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Original Article

Effect of Irrigation Interval, N Fertilizer Rate and Plant Density on Yield and Chemical Composition of Fenugreek (*Trigonella foenum-gracum* L.)

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Abstract

In order to study the effect of irrigation interval, N fertilization and plant density on yield and yield components of fenugreek, a split-split-plot experiment was conducted in Darmian, Iran in spring 2010. The experiment was conducted in randomized complete block design with three replications. The main plots were subjected to different irrigation intervals of 5 and 10 days. The sub-plots were treated with N fertilization at three different rates of 0, 75 and 150 kg N ha^{-1} from urea source and the sub-sub-plots were assigned with three varied plant density levels of 22, 33 and 66 plants m^{-2} respectively. Analysis of variance showed that the effect of irrigation interval on all measured traits was not significant. Also application of 150 kg N ha^{-1} had 33.9 and 66.1% higher pod number per plant, 37.5 and 68.1% higher pod number per m^2, 31.7 and 67.1% higher seed yield, 31.7 and 65.9% higher single-plant seed yield, 31.1 and 68.9% higher single-plant biomass yield and 30.6 and 67.8% higher biological yield than the application of 75 and 0 kg N ha^{-1}, respectively. Moreover, the results showed that increase in population from 22 to 33 plants m^{-2} decreased single-plant seed and biomass yields by 32.1 and 33.6%, respectively. Conclusively, considering the results of the present study and the importance of water saving, it is recommended to use an irrigation interval of 10 days with the application of 150 kg N ha^{-1} and a population of 66 plants m^{-2} for the cultivation of fenugreek in Darmian, Iran.

Key words: *Trigonella foenum-gracum* L., Irrigation interval, Population, Yield.

Introduction

Iran has a rich source of medicinal plants and is regarded as one of the best regions for the cultivation of these plants in the world due to its conducive climate, geography and background, but in comparison with other countries despite having high potentials and salient history, unfortunately their cultivation is not allotted the needed attention [1].

Fenugreek (*Trigonella* sp.) is native to western Asia (eastern Mediterranean) with 80 species out of which 30 species have been reportedly found in Iran [2,3]. Its seeds are medicinally used and some studies have shown that fenugreek contains nicotinic acid or niacin (vitamin B_3). Vitamin B_3 prevents Pellagra and atherosclerosis [4].

Because of inadequate annual precipitation and its inappropriate temporal and spatial distribution, most parts of Iran are arid and semi-arid and hence 95% of irrigation water is supplied by underground waters [5, 6]. Water deficiency has always been a limiting factor for the cultivation of most medicinal plants in Iran. The study of Shokhmgar [7] showed that irrigation had a significant effect on fenugreek seed yield and the highest and lowest seed yields were obtained at irrigation intervals of 4 and 12 days, respectively. Also, Lebaschi and Sharifieashorabad [8] studied the effects of irrigation treatments of 25, 50, 75 and 100% of field capacity on Isabgol, yarrow, marigold and chamomile and concluded that intensified drought stress decreased biological yield of the studied plants.

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In addition to available water, N is a key element in the structure of many compounds of cells and plays a crucial role in the growth and yield of plants. Shokhmgar [7] showed that N fertilization significantly affected fenugreek seed yield, and the highest seed yield was obtained with the application of 150 kg N ha^{-1}. In a study on the effect of three N fertilization rates (0, 80 and 120 kg N ha^{-1}) on yield of *Nigella sativa*, Khan [9] reported that the application of 80 kg N ha^{-1} produced the highest number of follicles per plant and seed yield and the application of 120 kg N ha^{-1} produced the highest 1000-seed weight, but the seed number per follicle was not affected by N fertilization. In addition, the application of 80 kg N ha^{-1} increased yield by 91% as compared with no-N fertilization treatment plot.

On the other hand, it should be noted that plant density is an important parameter influencing the yield of plants. There is an optimum plant density for each crop, over which assimilates are more utilized for vegetative growth and respiration increase rather than the reproductive growth. Also, under similar plant density conditions, although single-plant production increased, yield per area unit decreased [5, 10]. Sharma [11] indicated that the increase in plant density increased seed yield and decreased 1000-seed weight of fenugreek. Also, Gowda *et al.* [12] compared the effects of three sowing arrangements (15×15, 15×30 and 30×30 cm) on fenugreek and showed that the sowing arrangement of 15×15 produced the highest seed yield. Comparing the effect of four plant populations of 10, 20, 30 and 40 plants m^{-2} on fenugreek by Seghatoleslami and Ahmadi Bonakdar [13] indicated that plant density had no significant effect on 1000-seed weight and harvest index, but as the population was increased from 10 to 40 plants m^{-2}, total biomass increased from 67.3 to 142.8 g m^{-2}.

