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Effect of sowing date and irrigation regimes on yield components, protein and galactomannan content of guar (*Cyamopsis Tetragonoloba* L.) in Iran climate

Heidar Meftahizade¹, Yousef Hamidoghli^{2*}, Mohammad Hassan Assareh³, Majid Javanmard Dakheli⁴

¹Department of Horticultural Science, University Campus 2, University of Guilan, Rasht, Iran

²Department of Horticultural Science, University of Guilan, Rasht, Iran

³Research Institute of Forests and Rangelands, Karaj, Iran

⁴Department of Chemical Technologies, Iranian Research Organization for Science and Technology, Iran

*Corresponding author: hamdoghli@guilan.ac.ir

Abstract

In this study, the effects of irrigation regimes and sowing dates on growth, yield performance, protein and galactomannans of guar (*Cyamopsis tetragonaloba* L.) were monitored in Ilam province (latitude 37°8" min and Longitude 47°27"), Iran. A field experiment was conducted for two years during 2015 and 2016, using a split-split plot randomized block design including two sowing date (15th and 30th July), 4 irrigation regimes (once: after sowing, twice: after sowing and flowering, three times: after sowing, flowering and seed formation, and four times: after sowing, flowering, seed formation and before harvesting) and 3 genotypes (RGC1031, RGC 936 and RGC1066) with three replicates. Main seed yield attributes such as protein, and galactomannans content were evaluated. The results showed that late sowing date responded better in higher yield than to the earlier one (15th). There were not observed significant differences among studied genotypes in case of yield components, galactomannan and protein contents traits. According to ANOVA analysis, RGC-1066 with 4 times irrigation was suitable treatment in case of seed yield. Also the highest amount of protein and galactomannans was obtained in the late sowing date. These results indicated that guar can be put as an alternative crop and can be introduced as a new crop for cultivation in Iran climate.

Keywords: *Cyamopsis tetragonoloba* L., Iran, irrigation regimes, sowing date, yield components. **Abbreviations:** I_Irrigation regimes, G_Genotypes, D_Sowing dates, DAS_day after sowing.

Introduction

Guar (Cyamopsis tetragonoloba L.), belonging to the family Leguminosae. It is an industrial crop, adapted to the arid and semi-arid area, requiring low humidity and nutritious. The seeds are worthwhile for industrial gum (Sunil et al., 2013). Guar plant has a short life cycle (3-4 months) that can tolerate prolonged periods of drought and high salinity soil (Deepika and Dhingra, 2014). Guar Galactomannan (gum) used as an emollient, softening or thickening agent, flocculent, and stabilizer. This gum can also be used in hydraulic fracturing and a wide range of other industries such as the production of cosmetics, paper, textiles, detergents and food products (Gresta et al., 2014). The gum of guar is applied as a lowering cholesterol, blood pressure, and blood sugar levels. It also plays a positive role in general weight loss by promoting fullness and appetite suppression (Sharma et al., 2011). The extract of guar seeds powder has anticancer activities (Badr et al., 2014).

Sowing dates with high guar seed yields are varied from May to August, while mid-May planting of guar has been reported to produce the highest seed yields in a Mediterranean environment of Italy (Gresta et al., 2013). In the southwest United States, mid-April to late-May, and mid-May to early-July have been reported as optimum sowing date in South Texas and Central West Texas, respectively (Tripp et al., 2011).Guar can potentially be grown under semi-arid climatic conditions receiving less than 250 mm annual rainfall with minimum nutrients and abundant sunshine (Ashraf et al., 2005). Generally, guar is irrigated 3-4 times during cultivation period when it is cultured under irrigation system. It is an extremely drought tolerant plant and very easy to cultivate with a limited supply of water (Singh, 2014). Guar seeds have been produced with 2650 m3/ha average water supply (Gresta et al., 2013).

Seed rate is the main yield components, which many others yield factors, directly or indirectly has the positive correlation with it (Shekhawat and Singhania, 2005). Also, Seed weight showed the negative correlation with the galactomannan content but positive correlation with the protein content (Gresta et al., 2013).

