, respectively, over their corresponding traits in the control (F1). High nitrogen fertilization can increase photosynthesis and plant growth on several crops (Evans, 1989; Cechin and Fumis, 2004). Up to 75% of leaf N found in chloroplasts and most of it is part of ribulose bisphosphate carboxylase (Rubisco) alone. Under low nitrogen level the lower photosynthesis is often attributed to the reduction in chlorophyll content and Rubisco activity (Evans and Terashima, 1987; Fredeen et al., 1991).

Christos et al. (2007) reported that chlorophyll content was affected by the nitrogen treatments and it was higher at both nitrogen levels compared with the control. Useful effects of bacteria inoculation on increased chlorophyll content are due to higher availability of nitrogen to the growing tissue and organs supplied by free living N-fixing bacteria (Chandrasekhar et al., 2005). Rahi (2013) showed that bio-fertilizer (nitroxin) increased chlorophylls a, b, total Carotenoids and anthocyanin content of the plants linearly. No significant different for carotenoid was observed between the control and other fertilizer treatments Smolen and Sady (2009) showed that Nitrogen fertilization did not cause any significant changes in carotenoid compounds in carrot. The interaction effect of year and fertilizer on the amount of carotenoids was significant. The highest amount of carotenoids was obtained from second year and F4 treatment (Figure 1).

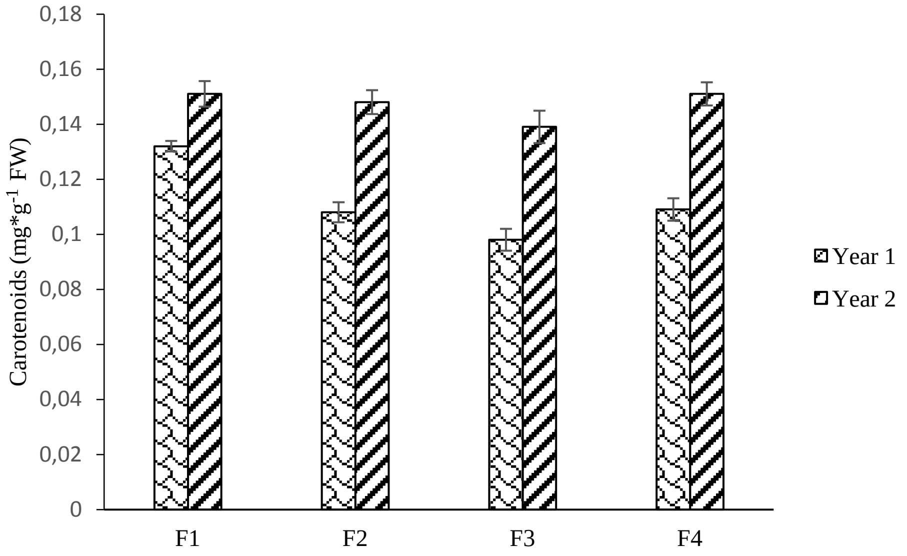

Figure 1. The effects of different fertilizer treatments on carotenoids during two growing seasons. F1: (control); F2: 100% chemical fertilizer (N); F3: bio-fertilizer (nitroxin); F4: (integration of bio-fertilizer (nitroxin) + 50% chemical fertilizer)

Yield and morphological traits of maize

The result of mean comparisons revealed that the highest plant height, number of green leaves per plant, ear dry weight, stem dry weight and total dry weight were achieved in second year (Table 3). The effect of two growing seasons on these traits was not the same and the difference between the growing seasons was significantly observed, and it is apparently contributed to the appropriateness of climate conditions especially in the early of seasonal growth in the second year. This condition, through providing suitable seedlings establishment, suitable photosynthesis, and identical growth, paved the way for having useful field with high yield.

The effects of different intercropping systems were significant on plant height, number of green leaves per plant, leaf dry weight, ear dry weight, stem dry weight and total dry weight. The data presented in Table 3 showed that among the different intercropping systems the highest plant height, number of green leaf per plant, leaf dry weight, ear dry weight, stem dry weight and total dry weight were obtained from sole cropping pattern. Among the different intercropping systems, sole cropping pattern in terms of plant height had no significant difference with corn + 25% sweet basil and corn + 50% sweet basil treatments. Due to high density of sweet basil intercropped with corn and its competition with corn over water and nutrient uptake, the height of corn in corn + 100% sweet basil pattern was apparently decreased as compared to corn's monoculture. Javanmard et al. (2013) reported that the height of corn intercropped with some legumes like hairy vetch (*Vicia villosa*) and bitter vetch (*Vicia ervilia* L.) was significantly decreased.