Given the importance of the study of medicinal plants, the current study was carried out to determine the optimum irrigation interval, N fertilization rate and plant density of fenugreek in Darmian, Iran.

Materials and Methods

The study was carried out in the seedling reservoir of Department of Natural and Water Resources of Darmian (Long. 59°55´56″ E., Lat. 32°56´50″ N., Alt. 1456 m) in the east of Birjand in 2010. The field had been left fallow in the previous year. The soil was sampled during field preparation operation in March 2010 from the depth of 0-40 cm for laboratory analysis the results of which are shown in Table 1. The plowing and levering was carried out by chisel plow. The furrows were created during late-March 2010 to prepare the field for sowing. According to soil analysis and recommended fertilization for fenugreek, the base fertilizer of phosphorous and potash was applied with the amount of 60 kg ha^{-1} K_2O from potassium sulfate source + 60 kg ha^{-1} phosphorous from golden biophosphate source (17% phosphorus) concurrent with making the furrows.

It was a split-split-plot experiment based on a randomized complete block design with three replications. The main plots were subjected to different irrigation internals of 5 and 10 days. The sub-plots were treated with N fertilization at three different rates of 0, 75 and 150 kg ha^{-1} and the sub-sub-plots were assigned with three varied plant density levels of 22, 33 and 66 plants m^{-2} respectively (with inter-plant spacing of 30 cm and inter-row spacing of 15, 10 and 5 cm). The furrows were 60 cm apart and the cultivation was carried out on both sides of them. Each experimental plot included six 6-m-long rows. The sub-sub-plots were 60 cm apart, the sub-plots were 120 cm apart, the main-plots were 180 cm apart and the replications were 2 m apart. The seeds were sown on April 9, 2010. The seeds had been disinfected with Carbendazim fungicide (2:1000) before sowing. Then, they were sown at an approximate depth of 0.5-1cm and were covered with soil. The plants were thinned at 4-5-leaf stage and the plots were weeded and fertilized with N (from urea source) at two stages, first at 4-5-leaf and second at 8-10-leaf stage. The plants were harvested during June 30-August 4 when 70-80% of the pods were completely ripe and the plants yellowed but they had not started to shed yet. Then, they were sown at an approximate depth of 0.5-1cm and were covered with soil. The plants were thinned at 4-5-leaf stage and the plots were weeded and fertilized with N (from urea source) at two stages, first at 4-5-leaf and second at 8-10-leaf stage. The plants were harvested during June 30-August 4 when 70-80% of the pods were completely ripe and the plants yellowed but they had not started to shed yet.

Table 1 Physical and chemical characteristics of study field at the depth of 0-40 cm

Soil Texture	OC (%)	Sand (%)	Silt (%)	Clay (%)	Total N (%)	Average P (%)	Average K (%)	pH	EC (ms/cm)
Silt clay	0.13	41.1	27.3	31.6	0.014	4.11	235	7.91	6.2

The plants were harvested from an area of 3 m^2 from the middle four rows of the plots and then, they were transferred to the laboratory in special bags. The measured traits included seed yield, total biomass, 1000-seed weight, harvest index, single-plant biomass and seed yield, pod number per plant and per m^2 and seed number per pod. After complete harvest and subsequently drying them in an oven, total biomass and seed yield were measured in terms of kg ha^{-1}. Single-plant biomass and seed yield were calculated by dividing the yield of the sampling area by the number of plants of the same area. Seed number per pod was calculated by dividing the number of seeds by the number of pods, and pod number per plant was calculated by dividing the number of pods by the number of plants of the sampling area. Harvest index was obtained by the following equation:

$$HI = \frac{SY}{BY} \times 100$$

Where HI, SY and BY depict harvest index, seed yield and biological yield, respectively.
The data were analyzed by statistical software SAS and the means were compared with Duncan Multiple Range Test at 5% probability level.

