

Investigation the Influences of Manure Sources and Chemical Fertilizers on Yield, Protein and Oil Content of Sunflower under Drought Stress

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Abstract: A field trial was carried out to investigation the effectiveness of organic and inorganic fertilizers alone and in combination on achene yield and quality of sunflower under drought stress in reproductive stage. Experiment was laid out in a randomized complete block design in a split plot arrangement with three replications in Gonabad, Iran, during spring season 2009. Drought stress treatment were kept in main plots and composed of: D₁: control (normal irrigation), D₂: drought stress at flowering stage and D₃: Drought stress at seed filling stage and fertilizer treatments in sub plots comprised of: F₁: control (no fertilization), F₂: cattle manure, F₃: sheep manure, F₄: poultry manure, F₅: chemical fertilizer, F₆: 50% cattle manure+50% fertilizer, F₇: 50% sheep manure+50% fertilizer, and F₈: 50% poultry manure+50% fertilizer. The results of present study clearly indicated that drought stress induced the decrease of seed yield and 100-achene weight and increase of seed hollowness, but had not any significant influence on head diameter, stem diameter and plant height. Correspondingly, among fertilizer treatments, sunflower plants treated with F₂, F₄ and F₆ treatments showed the highest and control had the lowest amounts in all quantitative traits measured in this experiment. Furthermore, obtained results showed that all the qualitative traits significantly affected by experiment treatments. Water stress at flowering stage caused an increase in seed protein content than other treatments. An adverse effect of drought stress on achene oil contents was observed, so that full irrigation treatment had the highest and drought stress at seed filling stage treatment had the lowest value. Protein yield and oil yield because of great effect of drought stress at reproductive growth stage on seed yield showed significant decreases. F₃ and F₄ treatments as a source of organic manures were statistically superior to other treatments in protein percentage and protein yield, respectively. On the other hand, maximum achene oil content and oil yield were recorded, when crop fertilized by cattle manure alone.

Key words: Drought stress, fertilizer, Sunflower, Yield, Protein, Oil.

INTRODUCTION

Drought stress is a major limiting factor for plant growth and development worldwide and, in Iran, too. Sunflower is a well adapted to drought crop, essentially because of the powerful water uptake due to its efficient root system (Belhassen, 1995). However, it has been found that both quantity and distribution of water has a significant impact on seed yield and seed quality in sunflower (Krizmanic *et al.*, 2003; Iqbal *et al.*, 2005). Intensity of yield reduction by drought stress depends on the growth stage of crop, the severity of the drought and tolerance of genotype (Lorens *et al.*, 1987). Petcu *et al.* (2001) showed that grain yield of sunflower hybrids was affected by drought stress with the low status treatment yielding 10-13% less than the control treatment. Iqbal *et al.* (2005) reported a trend in yield decline and reduce of yield components due to water stress treatments. Razi and Asad (1998) indicated that drought stress at flowering stage was observed to be a limiting factor for seed filling, so significant reduction of unfilled seeds was observed as a result of irrigation missing. Foroud *et al.* (1993) reported decrease in protein and oil content of soybean under high water deficit condition. Organic fertilizers may be used for the crop production as a substitute of the chemical fertilizers because the importance of the organic manures cannot be overlooked. Worldwide, there is growing interest in the use of organic manures due to depletion in the soil fertility (Farhad *et al.*, 2009). Organic agricultural practices aim to enhance biodiversity, biological cycles and soil biological activity so as to achieve optimal natural systems that are socially, ecologically and economically sustainable (Samman *et al.* 2008). The importance of organic manures in the soil has been recognized for centuries as the key to soil fertility and productivity. Organic manures, favorable by product of farming and allied industries contribute to plant growth through their favorable effect on physical, chemical and biological properties of soil (Aowad *et al.*, 2009).