In 2012, world demand for guar gum has increased and the price has approximately risen up to 2.3 times (Gresta et al., 2014). India is the world leader for the production of guar, contributing 80% of the total production in the world. Pakistan with 15% of world production is near to India with major guar production and the remaining is produced by United States of America, Sudan, Australia, Brazil and South Africa (5 percent) (DSIR, 2014). The average of guar seed yield is 400 kg/ha with a large year-to-year variation especially in years of drought (NRAA, 2014).

Cultivation of plants which can tolerate deficient water during growing periods in semi-arid area like Iran is necessary. Much is known about the cultivation of guar crop in India and Pakistan, while, no information is available on guar production in Iran. Therefore, the aim of the present study was to evaluate the possibility of cultivating of guar in semi-arid regions of Iran and to determine the best sowing date and irrigation regimes in order to explore a potential alternative crop for Iranian climate.

Results

Growth stages: Analysis of phonological and morphological data

In the two-year trial, 3 stages of flowering, formation of first pod and harvest took place in 31, 58 and 95 days after sowing for early sowing time (15 June) and 43, 86 and 110 days after sowing for late sowing time (30 July), respectively (Fig. 1, 2). In the current study, the 15th June planting required less time (100 days) to mature than late July plantings in both years, but in general, late sowing time (in both years) was more suitable for guar cultivation (Fig. 1, 2).

Yield and yield components

Plant height was significantly affected by irrigation regimes (I) $p \le 0.01$. In I4 irrigation regime a plant height of 95.9 cm was recorded, while I3, I2 and I1 showed plant height of 73.16, 25.27 and 3.13 cm, respectively (Fig. 3).

The number of branch per plant was significantly affected by irrigation and interaction of irrigation regimes and genotypes (I×G) (Fig. 3, 5). In the first treatment i.e. $GI \times II$, no branch was observed in plants, but in the treatment of I4 × G3, 10 branches per plant were recorded. The average numbers of branch per plant were 9.52, 6.80, 3.27 and 0.58 for I4, I3, I2 and I1 irrigation regimes, respectively (Fig. 3). Finally, the maximum number of branches per plant in all treatments recorded in D2 (late sowing date) (Fig. 9).

The number of pod per branch was significantly affected by sowing date (D), irrigation regimes (I) and interaction of I ×D. As shown in Fig. 7, treatments of D2×I4, D2×I3 and D1×I4 produced 7.88, 6.7 and 6 pods per branch respectively. Late sowing date produced 4.02 pods per branch, while early sowing date produced 3.22 pods per branch.

The number of pod per plant also influenced by sowing date (D), irrigation regimes (I), interaction (I×D) and (I×G). Plants treated with D2 × I4 and D1×I4 produced 82 and 51.22 pods per plant (Fig. 8). Interaction of G×I for this traits was sophisticated, G3 × I4 and G1 × I4 produced 81.08 and 74.75 pods per plant (Fig. 6). Mean comparison of different irrigation regimes of I4, I3, I2 and I1 produced 73, 40, 4.5 and 0.13 pod per plant respectively (Fig. 3). In all treatments, late sowing date and early sowing date produced 33.45 and 25.47 pods per plant (Fig. 9).

The number of seed per pod: Results of ANOVA analysis showed the positive impact of irrigation on the number of seed per pod in $p \le 0.01$ %. Plants irrigated with I4, I3, I2 and I1 regimes produced 5.94, 5.41, 1.55 and 0.02 seeds per pod respectively (Fig. 3). There are non-significant differences between guar verities and sowing date with respect to seed number per pod.

Seed number per plant: Seed number per plant were significantly influenced by I, D, D×I and I×G ($p \le 0.01$) (Fig. 3, 6, 8, 9). Also, the interaction of genotypes and irrigation regimes was significantly different ($p \le 0.01$) for the seed number per plant (Fig. 6). There was also a significant difference between sowing date for seed per plant (seed yield). In the current study, late sowing date and early sowing date produce 190 and 134 seeds per plant (Fig. 9). D2 × I4 and G3 × I4 produced 542 and 478 seeds per plant respectively (Fig. 6, 8).

