Iran Agricultural Research (2021) 40(1) 71-82

Research Article

Shiraz University

Selection of fenugreek (*Trigonella foenum-graecum* L.) landraces for fall planting and freezing tolerance

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DOI: 10.22099/IAR.2021.40312.1433

ARTICLE INFO

***Article history*:**

Received **19 April 2021**
Accepted **4 July 2021**
Available online **21 September 2021**

***Keywords*:**

Cold acclimation
Seed yield
Stepwise regression
Survival rate

ABSTRACT- Cultivation of medicinal plants, such as fenugreek, is important for producing healthy food and preparing raw materials for herbal medicines. Determining the appropriate planting date and tolerance of fenugreek to low temperatures can help to gain more economic yield. The effects of cold and fall planting on the Iranian fenugreek (*Trigonella foenum-graceum* L.) landraces were evaluated under controlled and field conditions in two years (2013-2015). Ten fenugreek landraces including Azari, Ardestan, Tall, Dwarf, Shiraz, Shirvan, Mashhad, Neyshabour, Hamedan, and Hendi landraces were planted on five planting dates (September 14, October 15, and November 14 in 2013; March 6 and April 4 in 2014). These landraces were also planted on four planting dates (September and October in 2013; March and April in 2014) in pots and were transferred to a thermogradient freezer to apply frost temperatures (-6^{o}C, -9^{o}C, -12^{o}C, and -15^{o}C). Four landraces including Dwarf, Shirvan, Mashhad, Neyshabour landraces were planted on five planting dates similar to the first year in the second year. Shirvan landrace showed more tolerance to freezing temperatures. Evaluation of survival rate showed that the controlled grown landraces had higher survival rates than the field-grown landraces. Mashhad (55%) and Shirvan (44%) had the highest and lowest yield reduction rates as a result of delaying the planting date from September to April. The branch number/plant in the first year of the field experiment and 1000-seed weight (TSW) in the second year had the most important impact on the seed yield. The most suitable sowing dates for the highest seed yield were October 15 and November 14. Shirvan landrace had the highest seed yield in all the planting dates in both years, and it can be recommended for cultivation in the regions.

INTRODUCTION

Fenugreek (*Trigonella foenum-graecum* L.) is an eatable and spicy annual herbaceous plant, which has pharmaceutical uses (lowers cholesterol and blood sugar levels). In addition, it is used as a source of raw materials in the pharmaceutical industry, especially for production of steroid hormones (Zandi et al., 2015). Moreover, this plant is considered a cover crop with the ability to fix nitrogen and increase soil fertility (Dadrasan et al., 2015). It can also be used in short-term rotation to produce forage and silage for animal feed (Zandi et al., 2015). Fenugreek seed extract contains antioxidant compounds and protects cellular structures from oxidative damages (Neelakantan et al., 2014).

Today, however, the need to cultivate medicinal plants, including fenugreek, is becoming more essential for producing healthy food and raw materials for herbal medicines, as well as remediating soil. These plants do not have a much-cultivated area. Fenugreek is cultivated in all parts of Iran with a small cultivation area (about 400 ha) and a seed yield of about 800 kg ha^{-1} (Mehrafarin et al., 2011).

Planting date is the most important factor affecting the growth, yield, and quality of fenugreek (Anitha et al., 2016). In fact, by selecting the appropriate planting date, the plant phenological stages can be adjusted according to the environmental conditions to achieve higher yields. Spring planting causes plants to be exposed to heat and drought during the growing season, and irrigation is essential for higher yield. On the other hand, water deficit is the most important factor in reducing the crop yield in arid and semi-arid regions (Karimzadeh Soureshjani et al., 2019). Therefore, in recent years, more attention has been paid to the early planting of plants in the spring or their fall-winter planting dates. The results of a previous study has been shown that the yield of some plants, such as broad bean (*Vicia faba* L.), in fall cultivation is often more than that in the common spring cultivation date and yield stability is also more in this planting date (Duzdemir and Ece, 2011). However, cold tolerance in the plants is essential for successful fall planting; otherwise, the plant may be damaged and even died in some cases (Yadav, 2010).

The planting date of fenugreek is a determining factor in its seed yield. Early or delayed planting may impair growth, yield, and crop quality. It has been shown that early planting leads to early flowering and may have a negative effect on yield if the genotype does not have a tolerance to cold and freezing stress (Marlene Pérez-barbeito et al., 2008). On the other hand, delayed planting might reduce the plant growth and yield due to the reduced length of the growing season (Bhutia and Sharangi, 2016). It has also been reported that improper planting dates cause the critical stages of the plant life cycle, such as flowering, to coincide with unfavorable temperatures, which reduces the number of fertile flowers, thus reducing the seed yield (Adamsen and Coffelt, 2005).