Results

Yield and yield components

The analysis of variance results showed that irrigation intervals had no significant effects on yield and its components, but N fertilizer significantly affected pod number per plant, pod number per m^2 and seed yield at 1% level. In addition, the change in plant density caused significant difference significant differences in pod number per plant at the 1% level. Also, the interaction between factors only significantly affected seed yield but had no significant effect on other traits (Table 2).
The comparisons of means showed that despite no significant differences in the traits related to yield and yield components of fenugreek between irrigation intervals of 5 and 10 days, the increase in irrigation interval from 5 to 10 days decreased pod number per plant, pod number per m^2, seed number per pod and seed yield by 21.9, 22, 6.5 and 22%, respectively (Table 3).
In total, the increase in N rate from 0 to 150 kg ha^{-1} had positive, significant effect on seed yield and some yield components, where at N rate of 150 kg ha^{-1} as compared with N rates of 75 and 0 kg ha^{-1}, the comparisons of means showed that pod number per plant increased by 33.9 and 66.1%, pod number per m^2 increased by 37.5 and 68.1% and seed yield increased by 31.7 and 67.1%, respectively (Table 3).
Also, the comparisons of means indicated that pod number per plant significantly increased by 32 and 62.3% as the density was increased from 22 to 33 and 66 plants m^{-2}, respectively (Table 3).
The interaction between factors only significantly affected seed yield but had no significant effect on the other traits (Table 2). The highest fruit yield (1362.83 kg ha^{-1}) was obtained at an irrigation interval of 5 days with 150 kg N ha^{-1} application treatment and the lowest fruit yield (575.18 kg ha^{-1}) was obtained at an irrigation interval of 10 days without nitrogen application treatment (Fig 1). Fig 2 and Fig 3 show interaction effects of nitrogen x plant density and irrigation x plant density on seed yield.

Biological yield and harvest index

According to the results of analysis of variance, the change in irrigation interval and plant density did not significantly affect biological yield and harvest index, but the impact of N fertilization was significant on biological yield at 1% level while its impact was not significant on harvest index. Moreover, the interactions between factors did not significantly affect these two traits (Table 2).
However, the comparisons of means indicated that as the irrigation interval was increased from 5 to 10 days, biomass yield and harvest index decreased 18.2% and 7.4%, respectively (Table 3).
Biological yield significantly increased with the increase in N rate, exhibiting 30.6 and 67.8% higher yield at an N rate of 150 kg ha^{-1} than that at N rates of 75 and 0 kg ha^{-1}, respectively (Table 3).
As comparisons of means showed, the highest biological yield (2738.4 kg ha^{-1}) and harvest index (37.8%) was obtained at the population of 66 plants m^{-2}, but it did not show significant difference in other population levels (Table 3).
Moreover, the interactions between factors did not significantly affect these two traits (Table 2). Fig. 4, Fig. 5 and Fig. 6 show interaction effects of irrigation x nitrogen, nitrogen x plant density and irrigation x plant density on seed yield.

Single-plant seed and biomass yield

The results indicated that irrigation interval did not significantly impact single-plant seed and biomass yield, but these two traits were significantly affected by applied N rates and plant density at the 1% level (Table 2).

Table 2 Results of analysis of variance for yield and yield components of fenugreek as affected by irrigation interval, N fertilization and plant density

