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Effect of deficit and adequate irrigation and nitrogen fertilizer levels on physiological traits of maize in Kermansha province – Iran

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ABSTRACT

In order to evaluation effects of drought stress and nitrogen levels on physiological traits of maize hybrid (Ksc703), a field study was conducted at Kermanshah province, Western Iran, during 2009 and 2010. Effects of irrigation (optimum, 85% and 60% of water requirement) and nitrogen (recommended, plus 25% and minus 25%) levels were studied by using a split plot model and 4 replications. The LAI, chlorophyll content and Chlorophyll fluorescence (Fv/Fm) were measured with a Sun Scan, chlorophyll meter (SPAD) and fluorimeter respectively. Chlorophyll content and Fv/Fm was found to decrease with diminishing of available water, hence increase of N resulted in increase of chlorophyll content and Fv/Fm in both normal and stress conditions. Drought stress reduced LAI in all N levels. The increase of N increased LAI in normal condition. In contrast LAI increased by increase of N usage, but in high level of N (more than recommended rate) LAI reduced. Drought stress reduced RWC, high level of N reduced RWC also.

INTRODUCTION

Maize (*Zea mays* L.) is an important crop that used as food, feed and industrial products. Maize is the third most important cereal after wheat and rice all over the world and the world's largest grain crop in term of total production on a MT basis. Maize planting area is about 184 million hectares in 125 countries and is the most important crop in 75 countries (FAOSTAT, 2015). Maize is one of the most important crops in the western part of Iran there is a shortfall in the production of animal feeds. Kermanshah is located in western Iran and Maize is the most important crop after wheat, grown on an area of 45,000 ha with the production of 382,500 tones

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with 8500 kg ha-1 average grain yield; the third and first place of Iran for area harvested and mean yield, respectively. The mean annual precipitation in Iran is 240 ml and it is seen as a dry or semidry country. Maize is an irrigated crop in Iran and recent drought periods in Iran imposed pressure on groundwater resources. The groundwater is the primary source of irrigation of maize production in province and in recent years, the groundwater levels has gradually decreased in this region mainly because of increasing annual irrigation and the dry climate (Agricultural Department of Kermanshah, 2015).

The dearth of water is one of the major factors challenging maize production. Among agro-meteorological hazards, drought has the greatest effect on yield stability (Vinocur and Altman, 2005). It can seriously affect the grain quality and grain output reducing average yields by 50% or even more (Wang et al., 2003). Water shortage due to decreasing annual precipitation and the dry climate, as well as low fertility and low percentage of organic matter in soil is major problems of maize production in Iran. For this reason, overuse of chemical fertilizers in Iran is rising, leading to environmental pollution and soil degradation. Yield losses include more than two- thirds of the total damage of abiotic stresses due to drought, salinity and other factors. Maize is highly sensitive to drought stress, specifically in flowering stage (Tollenaar and Lee, 2011).

Drought is a major problem for the production of the world's five principal cereals: maize, wheat, rice, pearl millet, and sorghum. Water stress reduced yield in crop, but interactive effects of water and nitrogen deficits on physiological traits and on physiological changes associated with leaf aging have received little attention (Valliyodan and Nguyen 2006). Soil water deficit reduces yield of maize (*Zea mays* L.) and other grain crops by three main mechanisms. First, whole canopy absorption of incident Photosynthesis Active Radiation (PAR) may be reduced, either by drought induced limitation of leaf area expansion, by temporary leaf wilting or rolling during periods of severe stress, or by early leaf senescence. Second, drought stress reduces the efficiency with which absorbed PAR is used by the crop to produce new dry matter. Third, drought stress may limit grain yield of maize by reducing the harvest index (Earl and Davis, 2003).