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In the other hand, continuous use of fertilizers creates potential polluting effect in the environment (Oad *et al.*, 2004). Synthesis of chemical fertilizers consumes a large amount of energy and money. However, an organic farming with or without chemical fertilizers seems to be possible solution for these situations (Prabu *et al.*, 2003). The integration of organic sources and synthetic sources of nutrients not only supply essential nutrients but also have some positive interaction with chemical fertilizers to increase their efficiency and thereby reduce environmental hazards (Ahmad *et al.*, 1996). Patil *et al.* (2008) studied the effect of use of manures and fertilizers alone and in combination on soybean and concluded that highest amounts of grain and straw yield and crude protein recorded by conjoint use of manures and fertilizers. Surlekov and Rankov (1989) reported that, the application of FYM along with NPK recorded higher plant height, plant canopy and yield of chickpea. Mallikarjun *et al.* (2000) conducted that integrated application of half of the recommended dose of phosphorus in the form of super phosphate and remaining through cattle urine enriched FYM and produced significantly higher seed yield of sunflower.

Scheiner *et al.* (2002) suggested that excessive nitrogen fertilization of sunflower not only generates that environmental risk, it may also affect the grain quality, decreasing its oil content and reduce yield through an increase of plant lodging. Akbari *et al.* (2011) found that the integrated fertilizer significantly increased the seed protein content and protein yield of sunflower. they also reported that oil content and oil yield significantly increased in response to combinations of organic and inorganic fertilization application compared to the organic alone or inorganic fertilization.

The objective of this study was to investigate sunflower responses to different sources of manure and chemical fertilizer under drought stress condition in reproductive stage.

MATERIALS AND METHODS

A field experiment was conducted in an experimental farm in the city of Gonabad, Iran (34° 21'N, 58°42'E & 1150 m a.s.l.) during growing season of 2010, to study the response of sunflower to combination of different sources of organic manure with inorganic fertilizer under drought stress at reproductive stage. The trial was laid out using a randomized complete block design with split-plot arrangement with three replications. Main plot composed of three levels included: D₁: control (normal irrigation), D₂: irrigation missing at flowering stage and D₃: irrigation missing at seed filling stage and sub plot composed of eight levels included: F₁: control, F₂: cattle manure, F₃: sheep manure, F₄: poultry manure, F₅: chemical fertilizer, F₆: 50% cattle manure+50% chemical fertilizer, F₇: 50% sheep manure+50% chemical fertilizer, and F₈: 50% poultry manure+50% chemical fertilizer. Before sowing, a mixed soil sample composed of different locations of the field prepared and transferred to the laboratory for its physical and chemical analysis. The analysis of the soil is shown in Table 1. Based on soil test, nitrogen (as urea), phosphorus (as triple super phosphate) and potassium (as potassium sulfate) at the rate of 220, 150 and 100 kg ha⁻¹ were applied in recommended chemical fertilizer treatment, respectively. Manure (cattle manure, sheep manure, and poultry manure at the rates of 30, 20 and 10 ton ha⁻¹, respectively) and fertilizer with except of half of urea were applied to the respective plots and incorporated into the soil before sowing. Seeds of sunflower cultivar were sown on May 10, 2009. The crop was sown in 65 cm apart rows and each sub plot had 8 rows. Planting was done by dibbling and placing 3 seeds per hill at 20 cm distance from each other.

At 3-4 leaf stage, one plant per hill was maintained through thinning in order to achieve proper plant population. At 6 leaf stage, remaining N fertilizer was applied. During growing season, two hoeing were done to control weeds. Irrigation in full irrigation treatment was regularly carried out at 7 days interval. In drought stress treatments, sunflower plants were not irrigated temporary for seven days interval.