The reason for decreasing the number of green leaves in the treatment of corn + 100% sweet basil pattern is related to their increasing competition for receiving nutrient materials especially nitrogen. Under nitrogen deficit, during the remobilization mechanism it is transferred from aged leaves located on the lower parts of a plant into young ones, and this factor causes to intensify the ageing and dryness of aged leaves located on the lower parts of a plant. The results of this research showed that corn's leaf dry weight (LDW) was only affected while intercropped with highest density of sweet basil, and LDW, as compared to other traits, had the least sensitivity to this condition. Considering the importance of corn leaves comparing to its stem, the more leaves are produced, the more the silage and forage palatability are increased. This trait is usually directed the farmers attentions to itself. Also, there was no significant difference between corn sole cropping and corn + 25% sweet basil treatment in terms of the ear dry weight (EDW) trait, but it was decreased in other intercropping treatments whenever the density of sweet basil was increased. It seems that whenever the density of sweet basil goes up, the rate of interspecific competition over nutrient uptake also elevates. This factor culminates in reduction of nutrient resources and followed by lessening leaf area and other effective parameters influencing on photosynthesis. Finally, it causes to diminish the amount of plant's assimilation and yield. Pilbem et al. (1994) found that the rate of corn's grain yield under monoculture condition was higher than that of in intercropping with legume. In this regard, in intercropping corn with cucumber (*Cucumis sativus* L.) whenever the density of cucumber goes up simultaneously led to reduce the corn's harvesting index (HI) (Ghanbari et al., 2006).

Among corn's traits investigated, stem dry weight (SDW) followed by LDW had the least sensitivity to high density of sweet basil. Concerning SDW trait, the difference between sole cropping, corn + 25% sweet basil, and corn + 50% sweet basil treatments was not significantly observed whereas a significant reduction between corn + 75% sweet basil and corn + 100% sweet basil was obtained compared to other treatments (Table 3). As comparing to monoculture condition, a significant reduction in the dry matter accumulation of wheat intercropped with corn has also been reported (Nasirimahlati et al., 2011).

The highest total dry weight was obtained from sole cropping pattern and corn + 25% sweet basil pattern (Table 3). Best utilization of nutrients, moisture, space and solar energy can be derived through sole cropping system (Aiyer, 1963). Bagheri et al. (2014) reported that fresh and dry weight of leaves, stem, and ear in sole culture of maize were more than intercrops. Increasing the proportions of sweet basil and/or borage decreased the weight of leaves, stem, and ear of maize. Higher yield of sole culture compared to intercrop may be due to minimal disruption of the plants habitat (Banik et al., 2006).

Table 3. The effect of different source of nitrogen on yield, yield components and some morphological traits of forage corn under different intercropping systems during 2014-2015 growing seasons

Treatment	Plant height (cm)	Number of green leaf per plant	Leaf dry weight ($ton*ha^{-1}$)	Ear dry weight ($ton*ha^{-1}$)	Stem dry weight ($ton*ha^{-1}$)	Total dry weight ($ton*ha^{-1}$)
Year 1	175.67[b]	10.05[b]	5.29[a]	6.58[b]	5.66[b]	17.76[b]
Year 2	177.31[a]	11.86[a]	5.61[a]	7.18[a]	6.08[a]	19.15[a]
I1	179.25[a]	11.4[a]	5.64[a]	7.19[a]	6.11[a]	19.22[a]
I2	178.38[a]	10.89[bc]	5.63[a]	7.03[ab]	6.08[a]	19[ab]
I3	177.96[a]	11.08[b]	5.49[a]	7[b]	5.95[a]	18.69[b]
I4	174.37[b]	10.84[c]	5.39[a]	6.76[c]	5.77[b]	18.17[c]
I5	172.49[c]	10.56[d]	5.1[b]	6.43[d]	5.45[c]	17.19[d]
F1	162.37[c]	9.93[d]	4.62[d]	6.19[c]	5.06[c]	16.1[d]
F2	185.53[a]	11.5[b]	6.22[a]	7.26[a]	6.43[a]	20.17[a]
F3	173.46[b]	10.67[c]	4.99[c]	6.99[b]	5.71[b]	17.96[c]
F4	184.60[a]	11.72[a]	5.98[b]	7.08[b]	6.28[a]	19.59[b]