It has been shown that delayed planting dates reduce the fertile pod, seed weight, and seed yield of fenugreek (Anitha et al., 2016). Delayed wheat (*Triticum aestivum* L.) cultivars (spring instead of fall) reduced the seed yield by about 16% (Wyzińska and Grabiński, 2018). Increased seed yield of fall planting date compared with spring planting date has been reported in other plants such as faba bean (Confalone et al., 2013; Duzdemir and Ece, 2011). It has also been reported that earlier planting dates of winter faba bean increases the biomass and the harvest index (HI) due to the increment of the node number, green area index, and absorbed photosynthetically active radiation (Neugschwandtner et al., 2019). Winter pea (*Pisum sativum*) cultivars also have higher plant height, HI, and seed yield than spring cultivars (Neugschwandtner et al., 2019).

The results of another study have indicated that delayed planting dates (July) of two soybean cultivars (*Glycine max* L.) decreased the number of branches, seed weight, and seed yield compared with early planting dates (April 3, June 23, and July 30); however, the reductions in these varieties were not similar (Tang et al., 2006). By another research, it has been shown that winter planting dates increase seed and biomass yields of peas and beans (*Phaseolus vulgaris*). Moreover, yield stability in winter peas was higher than that in spring peas (Urbatzka et al., 2011). However, the risk of winter cold in some or all of the years may reduce yield stability (Neugschwandtner et al., 2019). For example, a negative correlation has been found between winter mortality and seed yield in winter wheat (Sharma et al., 2014).

Since Iran has arid and semi-arid climate conditions, water deficit during plant growth is one of the main problems in spring planting dates. Therefore, considering the benefits of fall cultivation for providing the plant's water needs by fall, winter, and early spring rainfall, this experiment was performed aiming to (i) evaluate the response of Iranian fenugreek landraces to different fall and winter planting dates and (ii) to determine the effect of freezing temperatures on these landraces.

MATERIALS AND METHODS

Study location

This study was conducted during the two growing seasons in 2013-2014 at the research field of Ferdowsi University of Mashhad, Iran (36°15’ N, 59°28’ E, 985 m altitude). The weather data of the study location during the growing seasons in 2013-2015 are shown in Table 1.

Experiment Execution Process

Each experiment was conducted as a split-plot arranged in a randomized complete block design with three replications. Five planting dates (September 14, October 15, and November 14 in 2013; March 6 and April 4 in 2014) were used as the main plots and ten Iranian fenugreek landraces including Azari, Ardestan, Tall, Dwarf, Shiraz, Shirvan, Mashhad, Neyshabour, Hamedan, Hendi landraces were used as sub-plots. The characteristics of fenugreek landraces collection centers are shown in Table 2.

A pot experiment was also performed to evaluate the effect of freezing temperatures (-6^{o}C, -9^{o}C, -12^{o}C, and -15^{o}C) on the Iranian fenugreek landraces used in this study. In this regard, the fenugreek landraces were planted in the uniform pots on four different dates (September 14 and October 15 in 2013; March 6 and April 4 in 2014). Then, the pots were placed outdoors to get acclimation to the cold. The freezing temperatures were applied using a thermogradient freezer on January 5 for September and October planting dates and on May 5 for March and April planting dates. At the start of the test, the freezer temperature was 5^{o}C, and then it decreased to 2^{o}C per hour after placing the pots. To prevent the formation of ice nuclei in the seedlings, the plants were sprayed with an ice-nucleating active bacteria suspension at -2.5^{o}C to -3^{o}C (Nezami et al., 2012). The pots were kept for 1 h in each temperature treatment and then removed from the freezer. To reduce the melting rate of the plants, the pots were immediately transferred to a cold room with a temperature of 4 ± 1^{o}C and stored there for 24 h (Nezami et al., 2012). The pots were then placed in the greenhouse for 4 weeks to allow the plants to recover.

In 2014-2015, based on the seed yield in the 2013-2014 growing season and the survival percentage in the second step, four landraces including Dwarf, Shirvan, Mashhad, and Neyshabour landraces were selected and planted on five planting dates similar to the first step.

The plant density was 40 plants per square meter (70 cm row spacing and two cultivation lines on each row with a 35-cm distance), and each plot was 5.25 m^2 (2.1 m * 2.5 m). The distances between the main plots, subplots, and replication were 2, 1, and 2 m, respectively. The plants were irrigated every seven days until physiological maturity.

To calculate the survival percentage (S), the plant number was counted after establishment (n), and then the number of plants was recounted (m) after winter (in the field experiments) or after recovery (in the pot experiment). The survival percentage was calculated using Equation 1.

$$S(\%) = \frac{m}{n} * 100 \qquad (1)$$

In the field experiments, one square meter of each plot was harvested at physiological maturity. After oven drying (at 75 ^{o}C for 48 h) and threshing the pods, seed yield was measured, and HI was calculated using Equation 2. Yield components including the pod number and 1000-seed weight (TSW) were also determined using 10 randomly selected plants in each plot.

$$H \ (\%) = \frac{g \quad y}{a \quad g \quad b} * 100 \qquad (2)$$

Statistical Analysis

Analysis of variance was performed after the normality test of data using the Kolmogorov-Smirnov test. The least significant difference (LSD) test was also used to compare means at *P*-value = 0.05. All the analyses were performed using Minitab17 software (https:// www.minitab.com/en-us/products/minitab/).The Pearson correlation coefficients of the yield components were also calculated at *P*-value = 0.05. In addition, stepwise regression was used to determine the component with the greatest effect on the seed yield.