The crop was harvested by hand on September 16, 2009. At harvesting time, heads of the two inner ridges of each plot were harvested and left 10 days until fully air dried by sunshine and seed yield, 100-achene weight and seed hollowness were estimated. Also, six plants from each plot were selected at random and plant height, stem diameter and head diameter were measured and average was calculated from those six measured values. Seed protein content was determined by measuring the nitrogen content with the Microkjeldhal method and multiplying it by 6.25 to express to total protein content (Bremner, 1996). Seed oil content was determined according to A.O.A.C. (1990) using soxhlet apparatus and diethyl ether as a solvent. Protein yield and oil yield were calculated by multiplying seed yield by protein and oil percentage, respectively.

Finally, all data were subjected to analyses of variance using SAS program (SAS., 1990) and wherever appropriate, the mean values were compared according to Duncan's multiple range test at 5% probability level.

Table 1: Physical and chemical analysis of a representative soil sample from the experimental site.

| Depth (cm) | Soil texture | ECe (ds m ⁻¹) | pH | O.C (%) | N (%) | P (mg kg ⁻¹) | K (mg kg ⁻¹) |
|------------|--------------|---------------------------|-----|---------|-------|--------------------------|--------------------------|
| 0-30 | Clay loam | 3.4 | 7.8 | 0.6 | 0.05 | 3.5 | 147 |

RESULTS AND DISCUSSION

Yield and Yield Attributes:

Achene yield of sunflower was significantly affected by drought stress treatments (Table 2). Drought stress at flowering and seed filling stages resulted in decrease in achene yield, so that the highest amount (3872 kg ha^{-1}) was recorded in control, while the lowest values were obtained in drought stress at flowering stage (3391 kg ha^{-1}) and drought stress at seed filling stage (3375 kg ha^{-1}), so that did not differ significantly from each other with respect to this parameter (Figure 1). Karaata (1991) conducted experiments to identify highly drought sensitive growth stages of sunflower, and it was found that maximum reduction in yield occurred when drought was imposed during flowering stage. Mozaffari *et al.* (1996) concluded that drought stress for longer than 12 days at seed filling and flowering stage of sunflower was the most damaging in reducing the seed yield.

Data presented in Table 2 show that achene yield was significantly affected by different levels of manures and chemical fertilizer. Results of the comparison of treatments' means presented in Figure 2 showed that maximum achene yield (3752 kg ha^{-1}) was obtained from F_2 where 100% cattle manure was applied which was statistically equal to that of F_4 (100% poultry manure) and F_6 (50% cattle manure+50% chemical fertilizer) treatments giving seed yields of 3744 kg ha^{-1} and 3738 kg ha^{-1} , respectively. The lowest seed yield (2942 kg ha^{-1}) was obtained from F_1 (control). Similar findings were also reported by Gorttappeh *et al.* (2000) and Saeed *et al.* (2002), who stated that organic manure alone or in combination with synthetic fertilizers significantly increased achene yield of sunflower compared with control. The results presented in Table 2 revealed that head diameter was not significantly affected by drought stress treatments, but, different levels of manures and fertilizers had significant effect on this parameter. Among all treatments, F_2 and F_6 with means of 14.27 cm and 14.26 cm had the highest values and the lowest value (12.38 cm) was recorded in control (Table 3).

Drought stress significantly influenced 100-achene weight as shown in Table 2. Data in Table 3 clearly reveal that irrigation missed at seed filling stage significantly reduced this trait compared to control and followed by drought stress at flowering stage. The different sources of organic manures and chemical fertilizer had a significant effect on 100-achene weight (Table 2). The comparison of treatments' means reveals that maximum amount recorded from plots F_2 , F_6 , and F_8 which did not differ significantly from each other with respect to 100-achene weight of sunflower while control gave minimum value (Table 3). This significant increase may be due to improvement in translocation of assimilates to the seeds. Gorttappeh *et al.*, (2000) showed that application of nitrogen fertilizers and manure increased 100-achene weight of sunflower plants. Regarding the influence of drought stress on seed hollowness data in Table 2 showed that this variable significantly affected by drought stress treatments. It is clear from Table 3 that drought stress at flowering stage significantly exceeded seed hollowness than control, so that 27% increased this parameter than control. Results presented in Table 2 reveal that fertilizer treatments were affected seed hollowness in a significant way. The maximum value (14.94%) was noted with control and the minimum value (10.47%) was recorded from F_4 treatment, followed by F_6 by mean of 10.57% (Table 3).