I1, I2, I3, I4 and I5: sole cropping corn, the additive intercropping of corn + 25% sweet basil, corn + 50% sweet basil, corn + 75% sweet basil and corn + 100% sweet basil, respectively. F1: (control), F2: 100% chemical fertilizer (N), F3: bio-fertilizer (nitroxin), F4: integration of bio-fertilizer (nitroxin) + 50% chemical fertilizer. The means in each column having the same letter are not significantly different at 5% Level of probability based on Duncan test.

The effects of different fertilizer treatment were significant on plant height, number of green leaves per plant, leaf dry weight, ear dry weight, stem dry weight and total dry weight. The lowest plant height was achieved from F1 (control) treatment (Table 3). As the nutrient deficit plays a major role in the height of plants, it seems that, due to the deficit of nutritional matters, the control plants grew less than those were nourished properly through chemical and integrated fertilizers.

The highest number of green leaves per plant was achieved from F4 treatment. It seems that the reason for supremacy of F4 treatment over other fertilizer treatments in terms of the number of green leaves per plant is contributed to the proved effects of nitrogen-fixing bacteria, by which the premature ageing of leaves, through enhancing the nutritional uptake and nitrogen content of tissues, will be inhibited and consequently the number of dried leaves is significantly reduced (Bashan et al., 2004). By two-year planting two types of corn seeds in the field (i.e. the inoculated seeds with free living N-fixing bacteria and non-inoculated seeds as control group) under different treatments of consumption and non-consumption of nitrogen, Rohitashav et al. (1993) found that the yield of dry matter of forage and the ratio of green leaves per plant were higher due to inoculation benefits.

The comparison of the mean values of the leaf dry weight showed that among the different fertilizer treatments the highest and lowest leaf dry weight value shown by treatments F2 and F1 treatment, respectively (Table 3). This factor may be ascribed to the effect of nitrogen on the expanding of leaf area and continuance (Smiciklas and Below, 1990) and consequently improving photosynthesis resources and corn yield (Pandey et al., 2000). In this respect, Hernandez et al. (1995) also reported that inoculating seeds with nitrogen-fixing bacteria enhanced the fresh weight of aerial parts of plant and the number of leaves per plant.

Among the different fertilizer treatments the highest and lowest ear dry weight was obtained by treatments F2 and F1 treatment, respectively (Table 3). An increase in nitrogen leads to increase the leaf area, leaf area index and duration of leaf area, and this increase is stemmed from higher rate of assimilation and yield (Malakouti, 1998). Regarding the role of nitrogen deficit in increasing time span between releasing pollen and the emergence of corn silk, this agent is one of the main reasons of causing a sever reduction in ear weight and grain yield of control group (without applying fertilizers) owing to sterilized ears of corn.

In control group, stem dry weight (SDW) was significantly decreased in comparing to other fertilizer treatments apparently due to reduction in cell division and cell length and followed by reduction in internode length and stem height under nitrogen deficit condition. Molla Hoseini et al. (2006) reported that using 30% nitrogen more than that of recommended based on soil test increased the rate of total dry yield, stem, ear, and leaf.

The comparison of the mean values of the total dry weight showed that the highest and lowest total dry weight value shown by treatments F2 and F1 treatment, respectively (Table 3). Application of nitroxin biofertilizer (F3) resulted in 12% extra total dry weight of maize, over their corresponding trait in the control (F1). It seems that application nitrogen fertilizer plays a significant role in increasing plant’s vegetative growth and consequently paves the way for increasing the yield of forage corn. An increase in the rate of applied nitrogen and also its effect on physiological processes resulted in more photosynthesis, assimilation, dry matter and yield. In other words, due to nitrogen-fixing and the effects of nitrogen-fixing bacteria on secreting growth regulator as well as stimulating plant’s growth, the forage corn’s yield was increased through application nitroxin biofertilizer as compared to control group. It was also proved the positive role of nitrogen on qualitative and quantitative traits of corn such as dry weight (Cox et al., 1993). In this respect, Nanda et al. (1995) also showed that inoculating corn’s seeds with nitrogen-fixing bacteria under field conditions caused to increase complete soiling yield through treating plants with different levels of nitrogen fertilizer. The interaction effect of year and fertilizer on the amount of number of green leaves per plant was significant. The highest number of green leaves per plant was achieved from second year and F2 treatment (Figure 2). Also, the interaction between intercropping and fertilizer on plant height was significant. The sole cropping had the highest amount of plant height with F2 treatment application (Figure 3).