Table 1. The number of frosty nights, absolute monthly minimum temperature, monthly rainfall and total below zero temperatures during the growing seasons in 2013-2015

Date	The number of Frosty nights*		Absolute monthly minimum temperature (°C)		Monthly precipitation (mm)		Total temperatures below 0°C	
	2013-2014	2014-2015	2013-2014	2014-2015	2013-2014	2014-2015	2013-2014	2014-2015
Aug-Sep	-	-	11.6	11.9	0	0	-	-
Sep-Oct	-	-	5.2	3.2	11.4	16.7	-	-
Oct-Nov	2	4	-3.5	-3.3	16.8	30.6	-4.8	-7.1
Nov-Dec	11	12	-5.2	-4.9	8.7	21.8	-22.4	-27.9
Dec-Jan	25	12	-9	-2.3	0.6	20.9	-114.1	-10.2
Jan-Feb	20	10	-17.4	-4	6.5	38.7	-141.9	-25.2
Feb-Mar	10	14	-3.6	-4	1	41.6	-18.5	-23.3
Mar-Apr	3	4	-3.4	-4.5	67.4	26.1	-5.1	-14.1
Apr-May	-	-	10.4	7.6	30.5	23.8	-	-
May-Jun	-	-	12.1	12.2	22	0.3	-	-
Jun-Jul	-	-	16.9	18	0	0	-	-

*Based on the number of nights with a minimum temperature of less than 0°C.

Table 2. The characteristics of the locations in Iran where fenugreek landraces used in this study were gathered and evaluated during two growing seasons in Mashhad in 2013-2015.

Landraces	Location[1]	Geographical Coordinates[2]	Landraces	Location	Geographical Coordinates*
Azari	East Azerbaijan Province	$38^0$7’N/46^o20’E/+1351	Shirvan	North Khorasan Province	$37^0$40’N/57^o93’E/+1097
Ardestan	Isfahan province	$33^0$20’N/52^o25’E/+1238	Mashhad	Razavi Khorasan province	$36^0$20’N/59^o35’E/+1050
Tall	Gilan province	$37^0$8’N/50^o18’E/-19	Neyshabour	Razavi Khorasan province	$36^0$10’N/58^o50’E/+1250
Dwarf	Gilan province	$37^0$8’N/50^o18’E/-19	Hamedan	Hamedan province	$35^0$00’N/49^o00’E/+1850

Shiraz	Fars province	29[0]37'N/52º32'E/+1585	Hendi	Isfahan province	32[0]39'N/51º43'E/+1570

[1]Gathering Location [2]latitude/longitude/altitude

Statistical Analysis

Analysis of variance was performed after the normality test of data using the Kolmogorov-Smirnov test. The least significant difference (LSD) test was also used to compare means at *P*-value = 0.05. All the analyses were performed using Minitab17 software (https:// www.minitab.com/en-us/products/minitab/).The Pearson correlation coefficients of the yield components were also calculated at *P*-value = 0.05. In addition, stepwise regression was used to determine the component with the greatest effect on the seed yield.

RESULTS AND DISCUSSION

Weather Conditions

In the first year (2013-2014), there were 13 days with temperatures below 0ºC, and the absolute minimum temperature was -17.4ºC in this year (Table 1). It was also snowing for three days. The total precipitation during the growing season in 2013-2014 was 165 mm. In the second year (2014-2015), there were three days with temperatures below 0ºC, and the absolute minimum temperature was -4.9ºC. It was also snowing for seven days. The total precipitation during the growing season in 2014-2015 was 220 mm. The weather was slightly warmer in the second year.

Plant Survival Rate

Plant survival rate was affected by the interaction between planting dates and fenugreek landraces in the field in both years (Tables 3 and 4). On the September planting date, Neyshabour, Mashhad, Dwarf, and Shirvan landraces had a higher survival rate, and the survival percentage rate of Azari and Ardestan was about 50%. However, Tall, Dwarf, Shiraz, and Hendi had a 30% lower survival percentage (Table 5). Delaying the planting date for two months (fall planting in November) increased the survival percentage of all the landraces; therefore, all the landraces had a survival percentage of 100% on the November planting date. On this date, the highest increment of survival compared with the Sep 14 planting date was found in Shiraz and tall landraces (93% and 84% more, respectively).

Evaluation of the survival percentage in both controlled and field experiments (2013-2014) showed that landraces with more survival in the controlled experiment had higher survival rates in field conditions. Hence, on the September planting date, Neyshabour and Mashhad landraces were capable of tolerating a temperature of -12 ºC at the controlled condition. In this temperature, Neyshabour and Mashhad landraces had 100% and 89% survival, respectively (Table 5).