Sources of variation	df	Means of squares								
		Pod no. plant^{-1}	Pod no. m^{-2}	Seed no. pod^{-1}	1000-seed weight	Seed yield	Biological yield	Harvest index	Single-plant seed yield	Single-plant biomass yield
Replication	2	1733.130^{ns}	2027452.9^{ns}	6.741^{ns}	0.674^{ns}	1926137.605^{ns}	12680372.463^{ns}	36.727^{ns}	16.468^{ns}	112.290^{ns}
Irrigation interval (I)	1	785.852^{ns}	961333.79^{ns}	5.352^{ns}	1.340^{ns}	779055.014^{ns}	3527688.963^{ns}	111.836^{ns}	6.332^{ns}	25.992^{ns}
Main error	2	205.352	245096.13	1.407	0.613	215307.364	1490309.796	41.816	1.963	13.8674
Nitrogen rate (N)	2	1065.907**	1325671.79**	0.907^{ns}	0.888^{ns}	10749732.544^{ns}	7795147.241**	9.159^{ns}	8.774**	67.120**
I × N	2	105.907^{ns}	129263.35^{ns}	0.019^{ns}	1.298^{ns}	45692.167**	460029.241^{ns}	10.506^{ns}	0.279^{ns}	3.094^{ns}
Sub-error	8	66.769	52170.24	0.546	0.0608	34976.929	217132.907	27.461	0.531	2.886
Plant density (P)	2	3585.130**	97756.68^{ns}	0.574^{ns}	0.112^{ns}	82744.935^{ns}	360393.574^{ns}	7.921^{ns}	29.037**	221.690**
I × P	2	50.463^{ns}	2531.13^{ns}	0.241^{ns}	0.300^{ns}	6557.425^{ns}	70392.907^{ns}	10.342^{ns}	0.463^{ns}	0.809^{ns}
N × P	4	63.130^{ns}	1644.99^{ns}	0.935 ^{ns}	0.268^{ns}	8648.027^{ns}	27040.019^{ns}	3.042^{ns}	0.588^{ns}	6.187^{ns}
I × N × P	4	39.519^{ns}	28394.6^{ns}	0.157^{ns}	0.109^{ns}	24381.82^{ns}	139810.185^{ns}	6.167^{ns}	0.281^{ns}	1.477^{ns}
Sub-sub-error	24	61.315	41321.44	0.306	0.210	39518.249	205961.009	5.696	0.642	3.832
C.V. (%)		25.23	18.91	5.86	4.83	20.49	17.62	6.43	28.6	26.22

*, ** and ^{ns} show significant at 5 and 1% level and non-significance

Table 3 Results of comparisons of means for yield components of fenugreek as affected by irrigation interval, N fertilization and plant density

Treatment	Pod no. plant^{-1}	Pod no. m^{-2}	Seed no. pod^{-1}	1000-seed weight (g)	Seed yield (kg ha^{-1})	Biological yield (kg ha^{-1})	Harvest index (%)	Single-plant seed yield (g)	Single-plant biomass yield (g)
Irrigation interval (day)									
5	34.85 a	1208.63 a	6.74 a	9.33 a	1090.08 a	2830.67 a	38.53 a	3.14 a	8.16 a
10	27.22 a	941.78 a	9.11 a	9.65 a	849.86 a	2319.48 a	35.65 a	2.45 a	6.77 a
Nitrogen rate (kg N ha^{-1})									
0	23.28 b	795.11 c	9.67 a	9.31 a	729.7 c	1939.17 c	36.28 a	2.11 c	5.60 c
75	31.17 a	1093.56 b	9.22 a	9.41 a	960.92 b	2532.78 b	37.59 a	2.78 b	7.34 b
150	38.67 a	1336.94 a	9.39 a	9.74 a	1219.29 a	3253.28 a	37.42 a	3.502 a	9.46 a
density (plants.m^{-2})									
22	45.28 a	1020 a	9.22 a	9.4 a	922.10 a	2495.00 a	36.45 a	4.08 a	11.04 a
33	30.78 b	1046.72 a	9.56 a	9.52 a	940.25 a	2491.28 a	37.06 a	2.76 b	7.33 b
66	17.06 c	1158.89 a	9.5 a	9.54 a	1047.60 a	2738.44 a	37.77 a	1.54 c	4.03 c

Means with the same letter(s) in each column did not show significant difference at 5% level on the basis of Duncan Tes

Nonetheless, it should be noted that the increase in interval irrigation from 5 to 10 days led to 22 and 17% loss of single-plant seed and biomass yields, respectively (Table 3). Also, as N rate was increased from 0 to 75 and 150 kg ha^{-1}, single-plant seed yield significantly increased by 31.7 and 65.9% and single-plant biomass yield increased by 31.1 and 68.9%, respectively, and N fertilization rate of 150 kg ha^{-1} produced the highest single-plant seed and biomass yields (3.5 and 9.46 g, respectively) (Table 3).

The comparisons of means showed that as the population was increased from 22 to 33 plants m^{-2}, single-plant seed and biomass yields were decreased by 32.1 and 33.6%, respectively. In addition, as the population was increased up to 66 plants m^{-2}, single-plant seed and biomass yields decreased by 62.2 and 63.5%, respectively (Table 3).

Discussion

Although irrigation interval did not have a significant impact on yield traits of fenugreek in the current study, it should be taken into consideration that some of them noticeably decreased as the amount of available water dropped. Also, the results of the studies of Safikhani *et al.* [14] on *Deracocephalum moldavica* L. and Koocheki *et al*. [6] on fennel showed non-significant effect of irrigation interval on 1000-seed weight which is in agreement with the current study.