Mihailovic et al (1992) demonstrated one of the factors influencing physiological responses of plants to water stress is mineral nutrition. A significant role of nitrogen in regulating plant responses to water stress was established in a number of plant species. According to estimates of CIMMYT (International Maize and Wheat Improvement Centre) regarding abiotic stresses, the most significant causes of yield loss on farmers fields are low fertility (predominantly N deficiency) followed by drought and, less important, by plant competition related to low planting densities, weeds and intercrops (Ribaut and Poland, 1999). Some of the effects of drought stress on physiological traits of plants are suitable. Photosynthetic rate, leaf surface area and photosynthetic capacity enhanced with increase in nitrogen levels (Gungula and et al, 2005). Leaf area and LAI increase with increase in nitrogen levels (Oscar and Tollennar, 2006).

The chlorophyll meter (or SPAD meter) is a simple, portable diagnostic tool that measures the greenness or the relative chlorophyll concentration of leaves. Compared with the traditional destructive methods, this equipment might provide a substantial saving in time, space and resources. The Minolta Soil Plant Analysis Development (SPAD-502) chlorophyll meter is one tool that enables researchers to determine chlorophyll content by measuring leaf greenness (Peterson et al., 1993). The SPAD uses a silicon photodiode to derive the ratio of transmittance through the leaf tissue at 650 nm compared with transmittance at 940 nm, and a value is given based on that ratio. SPAD measures relative chlorophyll content in plant leaves. Because chlorophyll content is closely related to N supply (Pandey et al., 2000), SPAD is also used to diagnose maize N status and predict maize grain yield potential (Vetsch and Randall, 2004). Janos (2010) reported a close correlation between N fertilization and SPAD readings. Increasing N application increased N content and chlorophyll content in maize (Rambo et al., 2010). Factors affecting SPAD values include radiation differences between seasons, variety and species differences, plant and soil nutrient status (including N and other nutrients), and biotic and abiotic stresses (Peterson et al., 1993).

Atteya (2003) showed exposure of plants to drought lead to noticeable decrease in leaf water potential and relative water content (RWC) and Water stress changed the relation between leaf water potential and relative water content of maize so that stressed plants had lower water potentials than control at the same leaf RWC. The RWC measurement characterizes the internal water status of plant tissues and is also a convenient method for following changes in tissue water content without errors caused by continually changing tissue dry weight (Erickson et al., 1991). On the other hand using the chlorophyll fluorescence technique, is useful possible to estimate the parameters of actual photosynthetic efficiency of leaf, under various conditions at various times, and also the potential maximum of the quantum efficiency (Fv/Fm). Fv/Fm is the measurement of quantum yield potential of photosynthesis, or maximal photochemical efficiency of PSII. The Fv/Fm ratio has been shown to be a reliable indicator of stress (Duraes and et al, 2001). Photosynthesis, as a significant physiological process to yield is sensitive to water stress. The photosynthetic rate keeps decreasing while the intension of stress increases, which is the main reason for the reduction of yield by drought, Moreover, it is possible to determine if there is damage to light reaction systems in photosynthetic machinery during drought (Liu et al., 2012). Measuring chlorophyll fluorescence has become a very useful technique in obtaining rapid qualitative and quantitative information on photosynthesis (Rohácek, 1999), and it can provide information on the relationship between structure and function of photosystem II (PSII) reaction center (Rosenqvist and Van Kooten, 2003). Chlorophyll fluorescence provides useful information about leaf photosynthetic performance of many plants under drought stress (Baker and Rosenqvist, 2004).

Schlemmer et al (2005) reported a very strong relationship between the Minolta SPAD-502 chlorophyll meter readings and direct measurements of chlorophyll content in maize and soybean leaves. Since chlorophyll content is usually strongly related to N concentration, these meters can be used as indicators of need for agricultural N application. So, present research conducted in Kermansh, a drought stress prone province, to study of physiological aspects of water deficit and nitrogen levels on growth and development of maize hybrids.