Table 3. Analysis of variance of survival rates, plant height, branches number/plant, fertile pods/plant, 1000-seed weight (TSW), harvest index (HI) and seed yield of fenugreek landraces at different planting dates during the growing season in 2013-2014

		Mean of squares						
Sources of variation	DF	Survival rates	Height	Branches No/ plant	Fertile pods/plant	TSW	HI	Seed yield
Block	2	7ns	7ns	0.2ns	4ns	0.0003ns	4ns	70ns
Planting date (PD)	4	14666**	4427**	188.8**	2573**	1.6**	701**	21567**
Error a	8	5	7	0.3	6	0.04	22	301
Landrace (L)	9	1476**	702**	29.2**	115**	185.2**	537**	25343**
PD*L	36	701**	99**	2.9**	12ns	0.2**	270**	2368*
Error b	90	6	29	0.4	15	0.05	12	215

ns, * and **; non-significant, significant at *P*=0.05 and 0.01, respectively

Table 4. Analysis of variance of survival rates, plant height, branches number/plant, fertile pods/plant, 1000 seed weight (TSW), harvest index (HI) and seed yield of fenugreek landraces at different planting dates during the growing season in 2014-2015

		Mean of squares						
Source of variation	DF	Survival rates	Height	Branch No	Fertile pod/plant	TSW	HI	Seed yield
Block	2	0.9ns	5.9ns	0.04ns	2.6ns	0.1ns	5.0ns	118.7ns
Planting date (PD)	4	120.4**	1775.3**	98.1**	1127.7**	0.6**	97.0**	23488.7**
Error a	8	0.9	32.3	0.2	6.9	0.04	2.8	123.9
Landrace (L)	3	22.8**	1342.4**	66.4**	106.4**	274.2**	215.8**	22884.4**
PD*L	12	22.8**	890.8*	4.0**	8.1ns	0.05ns	65.1**	716.9ns
Error b	30	1.5	34.8	0.4	14.8	0.1	12.7	385.0

* and **, ns; significant at *P*=0.05 and 0.01, and no significant, respectively

Table 5. Effect of planting dates on survival rates of fenugreek landraces during the two growing seasons in 2013-2015

Planting date	Landraces	The survival rate in controlled condition (%)				The survival rate in field condition (%)	
		-6	-9	-12	-15	2013-2014	2014-2015
September 14	Azari	100	100	0	0	53	-
	Ardestan	100	100	0	0	49	-
	Tall	100	100	0	0	16	-
	Dwarf	100	100	0	0	80	94
	Shiraz	100	100	0	0	7	-
	Shirvan	100	100	0	0	76	85
	Mashhad	100	100	89	0	86	93
	Neyshabour	100	100	100	0	100	100
	Hamedan	100	100	0	0	28	-
	Hendi	100	100	0	0	20	-
October 15	Azari	100	100	39	0	60	-
	Ardestan	100	100	50	0	73	-
	Tall	100	100	0	0	59	-
	Dwarf	100	100	61	0	81	100
	Shiraz	100	100	0	0	70	-
	Shirvan	100	100	100	0	91	100
	Mashhad	100	100	100	0	93	100
	Neyshabour	100	100	100	0	100	100
	Hamedan	100	100	100	0	49	-
	Hendi	100	100	100	0	46	-
November 14	Azari	-	-	-	-	100	-
	Ardestan	-	-	-	-	100	-
	Tall	-	-	-	-	100	-
	Dwarf	-	-	-	-	100	100
	Shiraz	-	-	-	-	100	-
	Shirvan	-	-	-	-	100	100
	Mashhad	-	-	-	-	100	100
	Neyshabour	-	-	-	-	100	100
	Hamedan	-	-	-	-	100	-
	Hendi	-	-	-	-	100	-
March 6	Azari	100	0	0	0	100	-
	Ardestan	100	0	0	0	100	-
	Tall	100	100	0	0	100	-
	Dwarf	100	100	0	0	100	100
	Shiraz	100	100	0	0	100	-
	Shirvan	100	100	0	0	100	100
	Mashhad	100	100	0	0	100	100
	Neyshabour	100	100	0	0	100	100
	Hamedan	100	100	0	0	100	-
	Hendi	100	100	0	0	100	-
April 4	Azari	100	100	0	0	100	-
	Ardestan	100	100	0	0	100	-
	Tall	100	100	0	0	100	-
	Dwarf	100	100	0	0	100	100
	Shiraz	100	100	0	0	100	-
	Shirvan	100	100	0	0	100	100
	Mashhad	100	100	0	0	100	100
	Neyshabour	100	100	0	0	100	100
	Hamedan	100	100	0	0	100	-
	Hendi	100	100	0	0	100	-
$LSD_{0.05}$		2.1				3.5	2

LSD: Least Significant Difference at $P \leq 0.05$ probability level

The results showed that the effect of the interaction between the planting dates, landraces, and temperatures was significant on the plant survival rate under controlled conditions (Table 6). A drop in temperature to -12^{o}C under controlled conditions led to the death of Tall, Shiraz, Hendi, and Hamedan landraces, while Neyshabour, Mashhad, and Shirvan landraces had a 100% survival rate. The last three landraces also had the highest survival percentage in field conditions in the harsh winter during the 2013-2014 growing season (Table 5).

In spring planting dates, all the landraces had a survival percentage of 100% in field conditions. The minimum temperature in April was about -4^{o}C on these planting dates. However, in controlled conditions, since the plants were not sufficiently acclimated to cold, they could not survive in temperatures below -9^{o}C.