However, Shokhmgar [7] indicated that as the irrigation interval was increased, pod number per plant and seed number per pod significantly decreased. Non-significant differences in yield and yield components of fenugreek between irrigation intervals of 5 and 10 days could be related to the suitable amount and distribution of precipitation (as compared with the previous years) during the growing season of fenugreek and partial supply of plant water demand. However, the considerable difference in yield components under two irrigation levels showed that it is very likely that the decrease in the irrigation interval from 10 to 5 days considerably increases yield and yield components of fenugreek under the climate of the study location which usually experiences limited spring precipitation with still unsuitable distribution.

The 66.1% increase in pod number per plant as affected by the increase in N rate from 0 to 150 kg ha^{-1} could be associated with the positive effect of the fertilization on vegetative growth of fenugreek and the resulting increase in leaf area index and duration which increased plant height, branch number per plant and photosynthetic capacity of plants and paved the way for the production of more pods per plant and hence, the increase in pod number per m^2. Also, in a study on the effect of four fertilization rates of 0, 50, 150 and 200 kg N ha^{-1} on fenugreek, Shokhmgar [7] reported the positive effect of fertilization on pod number per plant and per m^2.

It seems that 1000-seed weight is one of the traits which is under a strict genetic control and enjoys a high heritability and thus, it is less affected by environmental parameters [15]. Also, in a study on the effect of the application of 0, 50, 100 and 150 kg N ha^{-1} on yield of *Nigella sativa*, Modi [16] concluded that N fertilization increased biological and seed yield, but it did not affect 1000-seed weight.

As the results showed, that seed number per pod of fenugreek was more affected by genetic parameters because the changes in irrigation, N and plant density had no significant effect on it and its numerical value showed slight fluctuations in different levels of irrigation, N and plant density.

The increase in fenugreek seed and biological yield resulting from N fertilization is an indicator of appropriate N absorption by plants which improves their vegetative growth and better utilization of solar radiation due to the increase in leaf area duration and index; and since fenugreek is an indeterminate plant, as vegetative growth and pod number per plant increase, its yield increases. Under the current study, it can be said that the highest effect of N on seed yield was through improving the potential of producing pods per plant which shows that pod number per plant is the most sensitive yield component. The studies of Husain *et al*. [17] and Whittaker [18] on soybean produced the same results.

The non-significant effect of N fertilization on harvest index in spite of its effect on seed and biological yield under the conditions of the current study implies that the decrease or increase in applied N rate equally affected seed and biological yield of fenugreek because the condition for producing a higher seed yield is the existence of enough foliage signifying higher biological yield. Additionally, lower vegetative growth decreases seed and biological yield under N-deficiency treatment and conversely, seed and biological yield increases due to optimum vegetative growth under adequate N fertilizer availability which results in partial constancy of harvest index. Also, the results of the study of Singh *et al.* [19] and Hasanzade [1] on sunflower and Taylor [20] on canola indicated non-significant effect of the increase N fertilizer rate on harvest index which is in concurrence with the results of the current study.