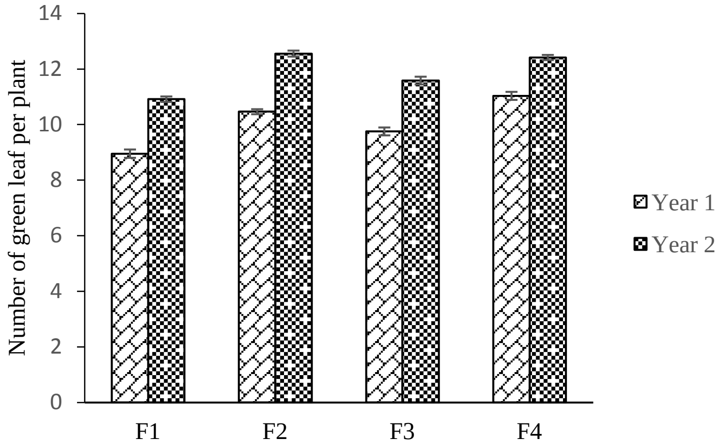

Figure 2. The effects of different fertilizer treatments on number of green leaves per plant during two growing seasons. F1: (control); F2: 100% chemical fertilizer (N); F3: bio-fertilizer (nitroxin); F4: (integration of bio-fertilizer (nitroxin) + 50% chemical fertilizer)

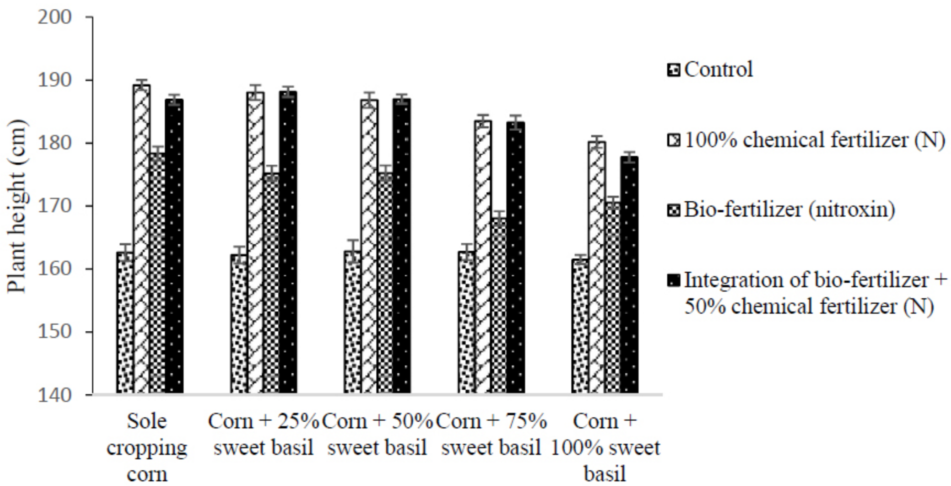

Figure 3. The effects of different fertilizer treatments on plant height under different intercropping systems