Table 6. Analysis of variance of survival of fenugreek landraces at different planting dates and temperatures in the controlled condition.

Source of variation	DF	Survival mean square
Planting date (PD)	3	2658**
Landrace (L)	9	722**
Temperature (T)	6	194185**
PD*L	27	368**
PD*T	18	1798**
L*T	54	644**
PD*L*T	162	394**
Error	560	1.7

* and **; ns;, significant at *P*=0.05 and 0.01, and no significant, respectively.

In the second year, Shirvan, Mashhad, and Dwarf landraces were damaged in the September planting date due to mild cold in winter, and their survival rate decreased by 15%, 7%, and 6%, respectively. In contrast, in the other planting dates, winter conditions did not damage the plants and the survival of all the landraces was 100% (Table 5). Due to extreme cold conditions (decrease in temperature to -17 ^{o}C), the survival percentage of all the landraces, except for Neyshabour, was lower in the first year of the field experiment (Table 5).

It has been reported that low temperatures reduce the plant's biosynthetic activity, prevent the normal function of physiological processes, and may cause permanent damage that eventually leads to death (Joshi et al., 2007). It has been found that faba bean and winter pea varieties enter the hibernation stage at the 2-4 leaves stage and have a survival rate of over 80% in both experimental years of Neugschwandtner et al., (2019) study. In this study it was observed that in the controlled experiment, since the plants cultivated in September and October and the plants cultivated in March were at a higher growth stage when cold stress was applied, the tested plants showed lower tolerance to low temperatures compared with the plants cultivated in April. The winter survival percentage of faba bean varieties during the three growing seasons in 2010-2013 revealed that in 2012-2013, the plants had a higher survival percentage due to the milder winter (Neugschwandtner et al., 2019). The results of the present study also showed that fall planting dates lead to more survival percentage compared with spring planting dates. This is due to this fact that in the fall planting dates, plants have ample opportunity to experience low temperatures and become acclimated to cold. On the contrary, plants had no opportunity to experience low temperatures on spring planting dates; hence, cold acclimation did not happen.

Plant Height

The effect of the interaction between planting dates and fenugreek landraces on plant height was significant (Tables 3 and 4). Delayed planting reduced the height of the plants of all landraces compared to those of the plants sowed in the September planting date (Table 7). Hamadan (26%), Ardestan (45%), Ardestan (49%), and Neyshabour landraces had the highest plant height increment in October, November (fall planting), March, and April planting dates, compared with the September planting date in the first year of the field experiment. Neyshabour landrace also had the highest height reduction in all planting dates compared with the September planting date in the first yield of the field experiments (Table 7). Bastidas et al. (2008) reported that delayed planting of soybean significantly reduced plant height in both tested years. It has been shown that delayed planting reduces the growth of the plant due to the reduction of the growth period and unfavorable temperatures (El Sherif and Khattab, 2016).

Branch Number per Plant

The branch number per plant was significantly affected by the interaction of planting dates and landraces (Tables 3 and 4). Delayed planting reduced the branch number of the plants of all the landraces in the first year of the field experiment. The highest branch number reduction per Plant was observed in Shiraz and Neyshabour landraces in the October planting date (fall) compared with the September planting date (Table 7). Moreover, in the second year, the highest branch number reduction per Plant was seen in Mashhad landrace in October (fall planting date), March, and April (spring planting dates) and in Dwarf landrace in the November planting date compared with the September planting date. It has been reported that unfavorable temperatures during the critical stages of the plant's life cycle and the plant's inability to make optimal use of environmental factors such as water and solar radiation, which result from improper planting dates, reduce plant growth (Caliskan et al., 2008). Decreasing the number of branches of plants is a component of reducing vegetative growth of the plants and is a symptom of declined growth (Linkemer et al. 1998).

Pod Number per Plant

The fertile pod number was significantly affected by the planting date and landraces (Tables 3 and 4). Delayed planting reduced fertile pods in both years; in the first year, the fertile pods were reduced by 5%, 41%, and 46% in November, March, and April planting dates, respectively, compared with those in the September planting date (Fig. 1). Fertile pod reduction due to delayed planting was 11%, 43%, and 49% in November (fall), March, and April (spring) planting dates, respectively, compared with those in the September planting date (Fig. 1)). However, the fertile pod number per plant did not show a significant reduction in the October planting date with those in the September planting date in both years (Fig. 1). The highest fertile pod number/plant was observed in the Neyshabour landrace in both years, which was not significantly different from those of Shirvan and Mashhad landraces (Fig. 2).