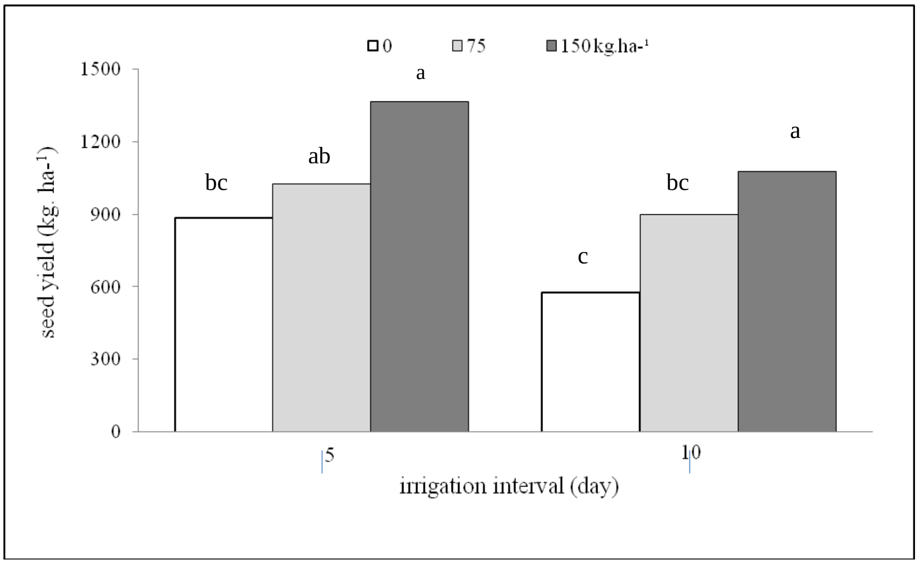

Fig 1 Interaction of irrigation and nitrogen on seed yield of fenugreek

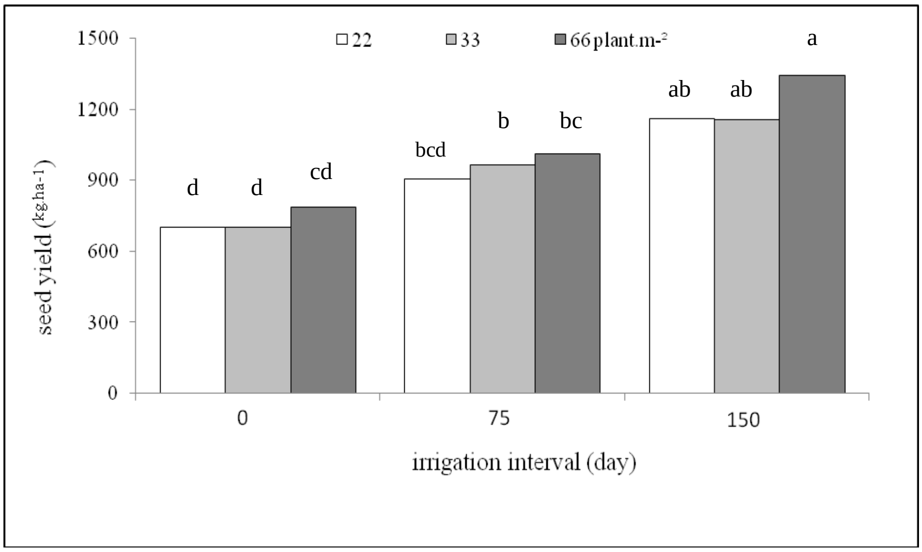

Fig 2 Interaction effect nitrogen and plant density on seed yield of fenugreek

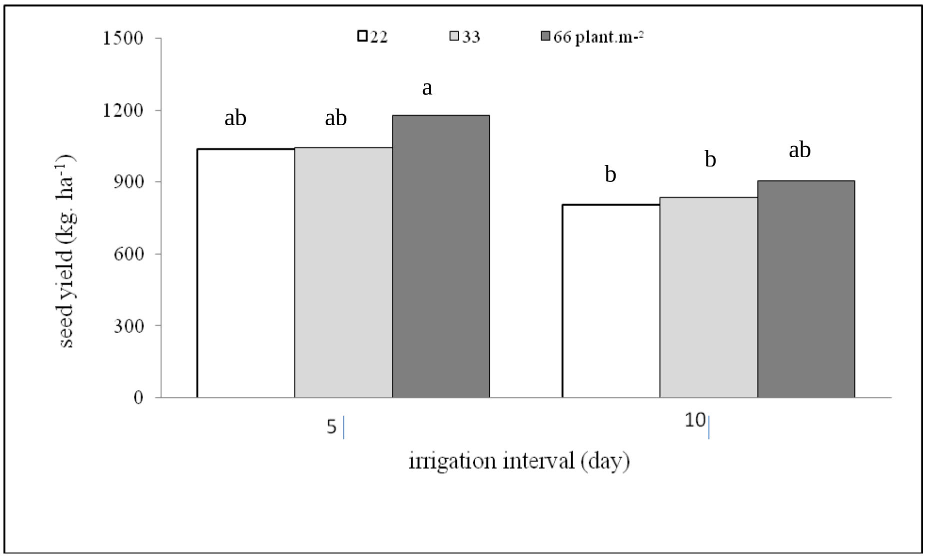

Fig 3 Interaction effect of irrigation and plant density on seed yield of fenugreek