MATERIAL AND METHODS

In order to evaluation effects of drought stress and nitrogen levels on physiological traits of maize hybrids, a field study conducted in Kermanshah province, western Iran, at 2016 and 2017 at the agricultural research farm, Agricultural and Natural Resources Research Centre in Kermanshah, Iran. This farm is located at 34.08 N, 46.26 E, 1345 m altitude, silty clay soil, pH=7.5-8, 450 mm precipitation Mediterranean climate.. The KSC703 (late maturating group) maize hybrid is dominate commercial cultivar of Kermansha province. Experiment plots were seeded with 75 cm row to row distance and plant density was 75000 plant/ha (conventional plant density). Seeds were sown 7 cm deep. Maize was planted in May and by experimental planter. Irrigation (optimum, 85% and 60% of water requirement) and nitrogen levels (recommended, plus 25% and minus 25%) arranged as main and sub plots respectively using a complete randomized block and 4 replication were used. LAI was measured with a Sun Scan canopy analysis system (Delta-T Devices, Cambridge, UK) and in stages V6, V10 and R1. Chlorophyll meter (SPAD-502, Minolta) readings were taken in all plots. SPAD reading were taken on the midpoint of the youngest fully expanded leaf.

Chlorophyll fluorescence (potential maximum of the quantum efficiency (Fv/Fm)) in leaves of non-stressed and water stressed plants was measured in R1 stage with chlorophyll fluorimeter (Pocket PEA). The RWC was measured using flag leaves after imposing drought conditions. Leaves were sealed within plastic bags and quickly transferred to the lab. Fresh weight (FW) was determined within 2 h after excision. Turgid weight (TW) was obtained after soaking leaves in distilled water in test tubes for 16 to 18 h at room temperature. After soaking, leaves were quickly and carefully blotted dry with tissue paper in preparation for determining turgid weight. Dry weight (DW) was obtained after oven drying the leaf samples for 72 h at 70°C. RWC was calculated from the formula:

RWC (%) = [fresh weight- dry weight/ turgid weight – dry weight] × 100

Where FW, TW and DW are fresh weight (g), turgid weight (g) and dry weight (g) respectively.

RESULTS AND DISCUSSION

Analysis of data showed that chlorophyll content at tasseling decreased by diminish of available water, hence increase of N, resulted in increase of chlorophyll content in both normal and stress condition. A direct close relationship of nitrogen levels with SPAD (Chlorophyll Meter Readings) was reported in maize (Schlemmer and et al 2005). Ciganda et al (2008) had similar results and reported that chlorophyll content is among the most important crop biophysical characteristics. Chlorophyll can be related to photosynthetic capacity, thus, productivity, developmental stage, and canopy stresses, also Munne-Bosch and Alegre (2000) reported the chlorophyll content was decreased with decreasing the irrigation water and this decrease was correlated with relative water content in leaves. Chlorophyll loss is a negative consequence of water stress.

Drought stress reduced LAI in all N levels. The increase of N increased LAI in normal condition. In contrast LAI increased by increase of N use, but in high level of N (more than recommended rate) LAI reduced. Similarly in accordance with our results, LAI was positively correlated with nitrogen application in normal condition (Oscar and Tollennar, 2006). For most plant species, the shortage of nitrogen or water causes a reduction in leaf area development, changes in leaf tissue composition, leaf cell structure and plant water content (Casa, 2003) and also in maize, drought reduces leaf area, leaf chlorophyll contents, photosynthesis and ultimately lowers the grain yield (Athar and Ashraf, 2005). Stone et al. (2001) reported that water deficit reduces crop growth

Table 1. AOVA table of data (2009 and 2010).

Source of variation	df	Mean squares (MS)			
		CH C	LAI	RWC	Fv/Fm
Replication	3	10.9	0.594	3.203	0.002
Year	1	398.5	2.584	54.028	0.002
Year* replication	3	3.358	4.23	2.425	0.001
Water levels	2	293.136**	4.871**	1312.09**	0.006**
Year* water	2	70.996	0.43	9.915	0.001
Error	12	6.156	0.07	24.556	0.001
Nitrogen levels	2	325.969**	0.56**	41.383**	0.001
Year* Nitrogen	2	8.211 ns	0.32 ns	0.156 ns	0.0010 ns
Water* Nitrogen	4	2.091 ns	1.71**	19.202*	0.001 ns
Water * Nitrogen * Year	4	0.976 ns	0.076	1.977	0.001
Error	36	5.108	0.54	7.065	0.001
C.V.		6.9	9.16	4.71	5.78

Table 2. The effect of Water levels on Chlorophyll content (Ch C), LAI, Relative water content (RWC), and Fv/Fm.