Table 7. Effect of planting dates on plant height and branches/plant of fenugreek landraces during the two growing years (2013-2015)

Planting date	Landrace	Plant height (cm)		Branches No/plant	
September 14	Azari	61.2	-	9.7	-
	Ardestan	71.8	-	9.9	-
	Tall	73.4	-	8.6	-
	Dwarf	44.7	45.4	8.6	8.9
	Shiraz	66.3	-	11	-
	Shirvan	75.3	74.1	11.3	10.9
	Mashhad	70.7	77.5	13.7	13.4
	Neyshabour	79.2	76	15.3	15.3
	Hamedan	73.3	-	11.7	-
	Hendi	53.3	-	11.6	-
October 15	Azari	55.7	-	9.7	-
	Ardestan	54.9	-	9.6	-
	Tall	72	-	8.5	-
	Dwarf	40.3	40.1	8.6	8.8
	Shiraz	55	-	9.6	-
	Shirvan	65.1	60.9	10.6	11.2
	Mashhad	65.7	69.9	13.7	10.7
	Neyshabour	63.4	61.3	14.7	15.5
	Hamedan	54.6	-	11.2	-
	Hendi	49	-	10.8	-
November 14	Azari	53.8	-	8.9	-
	Ardestan	39.2	-	9.6	-
	Tall	53.5	-	7.7	-
	Dwarf	42.1	36.2	7.7	6.8
	Shiraz	50.2	-	9.5	-
	Shirvan	67.9	51.9	9.7	9.7
	Mashhad	59.8	57.9	10.7	10.9
	Neyshabour	59.9	49.8	14.7	14.3
	Hamedan	49.2	-	10.8	-
	Hendi	54.3	-	10.8	-
March 6	Azari	48.9	-	7.1	-
	Ardestan	36.8	-	6.8	-
	Tall	38.1	-	5.7	-
	Dwarf	27.8	34.6	5.8	5.6
	Shiraz	35.8	-	6.5	-
	Shirvan	53.5	47.5	5.7	5.9
	Mashhad	48.8	48.1	6.5	6.8
	Neyshabour	45.3	38.9	7.6	7.9
	Hamedan	48.2	-	6.1	-
	Hendi	45.8	-	6.5	-
April 4	Azari	44.1	-	6.1	-
	Ardestan	36.5	-	6.4	-
	Tall	33.9	-	5	-
	Dwarf	25.1	31.4	5.2	5.2
	Shiraz	30.6	-	5.4	-
	Shirvan	40.6	46.7	5.3	5.2
	Mashhad	38.2	39.9	6.2	6.1
	Neyshabour	33.8	34.8	6.9	7.4
	Hamedan	40.2	-	5.9	-
	Hendi	37.3	-	5.6	-
$LSD_{0.05}$		8.7	9.8	1	1.1

LSD: Least Significant Difference in $P \leq 0.05$ probability level

Anitha et al. (2016) reported that delaying the planting date from October to December reduced the fertile pods by about 20% in all tested fenugreek varieties in their study. Delayed planting causes the sensitive stages of plant growth to be exposed to inappropriate temperatures; hence the green area and leaf area duration will be reduced, which leads to the reduction of the plant's photosynthetic capacity.

With the decrease of photosynthetic assimilation, fewer flowers are formed per plant, the flower abortion increases, and eventually the number of pods per plant decreases (Tsimba et al., 2013; Baogang et al., 2014).

1000-Seed Weight (TSW)

TSW was significantly affected by the interaction between planting dates and fenugreek landrace in the first year (Table 3) and in the second year (Table 4). Although delayed planting reduced the TSW of all the landraces (Table 8), some differences were found. The highest TSW reductions (13% and 9%) due to delaying the planting date from September to April were observed in Dwarf landrace in the first and second years of the field experiment, respectively. On the contrary, delaying the planting date from September to April had no significant effect on the TSW of Shiraz landrace in the first and second years of the field experiment (Table 8).

It has been previously reported that delaying the planting date from October to December reduces the TSW of fenugreek varieties by 12-16% in India (Anitha et al., 2016). Delayed planting reduces the length of plant growth and accelerates the completion of reproductive stages. Moreover, lower photosynthetic assimilates are exhibited by the seeds, and therefore, the weight of the seeds is reduced (Nielsen et al., 2002).

Harvest Index (HI)

HI was significantly affected by the interaction between planting dates and fenugreek landraces (Tables 3 and 4). The HI of most landraces was decreased due to delaying the planting date from September to April (Table 8). The highest and lowest HI reductions were found, respectively, in Dwarf (16%) and Shiraz (9%) landraces in the first year and Mashhad (14%) and Dwarf (2%) landraces in the second year. However, the HI of Neyshabour landrace slightly increased in April planting compared with September planting in the first (1%) and the second (3%) year of the field experiment.

The flowering of fall-sown plants begins in late April, which was about a month earlier than that of spring-sown plants. The high temperature during flowering is a limiting factor for spring crops (Bodner et al., 2018). Earlier planting dates prevent the flowering and pod filling stages from being exposed to heat and drought stress and prevent yield reduction (Sita et al., 2017). Furthermore, the duration of the developmental stages in fall crops is higher than in spring crops. In other words, the length of vegetative and reproductive growth stages such as the flowering period and the pod filling period is longer in fall crops which can increase the seed yield and HI (Bodner et al., 2018). In the current study, there was a significant positive correlation between the fertile pods/plant and HI (Table 9), and an increase in the number of fertile pods increased the HI.