Pod length: There was a positive significant for treatments of D, I, D×I as well as I×G for pod length. Results showed that treatment of D1× I4 had the longest pod length (6.12) among

all treatments, afterward D2 ×I4, D1×I4 and D2 ×I3 has 5.77, 6.12 and 5.13 cm respectively (Fig. 7). The longest pod (6.44 cm) was recorded in treatment of G3 ×I4 (Fig. 5). As seen in Fig. 3, I4 and I3 produced pod with the length of 6.46 and 6.13 cm respectively. Generally, the average pod length in late sowing date and early sowing date was 3.7 and 3.08 (Fig. 9). There were no significant differences among studied genotypes.

Seed weight: ANOVA analysis showed a significant difference between irrigation regimes, and also interaction of $D \times I$ in $p \le 0.01$. The average 100 seed weight of $D1 \times I4$ was higher than all other treatments (4.83 g/100 NO seed) (Fig. 7).

Seed protein and galactomannan

As shown in Table 1, sowing date and irrigation alone have significantly affected seed protein and galactomannan. But in the case of galactomannan, not only sowing date and irrigation but also interaction between D×I showed significant differences ($p \le 0.01$) (Fig. 8). In all treatments, late and early sowing date recorded 24.38 % and 21.58% gum, respectively. In the case of protein content, our results showed that, late sowing date produced 24.97% vs. 20.97% in early sowing date. As can be seen from Fig.4, among all treatments of irrigation regimes, I4 has recorded the maximum amount of galactomannan and protein, i.e. 32.5% and 32 %, respectively.

Effect of sowing date on investigated traits: the results demonstrated that there are significant differences ($p \le 0.01$) between sowing dates with respect to pod number per branch, pod number per plant, the number of seed per plant, pod length, seed protein and seed galactomannan. Therefore, late sowing date is more suitable for cultivation of guar in studied climate.

Discussion

Time of planting is one of the main important factors which determined seed yield of guar. In the current study, late July, was the best sowing date. Phonological period of late sowing date was longer than first sowing date. It can be deduced that accumulation of nutrition during this long period lead to high performance and quality. There are number of studies which indicate the suitable sowing time in various climates. Gresta et al. (2013) reported increase of 9% higher seed yield in early May planting compared to late-June planting in the Mediterranean environment of Italy. The optimum sowing date in India is July, Long days in June may cause produce taller plant (Gupta et al., 2001).

The impact of irrigation regimes on yield components and water requirement

In this study, irrigation regimes significantly influenced all other investigated traits. It seems, irrigation at the beginning of reproductive phase i.e. flowering formation is critical, that is occurred around 60-70 days after sowing. Studies suggest that one life-saving irrigation at 60 days of sowing increased the grain yield significantly over no irrigation (Meena et al., 1991). Other researchers also recommended 1–2 irrigations for higher crop yield (Singh, Singh, 1988). Guar does not require irrigation at the early vegetative stage but irrigation at flowering and pod formation stage helps the crop in higher



PHENOLOGICAL STAGES

Fig 1. Mean comparisons of phenological stages of guar varieties cultivated in 15 June.



PHENOLOGICAL STAGES

Fig 2. Mean comparisons of phonological stages of guar verities cultivated in 30 July.



Fig 3. Mean comparison of Irrigation regimes in investigated traits of guar. I_1 : one time irrigation, I_2 : twice irrigation, I_3 : three times irrigation and I_4 : four times irrigation. Different letters show significant difference of means at 1% level Duncan's test.



Fig 4. Mean comparison of irrigation regimes on seed number per plant during two years trials. I_1 : one time irrigation, I_2 : twice irrigation, I_3 : three times irrigation and I_4 : four times irrigation. Different letters show significant difference of means at 1% level Duncan's test.



interaction genotypes × irrigation regimes

Fig 5. Mean comparison of interaction $G \times I$ on Branch/plant and pod length in guar. G1, G2 and G3 are genotypes 1, 2 and 3, respectively. I_1 : one time irrigation, I_2 : twice irrigation, I_3 : three times irrigation and I_4 : four times irrigation. Different letters show significant difference of means at 1% level Duncan's test.



interaction of genotypes \times irrigation regimes

Fig 6. Mean comparison of interaction $G \times I$ on pod/plant and seed/plant traits in guar. G1, G2 and G3 are genotypes 1, 2 and 3, respectively. I_1 : one time irrigation, I_2 : twice irrigation, I_3 : three times irrigation and I_4 : four times irrigation. Different letters show significant difference of means at 1% level Duncan's test.