Water levels	CH C	LAI	RWC	Fv/Fm
Optimum (100%)	41.44	3.03	79.15	0.81
80% requirement	38.96	2.60	71.66	0.789
60% requirement	34.54	2.13	64.36	0.781

Table 3. The effect of Nitrogen levels on Chlorophyll content (Ch C), LAI, Relative water content (RWC), and Fv/Fm.

Nitrogen levels	CH C	LAI	RWC	Fv/Fm
Recommended - 25%	34.45	2.49	73.02	0.78
Recommended	38.45	2.51	71.76	0.79
Recommended + 25%	41.8	2.06	70.39	0.80

and morphological characteristics of maize plant. In maize, reproductive growth after the silking and flowering stages is the critical period for yield, and chlorophyll content and intact chloroplast structure are key factors for accumulation of dry matter and high yields (Yu et al., 2010).

The RWC measurement characterizes the internal water status of plant tissues and is also a convenient method for following changes in tissue water content without errors caused by continually changing tissue dry weight. Drought stress reduced RWC. The RWC was 80 to 64 for normal and drought stress condition respectively. High level of N reduced RWC also. Jabasingh and Saravana Babu (2014) had similar results and reported that the relative water content in leaves of different maize cultivars decreased significantly and with drought stress, the membrane permeability of the leaf cell markedly increased. Also Higher RWC indicates better growth

Table 4. The effect of Nitrogen and water levels on Chlorophyll content (Ch C), LAI, Relative water content (RWC), and Fv/Fm.

Water levels	Nitrogen levels	CH C	LAI	RWC	Fv/Fm
Optimum (100%)	Recommended - 25%	37.6	2.69	78.8	0.80
	Recommended	41.83	2.9	79	0.81
	Recommended + 25%	44.9	3.7	79.5	0.82
80% requirement	Recommended - 25%	34.9	2.4	74	0.78
	Recommended	38.9	2.7	72.1	0.79
	Recommended + 25%	43.03	2.5	68.1	0.79
60% requirement	Recommended - 25%	30.84	2.3	66	0.76
	Recommended	35.33	2.1	64.1	0.76
	Recommended + 25%	37.46	1.9	62.8	0.77

and development, which in turn depends on leaf area (Sivakumar, 2014).

The results showed that with reduction of water availability, the quantum efficiency (Fv/Fm) decreased. On the other hand with increase of nitrogen level, Fv/Fm increased but not statistically significant. Duraes et al (2001) reported that the Fv/Fm will reduce by drought stress in maize hybrids. Photochemical chlorophyll fluorescence quenching, photosystem II quantum yield and electron transport rate and more heat dissipation as compared to controls (Dias and Bruggemann, 2010). Light energy absorbed by chlorophyll molecules in a leaf can undergo one of three fates: it can be used to drive photosynthesis (photochemistry), excessive energy can be dissipated as heat or it can be re-emitted as light-chlorophyll fluorescence. These three processes occur in competition that is any increase in the efficiency of one will result in a decrease in the yield of the other two (Maxwell and Johnson, 2000).

CONCLUSION

Results showed that under drought stress RWC, chlorophyll content, LAI and the quantum efficiency (Fv/Fm) decreased. Therefore, reducing of RWC, LAI and Chlorophyll content or quantum efficiency (Fv/Fm) could be indicative of water stress. On the other hand increase of nitrogen, resulted in increase of chlorophyll content and Fv/Fm in both normal and stress conditions and with increasing amounts of nitrogen up to 2^{nd} level, LAI increased, but RWC decreased.

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