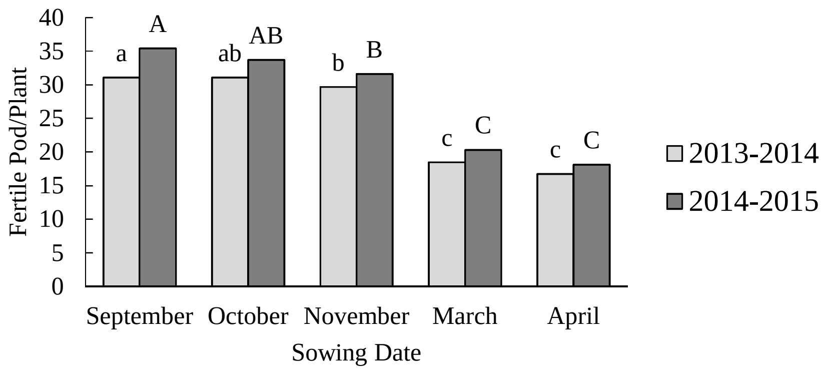

Fig. 1. The fertile pods number per plant of fenugreek landraces in different sowing dates during 2013-2014 (lower case letters), and 2014-2015 (upper case letters) growing seasons. Means followed by similar letters are not significantly different by LSD test ($P \leq 0.05$).

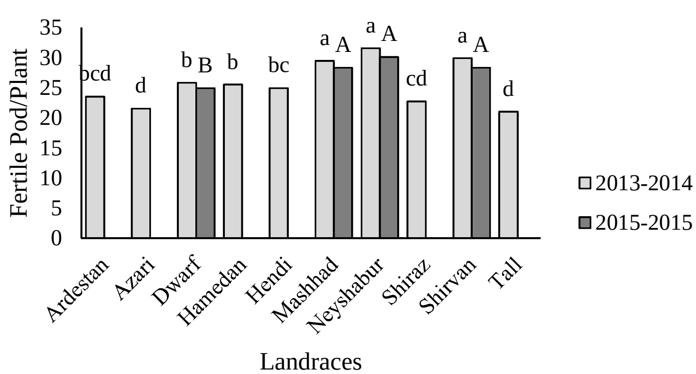

Fig. 2. The fertile pods number per plant of fenugreek landraces in 2014-2015 (lower case letters), and 2014-2015 (upper case letters) growing seasons. Means followed by similar letters are not significantly different by LSD test ($P \leq 0.05$).

Table 8. Effect of planting dates on 1000-seed weight, seed yield and HI of fenugreek landraces during the two growing seasons in 2013-2015.

Planting date	Landraces	1000 seed weight (g)		HI (%)		Seed yield ($g.m^{-1}$)	
		2013-2014	2014-2015	2013-2014	2014-2015	2013-2014	2014-2015
September 14	Azari	19.2	-	31	-	89	-
	Ardestan	14.6	-	30	-	66	-
	Tall	8.70	-	33	-	17	-
	Dwarf	9.00	8.50	40	32	119	113
	Shiraz	15.2	-	31	-	14	-
	Shirvan	18.6	18.4	47	40	216	212
	Mashhad	12.5	12.5	47	46	199	213
	Neyshabour	10.8	10.8	32	30	209	198
	Hamedan	14.4	-	40	-	59	-
	Hendi	14.1	-	35	-	39	-
October 15	Azari	18.9	-	29	-	95	-
	Ardestan	14.7	-	29	-	89	-
	Tall	8.60	-	28	-	54	-
	Dwarf	8.90	8.30	36	32	105	122
	Shiraz	15.1	-	26	-	94	-
	Shirvan	18.3	18.5	42	43	198	233
	Mashhad	12.5	12.3	44	41	186	189
	Neyshabour	10.5	10.9	31	37	168	194
	Hamedan	14.3	-	32	-	83	-
	Hendi	14.1	-	32	-	77	-
November 14	Azari	Azari	18.0	-	27	-	131
	Ardestan	Ardestan	14.4	-	29	-	127
	Tall	Tall	8.80	-	28	-	95
	Dwarf	Dwarf	8.90	8.10	38	35	159
	Shiraz	Shiraz	14.9	-	29	-	116
	Shirvan	Shirvan	18.1	18.5	45	44	229
	Mashhad	Mashhad	12.3	12.3	34	34	169
	Neyshabour	Neyshabour	10.8	10.8	33	34	171
	Hamedan	Hamedan	14.1	-	29	-	151
	Hendi	Hendi	14.0	-	28	-	145
March 6	Azari	17.9	-	17	-	72	-
	Ardestan	14.2	-	15	-	60	-
	Tall	8.40	-	19	-	54	-
	Dwarf	8.00	8.10	32	32	71	71
	Shiraz	14.8	-	23	-	91	-
	Shirvan	18.2	18.1	42	43	164	165
	Mashhad	11.9	12.1	34	30	112	100
	Neyshabour	10.4	10.8	29	26	109	99
	Hamedan	14.0	-	24	-	83	-
	Hendi	13.9	-	23	-	82	-
April 4	Azari	18.8	-	19	-	82	-
	Ardestan	14.0	-	19	-	63	-
	Tall	8.30	-	19	-	49	-
	Dwarf	7.80	7.80	24	30	54	57
	Shiraz	15.1	-	22	-	79	-
	Shirvan	18.1	17.8	33	31	123	118
	Mashhad	12.0	12.1	32	32	94	95
	Neyshabour	10.3	10.4	33	33	99	98
	Hamedan	13.9	-	26	-	81	-
	Hendi	13.8	-	25	-	79	-
LSD $_{0.05}$		0.4	0.5	6	6	24	24