interaction of sowing date × irrigation regimes

Fig 7. Mean compare of interaction of $D \times I$ on investigated traits in guar. D1: early sowing date D2: late sowing date. I₁: one time irrigation, I₂: twice irrigation, I₃: three times irrigation and I₄: four times irrigation. Different letters show significant difference of means at 1% level Duncan's test.



interaction of sowing date × irrigation regimes

Fig 8. Mean comparison of interaction $D \times I$ on seed/plant, pod/plant and Galactomanan% in guar. D1: early sowing date D2: late sowing date. I₁: one time irrigation, I₂: twice irrigation, I₃: three times irrigation and I₄: four times irrigation. Different letters show significant difference of means at 1% level Duncan's test.



Fig 9. Mean compare of Sowing Date (D) on investigated traits in guar. D1: early sowing date D2: late sowing date. Different letters

show significant difference of means at 1% level Duncan's test. yields and productivity (Singh, Singh, 1988), which is in agreement with current results. In both sowing date in two years, there were not any rainfall. In agreement with current results, Gresta et al. (2013) reported that 2200–3000 m3/ ha water is needed for seed production.

Plant height: According to current results, as plant height was taller, the seed number per pod was greater, which this is consistent with Rai, Dharmatti (2014), who reported the significant positive correlation of plant height with seed yield and seed per pod. Many previous studies have indicated that, the most influenced growth trait by water tension was plant height (Cakir, 2004). Deng Manasseh et al. (2016) reported that there are significant differences in plant height between water regimes treatments. Similar results of the decreased plant length during water stress have been reported by (Khanzada et al., 2002), which attributed the effect of plant water tension on the length of internodes to the impact on the elongation of developing cells and cell enlargement, rather than the increase in number of cells. Additionally, in a study conducted by Sudhir et al. (2016) reported, the mid-June planted guar grew taller than early July planting. Our results showed that there are not significant differences among genotypes on plant height; this result is not in agreement with Sanghi et al. (1964) who recorded that there was a wide range of phenotypic variability observed in plant height. This may be due to the genotypic factors.

The number of branches per plant: current finding showed that the number of branches per plant was decreased with water stress. This decreasing trend could be ascribed to the negative effect of reduced plant water tension on the initiation of vegetative primordial. Boutraa, Sanders (2001) found that water stress during flowering and pod-filling stages reduced number of main branches of two cultivars of guar.

The number of pods per branch/plant: irrigation in podfiling for formation of the number of pod per plant is one of the main fundamental stages for guar cultivation. Results of Boutraa, Sanders (2001) are consistent with our results, they reported that water stress during both flowering and podfilling stages decreased seed yield. On contrary to our results, Mahmoud et al. (2011) reported there were no significant differences in the number of pods per plant under water regime treatments in both seasons. Current results showed that, increasing pod per plant can increase the seed yield, which is consistent with Sultan et al. (2012) who found significantly positive correlation of pods per plant with seed yield, plant height and branches per plant.

Weight of 100 seeds: seed weight is a known component of yield, which reflects relationship between source and sinks of photosynthetic during pod filling stage and it is where compensation for earlier losses of pods may occur, thus enhancing the final yield (Dantuma , Thompson, 1983). In current results, to access high seed with suitable weight, irrigation in flowering and pod-filing is necessary; however, for seed filling, the last irrigation is necessary. In terminal tension i.e. post-flowering, it may weight of seed will be decreased. On contrary to current results, Mahmoud et al. (2011) reported the weight of 1000 seeds was not significantly affected by the different water regimes in both seasons. Shekhawat, Singhania (2005) reported that, Pods per plant, seeds per pod and 100-seed weight had the maximum positive impact on seed yield per plant. Gresta et al. (2013) reported that, seed weight showed negative correlation with galactomannan content, but positive correlation with protein content, which our report with respect to positive relation with increasing protein is in agreement with Gresta et al. (2013).