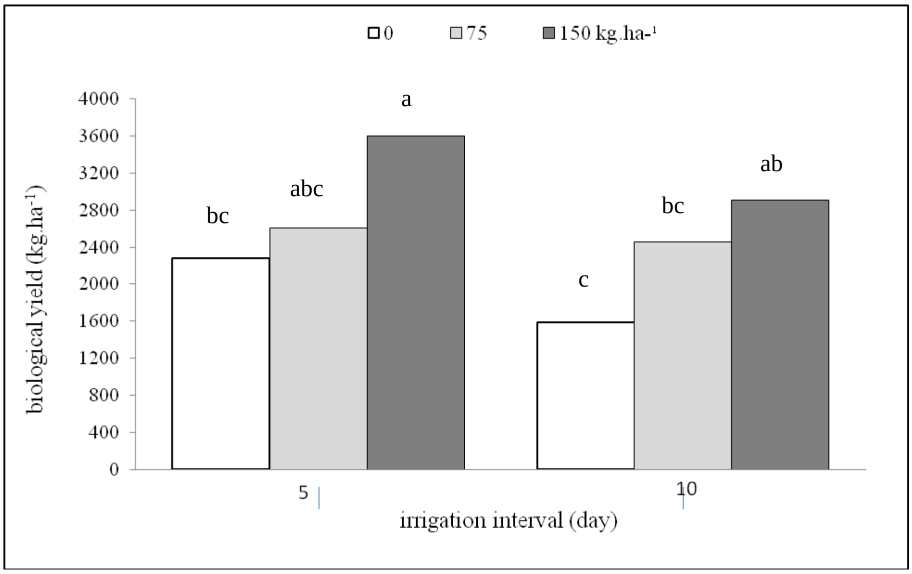

Fig 4 Interaction effect of irrigation and nitrogen on biological yield of fenugreek

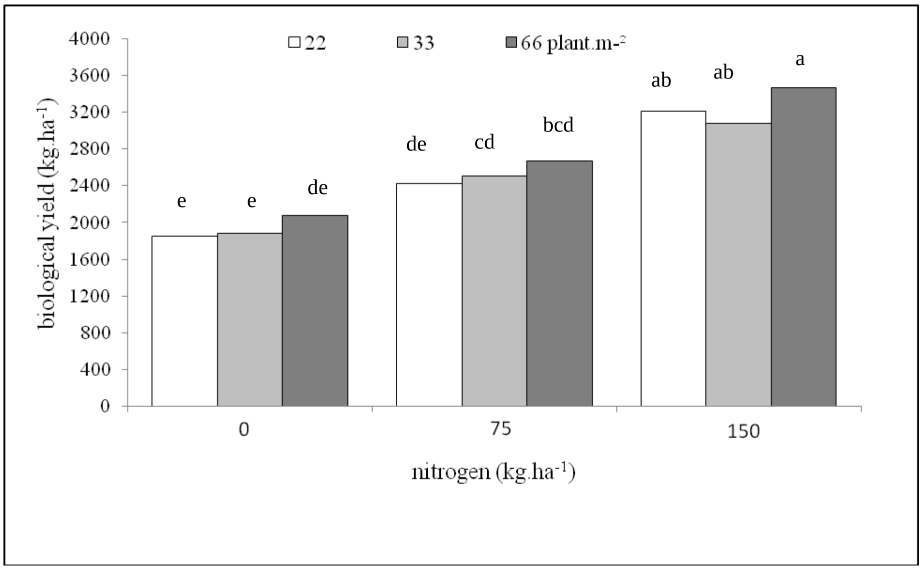

Fig 5 Interaction effect of irrigation and nitrogen on biological yield of fenugreek