Analysis of variance for the data for plant height revealed that this trait was not significantly affected by the drought stress treatments, while significant differences calculated in plant height between different fertilizer treatments (Table 2). Data cited in Table 3 show that the conjoint use of cattle manure and chemical fertilizer (F_6 treatment) produced the tallest plants (193.1 cm), while the lowest value (170.8 cm) observed in control. As shown in Table 2, there was no significant difference in the stem diameter of sunflower affected by drought stress treatments. But, our analyses showed that fertilizer treatments influenced this parameter, significantly. Stem diameter was significantly higher in F_2 , when crop was given 100% cattle manure which was statically equal to that F_6 , when sunflower plants fertilized by 50% cattle manure+50% chemical fertilizer. In contrast, the lowest value was recorded from control. Aowad *et al.* (2009) evaluate the effects of manure and mineral fertilizer on sunflower characteristics and found that the highest values of stem diameter and plant height produced due to combination of nitrogen fertilizer with farm yard manure.

Achen Protein Content:

Highly significant ($P \leq 0.01$) differences were observed among water stress treatments with respect to seed protein content (Table 2). The comparison of treatments' means (Table 3) indicate that irrigation missing at flowering stage significantly enhanced protein percentage of sunflower than other treatments, so that plants treated with this treatment had the highest value (21.9%). Our results are in agreement with the findings of Ahmad *et al.*, (2009) who reported that seed protein content of sunflower was significantly higher in irrigation missing at flowering stage than in control, when crop was given normal irrigations.

Results obtained from the variance analysis shows that fertilizer treatments have a significant difference in the case of protein content (Table 2). Among all treatments, the maximum value (22.7%) related to F_3 treatment (100% sheep manure) and the minimum amount of it (19%) related to control (Table 3). Results are in line with findings of Nanjundappa *et al.*, (2001) who reported an increase in seed protein content of sunflower due to application of organic sources.

Difference between sunflower plants in the case of protein yield was significant in response to drought stress and fertilizer treatments (Table 2). According to the table of means comparison (Table 3), protein yield was significantly lower in water-stressed plants than in plants grown under full irrigation. The highest amount (842 kg ha^{-1}) was obtained in plants irrigated in whole growing period, while the lowest values were obtained in drought stress at flowering stage (848.7 kg ha^{-1}) and drought stress at seed filling stage (676.7 kg ha^{-1}), so that had not significant difference from each other with respect to protein yield.

It can be noticed generally from the data recorded in Table 3 that among all fertilizer treatments, the highest value of protein yield (847.1 kg ha^{-1}) was achieved by application of 100% poultry manure and the lowest value (568 kg ha^{-1}) was obtained from control. Chawale *et al.* (1995) also reported that fertilization with FYM increased seed oil of groundnut.

Achene oil Content:

Data (Table 2) showed that drought stress and different organic and inorganic fertilizers had significant effect on the oil content of sunflower. There was a significant negative effect of drought stress at seed filling stage on this trait. The highest oil content (39.2%) was recorded in plants that not exposed to drought stress which was statistically at par with plants that exposed to drought at flowering stage (38.7%). The lowest oil content (37.4%) was related to drought stress at seed filling stage (Table 3). The low oil content may be due to the short seed filling stage caused by the temporary irrigation missing during this period. Iqbal *et al.*, (2005) examined the effect of water stress on sunflower lines and concluded that drought stress in reproductive stage reduced 4 and 7% achene oil content of Suncross and Gulshan-98 lines compared to control, respectively.