Conclusions

Integration of bio-fertilizer + 50% chemical fertilizer can be served as a procedure in order to reduce the consumption of chemical fertilizers in line with sustainable agriculture. Results of this research clearly demonstrated the useful effect of nitroxin biofertilizer to increase yield of forage maize and the integrated use of chemical fertilizers and biofertilizer provided encouraging results in increasing yield and improving growth parameters of forage maize under Khorramabad environmental condition. Also, the results of this research revealed that on the majority of traits in this research there was no significant difference between monoculture of corn and its intercropping with sweet basil at different proportions including corn + 25% sweet basil pattern and corn + 50% sweet basil pattern. Some advantages such as difference between their height, separate ecological nests for using resources, and lower nutrient competition are of the favorable features for intercropping corn with sweet basil which may lead to an acceptable biological yield. As a rule, one of the main criteria for evaluating the rate of success in plant production is its ultimate yield (biological or economical yield depending on the purpose). The higher yield gained by applying a treatment of integration of bio-fertilizer + 50% chemical fertilizer and intercropping treatments of forage corn + 25% sweet basil and forage corn + 50% sweet basil are considered to be a good indicator and criterion for assessing the success of this approach and practically using it by farmers in order to reduce the consumption of chemical fertilizers generating pollution and also serving as an efficient tool for access to the sustainable agriculture.

References

Agegnehu, G., Ghizaw, A., Sinebo, W. (2008) Yield potential and land-use efficiency of wheat and faba bean mixed intercropping. Agronomy for Sustainable Development, 28, 257-263. DOI: 10.1051/agro:2008012

Aiyer, A.K.Y.N. (1963) Principles of crop husbandry in India. 4th edition. Bangalore: Bangalore Press

Bagheri, M., Zaefarian, F., Bicharanlou, B., Ghanizadeh, H. (2014) A study of intercropping of maize with sweet basil and borag. Cercetari Agronomice in Moldova, 47 (2), 13-28. DOI: 10.2478/cerce-2014-0013

Banik, P., Midya, A., Sarkar, B.K., Ghose, S.S. (2006) Wheat and chickpea intercropping systems in an additive series experiment: Advantages and weed smothering. European Journal of Agronomy, 24, 325-332. DOI: 10.1016/j.eja.2005.10.010

Bashan, Y., Holguin, G., De-Bashan, L.E. (2004) *Azospirillum*- plant relationships: physiological, molecular, agricultural, and environmental advances. Canadian Journal of Microbiology, 50 (8), 521-577. DOI: 10.1139/W04-035

Bibi, Z., Khan, N., Akram, M., Khan, Q., Khan, M.J., Batool, S., Makhdum, K. (2010) Integrating cultivars with reduced herbicides rates for weed management in maize. Pakistan Journal of Botany, 42 (3), 1923-1929.

Cechin, I., Fumis, T.F. (2004) Effect of nitrogen supply on growth and photosynthesis of sunflower plants grown in the greenhouse. Plant Science, 166, 1379-1385. DOI: 10.1016/j.plantsci.2004.01.020

Chandrasekar, B.R., Ambrose, G., Jayabalan, N. (2005) Influence of biofertilizers and nitrogen source level on the growth and yield of *Echinochloa frumentacea* (Roxb.) Link. Journal of Agricultural Technology, 1, 223-234.

Chen, X., Cui, Z., Fan, M., Vitousek, R., Zhao, M., Ma, W., Wang, Z., Zhang, W., Yan, X., Yang, J., Deng, X., Gao, Q., Zhang, Q., Guo, S., Ren, J., Li, S., Ye, Y., Wang, Z., Huang, J., Tang, Q., Sun, Y., Peng, X., Zhang, J., He, M., Zhu, Y., Xue, J., Wang, G., Wu, L., An, N., Wu, L., Ma, L., Zhang, W., Zhang, F. (2014) Producing more grain with lower environmental costs. Nature, 514, 486-491. DOI: 10.1038/nature13609

Cocking, E.C. (2003) Endophytic colonization of plant roots by nitrogen-fixing bacteria. Plant soil, 252, 169-175. DOI: 10.1023/A:1024106605806

Cox, W.J., Kalonge, S., Cherney, D.J.R., Reid, W.S. (1993) Growth, yield and quality of forage maize under different nitrogen management practices. Agronomy Journal, 85, 344 -347. DOI: 10.2134/agronj1993.00021962008500020033x

Dobbelaere, S., Croonenborghs, A., Thys, A., Ptacek, D., Okon, Y., Vanderleyden, J. (2002) Effect of inoculation with wild type *Azospirillum brasilense* and *A. irakense* strains on development and nitrogen uptake of spring wheat and grain maize. Biology and Fertility of Soils, 36, 284–297. DOI: 10.1007/s00374-002-0534-9