Seed yield: The seed yield of any crop is the integrated results of number of physiological processes. Seed yield was significantly and positively correlated with biological yield, number of clusters and pods per plant and plant height. The results showed that late sowing date and I4 showed the most seed yield and impact interaction of G3 (RGC1066) with I4 are more suitable treatment in relation to yield parameters. Shekhawat, Singhania (2005) indicated that pods per plant, branches per plant, 100-seed weight and plant height had direct and positive effect on seed yield. In another study, Kumar, Ram (2015) suggested that number of pods per plant and plant height are the key traits for selection to yield improvement in guar.

Protein and Galactomannan: some studies revealed that seed yield and gum content has positive correlation, while seed weight has negative correlation with gum content (Pathak et al., 2011).

Materials and methods

Plant Material and Experimental Design

The experiment was conducted in an experimental field at Darrehshar region (latitude 37°8" min and Longitude 47°27"), Ilam, Iran, in 2015 -2016. The seeds of three genotypes (RGC1031, RGC 936 and RGC1066 as G1, G2, and G3, respectively) were provided from Rajhestan Agricultural Research Institute, India. The seeds were sown in $(5m \times 5m)$ with a density of 25 seeds / m2 (50 cm between rows, 8 cm between plants). Two sowing date at 15 June (D1) and 30 July (D2) were randomized in the main plots. Four irrigation regimes (one time irrigation: after sowing (I1), twice irrigation: after sowing and flowering (I2), three times irrigation: after sowing, flowering and seed formation (I3), and four times irrigation: after sowing, flowering, seed formation and before harvesting (I4) were managed in subplot and three genotypes (G1, G2, and G3) in sub-sub plot. Flood irrigation was done for any plots base on designed plan. The amount of applied water was recorded for any plot. The crop was kept weed free by hand weeding at 21 and 33 DAS (day after sowing) in the two seasons. No chemical fertilizers were used before sowing.

Soil texture and environmental condition

Soil texture was loam-clay type and no chemical fertilizer has been used during growth cycle. Also, there was no rainfall during the growth period of the guar, and the average temperature has been reported 32 $^\circ$ C during the growth cycle.

Measurement of traits

Plant height, the number of branch per plant, number of pod per branch, number of pod per plant, number of seed per pod, the number of seed per plant, pod length and seed weight were recorded. For measuring of each trait, three samples were harvested by hand and recorded precisely.

Guar gum manual purification and seed components determination

The guar endosperms were separated using the protocol reported by (Sabahelkheir et al., 2012). First, guar seeds were

soaked in distilled water for 10-12 hours, to imbibe water. The outer layer (hull) was removed easily and the medium layer (endosperm) was opened into two to separate the inner portion (the germ). Each part (hull, germ, and endosperm) was weighed separately until constant weight was reached. These were oven dried at a temperature 100°C and ground.

Crude protein

Crude protein was determined according to AOAC (1983) using a Kjeldahl system. 2 gram seed powder was weighted, then 7 ml of sulphuric acid were added to it. Afterward, 5 ml of hydrogen peroxide were added. Digestion of the sample was carried out at 420°C for 40 min. After cooling, the digested sample was diluted by adding 25 ml of distilled water. 25 ml of boric acid (4%) were used. The tube containing the sample was connected to the distilling unit 50 ml of NaOH was dispensed. Distillation was carried out for about 10 min. The receiver flask was then titrated against hydro chloric acid 0.1M. The crude protein percentage was calculated.

Statistical analysis

Differences between treatments were determined by using a three-way ANOVA model, with multiple mean comparisons according to the Duncan multiple tests at 1% level, SPSS.

Conclusion

The finding of this study provides new information about effect of sowing date and irrigation regimes on guar cultivation in Iran climates, Ilam province. Because of short life cycle of guar among all crop, it can be considered as an alternative crop during 3-4 month. As results shows, late sowing date (late of July) is suitable time for guar cultivation in this climate. Approximately irrigation regimes were significant differences in all parameters. This show that turns and amount of irrigation for high yield is important. Genotypes had no effect on much parameters. I4 is the best irrigation regimes in all parameters except in protein content and galactomannan amount and interaction of G3 (RGC1066) with I4 is the suitable treatment in relation to yield attributes. In future is needed to focus on irrigation amount, adaptation of more genotypes, plant density and so on to obtain the suitable condition for guar cultivation in Iran. So, current results indicated that guar can be an alternative crop and compatible with semi-arid climate of Iran.

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