As shown in Table 3, seed oil differed among fertilizer treatments. Sunflower plants which fertilized with 50% cattle manure and 50% chemical fertilizer had the highest oil content (39.7%) and plants fertilized with chemical fertilizer alone had the lowest value (37.2%). Bakht *et al.* (2010) reported that seed oil of sunflower decreased with an increase in the rate of NP fertilizers. Such results may be due to the adverse effect of nitrogen on oil content, is offset by an increase in protein content and a reduced availability of carbohydrates for oil synthesis at high N supply.

Table 2: Analysis of variance of measured parameters of sunflower as affected by experiment treatments.

| S.O.V | df | Achene yield | Head diameter | 100-Achene weight | Seed hollowness | Plant height | Stem diameter | Protein content | Protein yield | Oil content | Oil yield |
|-------------|----|--------------|---------------|-------------------|-----------------|--------------|---------------|-----------------|---------------|-------------|------------|
| Drought | 2 | 1258317.0** | 1.007ns | 0.503** | 45.9** | 0.073ns | 0.002ns | 22.93** | 164834.7** | 20.22** | 318535.7** |
| Error a | 4 | 64812.8 | 0.92 | 0.097 | 0.05 | 113.6 | 0.004 | 20.22 | 10671.1 | 7.54 | 7878.9 |
| Fertilizer | 7 | 641319.0** | 3.47** | 1.009** | 3.47** | 374.1** | 0.077** | 9.45* | 63739.3** | 4.62* | 105659.3** |
| Interaction | 14 | 74272.6ns | 0.50ns | 0.026ns | 0.63ns | 22.7ns | 0.001ns | 3.40ns | 11277.2ns | 6.15ns | 11638.5ns |
| Error b | 42 | 78808.6 | 0.18 | 0.66 | 0.66 | 32.3 | 0.003 | 4.30 | 9207.7 | 2.10 | 13464.1 |
| CV (%) | | 7.98 | 3.12 | 1.92 | 8.04 | 3.06 | 2.28 | 9.86 | 12.7 | 3.75 | 8.6 |

Ns= Non significant; * and ** = Significant at 5% and 1% probability, respectively.

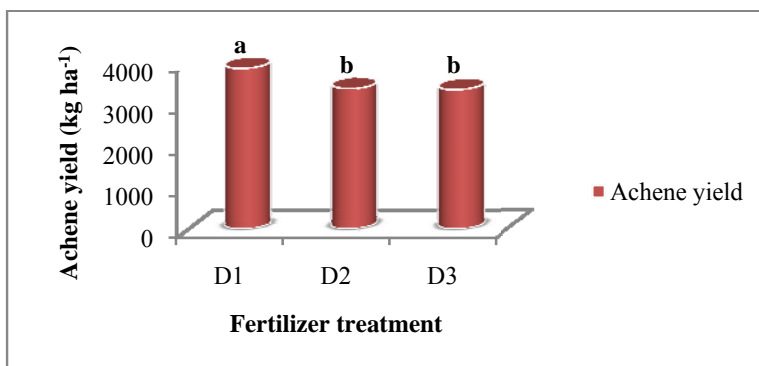


Fig. 1: Achene yield of sunflower as affected by drought stress.

Data presented in Table 2 show a significant effect due to experiment treatments on oil yield of sunflower. Drought stress results in the decrease of the oil yield of sunflower. The highest amount of oil yield (1483 kg ha^{-1}) was achieved under the condition of no drought stress but the lowest value (1261 kg ha^{-1}) was related to the treatment of drought stress at seed filling stage. Although, the difference between it and drought stress at flowering stage (1316 kg ha^{-1}) was not statically significant (Table 3). El Naim and Ahmed (2010) found that irrigation every 7 days increased oil content and oil yield; while the extended irrigation interval had reduced oil percentage and oil yield of sunflower.