Dordas, C., Sioulas, C. (2008) Safflower yield, chlorophyll content, photosynthesis, and water use efficiency response to nitrogen fertilization under rainfed conditions. Industrial Crops and Products, 27, 75-85. DOI: 10.1016/j.indcrop.2007.07.020

Evans, J.R., Terashima, I. (1987) Effects of nitrogen nutrition on electron transport components and photosynthesis in spinach. Australian Journal of Plant Physiology, l4, 281-292. DOI: 10.1071/PP9870059

Evans, J.R. (1989) Photosynthesis and nitrogen relationship in leaves of C3 plants. Oecologia, 78, 9-19. DOI: 10.1007/BF00377192

Fredeen, A.L., Gamon, J.A., Field, C.B. (1991) Responses of photosynthesis and carbohydrate partitioning to limitations in nitrogen and water availability in field grown sunflower. Plant Cell And Environment, 14, 963-970. DOI: 10.1111/pce.12952

Ghanbari, A., Ghadiri, H., Jokar, M. (2006) Effect of intercropping of maize and cucumber on controlling weeds. Pajouhesh and Sazandegi, 73, 193-199.

Girma, A. (2015) Yield advantage and economic benefit of maize basil intercropping under different spatial arrangements and nitrogen rates. Scholarly Journal of Agricultural Science, 5 (8), 296-302.

Guo, J.H., Liu, X.J., Zhang, Y., Shen, J.L., Han, W.X., Zhang, W.F., Christie, P., Goulding, K.W.T., Vitousek, P.M., Zhang, F.S. (2010) Significant acidification in major Chinese croplands. Science, 327, 1008-1010. DOI: 10.1126/science.1182570

Hernandez, A.N., Hernandez, A., Heydrich, M. (1995) Selection of rhizobacteria for use in maize cultivation. Cultivar Tropicales, 6, 5-8.

Hirpa, T. (2014) Response of maize crop to spatial arrangement and staggered inter seeding of haricot bean. International Journal of Environment, 3, 126-138. DOI: 10.3126/ije.v3i3.11072

Hungria, M., Campo, R.J., Souza, E.M., Pedrosa, F.O. (2010) Inoculation with selected strains of *Azospirillum brasilense* and *A. lipoferum* improves yields of maize and wheat in Brazil. Plant Soil, 331, 413–425. DOI: 10.1007/s11104-009-0262-0

Javanmard, A., Dabbagh Mohammadi Nasab, A., Javanshir, A., Moghaddam, M., Janmohammadi, H., Nasiri, Y., Shekari, F. (2013) Evaluation of some agronomic and physiological traits and forage quality in maize - legume intercropping as double cropping. Journal of Agricultural Science and Sustinable Production, 23 (2), 1-18.

Kader, M.A., Main, M.H., Hoque, M.S. (2002) Effects of *Azotobacter* inoculant on the yield and nitrogen uptake by wheat. Journal of Biological Sciences, 2, 259-261. DOI: 10.3923/jbs.2002.259.261

Kennedy, I.R., Choudhury, A.T.M.A., Kecskes, M.L. (2004) Non-symbiotic bacterial diazotrophs in crop-farming systems: can their potential for plant growth promotion is better exploited?. Soil Biology and Biochemistry, 36, 1229-1244. DOI: 10.1016/j.soilbio.2004.04.006

Koocheki, A., Khajeh Hosseini, M. (2008) Modern agronomy. Mashhad: Jehade Daneshgahi Mashhad Press.

Lichtenthaler, H.K. (1987) Chlorophylls and cartenoides: pigments of hotosynthetice bio-membranes. In: S.P. Colowick, N.O. Kaplan, ed. (1987) Method in Enzymol. NewYork: Academic Press. 48, 350-382.

Lithourgidis, A.S., Vlachostergios, D.N., Dorbas, C.A., Damalas, C.A. (2011) Dry matter yield, nitrogen content, and competition in pea-cereal intercropping systems. European Journal of Agronomy, 34, 287-294. DOI: 10.1016/j.eja.2011.02.007

Malakouti, M.J. (1998) Diagnosis and recommendation integrated system (DRIS) an appropriate method for balanced fertilization on the calcareous soils of Iran. Tehran: Tarbiat Modars University Press.