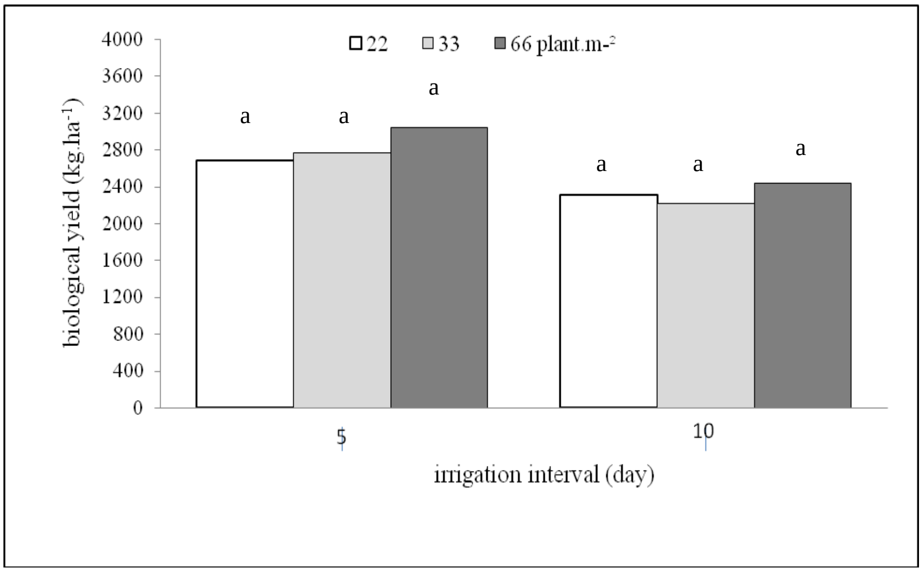

Fig 6 Interaction effect of irrigation and plant density on biological yield of fenugreek

The positive and significant effect of the increase in N fertilization on single-plant seed and biomass yields could be related to the less inter-plant competition of fenugreek on accessing N and consequently, optimum utilization of it for vegetative and reproductive growth.

According to the results, seed yield did not show significant changes as plant density per area unit was changed (Table 3) The main reason was the lack of considerable differences in pod number per m^2 and other yield components (seed number per pod and 1000-seed weight) under different plant densities. In other words, although the increase in plant density significantly decreased pod number per plant, but higher plant density compensated the decrease in pod number per plant and in total, it non-significantly increased pod number and seed yield per area unit.

The non-significant effect of plant density on seed number per pod, 1000-seed weight and seed yield of fenugreek has been also observed in the study of Seghatoleslami and Ahmadi Bonakdar [13]. Also, the studies of Rosalind *et al*. [21] on bean and Shirtliffe and Johnston [22] on soybean showed that 1000-seed weight did not significantly change in different plant densities. However, it should be noted that the studies of Singh *et al.* [23], Gowda *et al.* [12] and Yadav *et al*. [24] on the effect of different plant densities on seed yield of fenugreek (with more density difference) depicted that it was not able to compensate the decrease in plant density per area unit by increasing pod number and main branch number per plant which resulted in a significant decrease of yield with the decrease in plant density.

The reason for the 62.3% loss of pod number per plant as plant density was increased from 22 to 66 plants m^{-2} could have been the intensified inter-plant competition on environmental parameters (light, water, space) which brought about the decrease in the production of assimilates and its partitioning, and the loss of photosynthesis resulted in the reduction of vegetative growth (plant height and branch number per plant) and finally, the loss of pod number per plant. At the same time, it should be taken into account that higher number of plants per area unit leads to more efficient use of environmental parameters and so, more pods are produced per m^2. Also, with comparative row spacing studies of fenugreek *viz.* 22.5 and 30 cm, Singh *et al.* [23] showed that higher row spacing (lower density) increased pod number per plant. The results of the current study are consistent with the results of the studies of Seghatoleslami and Mousavi [25] and Martin and Deo [26] on *Calendula officinalis*.

As plant density is increased, biological yield of crops usually increases. Under the conditions of the current study, although the increase in plant density numerically increased biological yield, the reason for the non-significant difference between different density levels could be related to greater growth of plant (producing higher foliage, height and pods) at lower densities due to the less inter-plant competition and having greater share in resources, specially solar radiation and water, so that biomass yield per plant was 2.64 times as great at the minimum density (22 plants m^{-2}) as at the maximum density (66 plants m^{-2}) (Table 3). In other words, it can be said that although density was tripled, single-plant biomass yield proportionally decreased with the increase in plant density and in total, no significant difference was observed in biological yields of different plant densities.

Also, the increase in single-plant seed and biomass yields under lower plant densities could be related to the possibility of using more resources by plants on one hand and to the increase in net photosynthesis due to the absence of shading and the decrease in respiration on the other hand.

Conclusions

The results revealed that the irrigation interval of 10 days can be recommended for the cultivation of fenugreek in the study region because it is important to save water and increase efficiency of the water utilization. Besides, the application of 150 kg N ha^{-1} can be recommended because it not only stimulates vegetative growth and increases leaf area index and duration but also boosts photosynthetic potential and economical yield. In addition, it can be recommended to use the population of 66 plants m^{-2} because it ensures faster formation of canopy and optimum utilization of solar radiation.

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