It is evident from the Table 3 that, all fertilizer treatments significantly increased the oil yield as compared to control. Among all treatments, the highest oil yield (1482.4 kg ha⁻¹) was recorded under conjoint use of cattle manure and chemical fertilizer followed by application of cattle manure alone (1456.3 kg ha⁻¹). In this experiment the lowest amount of oil yield (1143 kg ha⁻¹) was related to control. These results were similar with what was obtained by Gorttappah *et al.* (2000), who reported that the highest amount of oil yield in sunflower plants was recorded due to conjoint use of fertilizer and organic manure.

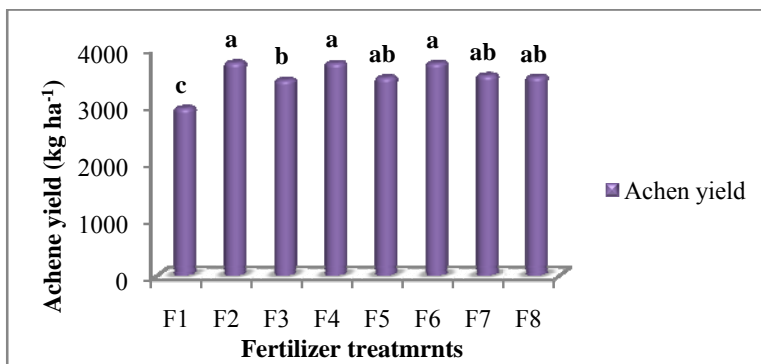


Fig. 2: Achene yield of sunflower as affected by organic and inorganic fertilizers.

Table 3: Mean comparison of the effects of drought stress and fertilizer treatments on selected parameters of sunflower.

| Treatment | Head diameter (cm) | 100-Achene weight (%) | Seed hollowness (%) | Plant height (cm) | Stem diameter (cm) | Protein content (%) | Protein yield (kg ha ⁻¹) | Oil content (%) | Oil yield (kg ha ⁻¹) |
|----------------|--------------------|-----------------------|---------------------|-------------------|--------------------|---------------------|--------------------------------------|-----------------|----------------------------------|
| Drought | | | | | | | | | |
| S ₁ | 14.0a | 10.0a | 10.0c | 187.3a | 2.48a | 21.1ab | 842.0a | 39.2a | 1483a |
| S ₂ | 13.8a | 9.8ab | 12.7a | 183.4a | 2.48a | 21.9a | 748.7b | 38.7a | 1316.b |
| S ₃ | 13.6a | 9.8b | 11.9b | 185.5a | 2.46a | 20.0b | 676.7b | 37.4b | 1261.6b |
| fertilization | | | | | | | | | |
| F ₁ | 12.4d | 9.1c | 14.9a | 170.8c | 2.30d | 19.0c | 568.0c | 38.7ab | 1143.0e |
| F ₂ | 14.3a | 10.1a | 10.9bcd | 187.6b | 2.58a | 20.8abc | 782.0ab | 38.8ab | 1456.3ab |
| F ₃ | 13.8c | 9.8b | 11.4bc | 184.5b | 2.42c | 22.7a | 783.0ab | 38.3abc | 1320.2d |
| F ₄ | 14.2ab | 10.0ab | 10.5d | 187.8ab | 2.50b | 21.5ab | 847.1a | 38.2bc | 1431.1abc |
| F ₅ | 13.6c | 9.9ab | 11.5b | 188.1ab | 2.53ab | 21.2ab | 806.0ab | 37.2c | 1299.0d |
| F ₆ | 14.3a | 10.1a | 10.6cd | 193.1a | 2.57a | 20.7bc | 789.8ab | 39.7a | 1482.4a |
| F ₇ | 13.8b | 10.0a | 11.2bcd | 185.5b | 2.42c | 20.8abc | 736.4b | 38.7ab | 1356.4bcd |
| F ₈ | 14.0ab | 10.0a | 11.1bcd | 185.4b | 2.49b | 21.1ab | 734.6b | 38.2bc | 1333.0cd |

Means followed by same letter in each column are not significantly different at the 5% level.

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