McGuirk, S.M., Semard, S.D. (2005) Toxicologic emergencies in cattle. Veterinary clinics of North America. Food Animal Practice, 21, 729-749. DOI: 10.1016/j.cvfa.2005.08.001

Molla Hoseini, H., Zand, B., Silespour, M. (2006) Evaluation of different nitrogen levels and plant population on quantitative and qualitative yield of silage corn (704) in Varamin. Iranian Journal of Crop Sciences, 8 (3), 250-258.

Nanda, S.S., Swain, K.C., Panda, S.C., Mohanty, A.K., Alim, M.A. (1995) Effect of nitrogen and biofertilizers in fodder rainfed upland conditions of Orisa. Current Agricultural Research, 8, 45-47.

Nasiri Mehallati, M., Koocheki, A.R., Jahan, M. (2010) Light absorption and use efficiency in winter wheat and maize in sequential cropping and relay intercropping. Iranian Journal of Field Crops Research, 8 (6), 878-890.

Olowe, V.I.O., Adeyemo, A.Y. (2009) Enhanced crop productivity and compatibility through intercropping of sesame and sunflower varieties. Annals of Applied Biology, 155, 285–291.

Pandey, R.K., Maranville, J.W., Admou, A. (2000) Deficit irrigation and nitrogen effects on maize in a Sahelian environment. I. Grain yield and yield components. Agricultural Water Management, 46, 1–13. DOI: 10.1016/s0378-3774(00)00073-1

Pilbeam, C.J., Okalebo, R., Simmonds, L.P., Gathua, K.W. (1994) Analysis of maize-common bean intercrops in semiarid Kenya. Journal of Agricultural Science – Cambridge, 123, 191-198. DOI: 10.1017/S0021859600068453

Rahi, A.R. (2013) Effect of nitroxin biofertilizer on morphological and physiological traits of *Amaranthus retroflexus*. Iranian Journal of Plant Physiology, 4 (1), 899-905.

Rohitashav, S., Sood, B.K., Sharma, V.K., Singh, R. (1993) Response of forage maize to *Azotobacter* inoculation and nitrogen. Indian Journal of Agronomy, 38, 555-558.

Safikhani, S., Chaichi, M.R., Poor-Babaei, A.A. (2014) The effect of different N fertilizer sources on forage yield of Berseem clover (*Trifolium alexandrinum* L.) in an additive intercropping system with basil (*Ocimum basilicum* L.). Iranian Journal of Crop Science, 44 (2), 237-248.

Shobeiri, S., Habibi, D., Kashani, A., Paknejad, F., Jafari, H. (2015) Study of physiological traits of grass pea with barely in pure and mixed cropping under dry land and irrigated conditions. Agronomy Journal (Pajouhesh & Sazandegi), 107, 91-98.

Smiciklas, K.D., Below, F.E. (1990) Influence of heterotic pattern on nitrogen use and yield of maize. Maydica Journal, 35, 209–213.

Smidt, N.W., Brimer, L. (2005) The use of herbs in pastures: An interview survey of biodynamic and organic farmers with dairy cattle. Agriculture and Human Values, 22, 355-363. DOI: 10.1007/s10460-005-6051-3

Smolen, S., Sady, W. (2009) The effect of various nitrogen fertilization and foliar nutrition regimes on the concentrations of nitrates, ammonium ions, dry matter and N-total in carrot (*Daucus carota* L.) roots. Scientia Horticulturae, 119 (3), 219-231. DOI: 10.1016/j.scienta.2008.07.030

Tagne, A., Feujio, T.P., Sonna, C. (2008) Essential oil and plant extracts as potential substitutes to synthetic fungicides in the control of fungi. International Conference Diversifying crop protection, 12-15 October 2008, France: La Grande-Motte.

Thobatsi, T. (2009) Growth and yield responses of maize (*zea mays* L.) and cowpea (*vigna unguiculata* L.) in an intercropping system. M Sc Thesis, University of Pretoria. Pretoria, South Africa.

Weiss, E.A. (2000) Oilseed Crops. London: Blackwell Publishing Limited.

Zegada-Lizarazu, W., Izumi, Y., Iijima, M. (2006) Water competition of intercropped pearl millet with cowpea under drought and soil compaction stress. Plant Production Science, 9 (2),123-132. DOI: 10.1626/pps.9.123