LSD: Least Significant Difference in $P \leq 0.05$ probability level

Seed Yield

The interaction between planting dates and landraces had a significant effect on fenugreek seed yield in both years (Tables 3 and 4). Shirvan, Mashhad, and Neyshabour landraces had the highest seed yield in fall plantings (September and October) in the first year (Table 8). Dwarf, Shirvan, Mashhad, and Neyshabour landraces had the highest seed yield (12%, 7%, 8%, and 20%, respectively) in the October planting date in the first year, but the seed yield of Mashhad and Neyshabour landraces decreased (11% and 2%, respectively) in the second year compared with the September planting date. Delaying the planting date from September to November reduced the seed yield of Mashhad and Neyshabour landraces in the first year (15% and 18%, respectively) and in the second year (22% and 11%, respectively), while the seed yield of other landraces increased in November compared with September planting dates mainly due to the increment of the survival rate (Table 5). The seed yield of Mashhad and Neyshabour landraces reduced in November compared with the September planting date in both years, whereas the seed yield of the other landraces increased. The seed yield of all the landraces reduced in March and April in the second year compared with the September planting date. In the first year, the seed yield of other landraces, except Tall, Shiraz, Hamedan, and Hendi landraces, also decreased in these two planting dates (Table 8). Mashhad, Neyshabour, and Shirvan landraces had the highest seed yield in both years. The most suitable planting date for these landraces for better seed yield was September, and for the other landraces was November. The seed yield was positively correlated with the survival percentage, height, branch number, fertile pods number, seed weight, and HI in both years (Table 9).

Evaluation of the effect of the five fall planting dates on five fenugreek varieties showed that delaying the planting date from October 15 to December 15 reduced the seed yield by 48%. Anitha et al. (2016) found that the highest reduction (53%) in the seed yield of fenugreek occurred in delayed planting of Co-2 and Rmt-1 genotypes. In the current study, increasing the length of vegetative and reproductive growth of the plant increased the plant height and the number of branches. Thereby, increasing biomass production, due to the increment of the growth duration, enhances the number of fertile pods per plant and the seed yield. It is also important that sensitive stages of the plant do not encounter unfavorable weather conditions.

The results of the stepwise regression analysis showed that the branch number in the first year of the field experiment and TSW in the second year of the field experiment had the most important influence on the seed yield. Moreover, the plant height had the least effect on the seed yield in both years (Table 10). In other words, increasing the branch number and TSW had the greatest effect on increasing the seed yield. In fact, by preventing the reduction of these two factors, the reduction of seed yield due to improper planting dates can be greatly inhibited.

CONCLUSIONS

The results of this study showed that the tested fenugreek landraces had a higher branch number per plant, TSW, and fertile pods per plant in the fall compared with spring planting dates. Neyshabour, Mashhad, Dwarf, and Shirvan landraces showed higher survival rates on the September planting date, while the survival percentage rate of Azari and Ardestan was about 50%. On the other hand, Tall, Dwarf, Shiraz, and Hendi had a 30% lower survival percentage. Mashhad, Neyshabour, and Shirvan landraces had the highest seed yield in both years tested in this study. The best planting date for these landraces to gain a higher seed yield was September and for the others was November. Evaluating physiological, biochemical, and molecular underlying mechanisms in the next studies can help us understand fenugreek's response to freezing stress.

Table 9. Correlation matrix of fenugreek landraces traits in 2013-2014 (above diameter) and 2014-2015 (below diameter).

Traits	Survival rate	Plant height	Branches No./plant	Fertile pods/plant	TSW	Seed yield	Harvest index
Survival	**1**	-0.49**	-0.34*	-0.35*	-0.09	0.44**	-0.23
Plant height	-0.47**	**1**	0.75**	0.73**	0.23	0.38*	0.55**
Branches no/plant	-0.33**	0.73**	**1**	0.89**	0.06	0.51**	0.52**
Fertile pods/plant	-0.33**	0.69**	0.86**	**1**	0.02	0.60**	0.79**
TSW	-0.09	0.21*	0.06	0.02	**1**	0.21	0.06
Seed yield	0.43**	0.38**	0.51**	0.61**	0.23**	**1**	0.63**
HI	-0.22*	0.52**	0.51**	0.80**	0.06	0.61**	**1**

*and **: significant in the probability levels of 5%, and 1%, respectively.

Table 10. Result of stepwise regression analysis for seed yield of fenugreek landraces in two years (2013-2015)

	2013-2014				2014-2015			
Step	Variable entered	Parameter Estimate	Cumulative R-Square	p-Value	Variable entered	Parameter Estimate	Cumulative R-Square	p-Value
1	Fertile pods/plant	2.04	0.375	**	Fertile pods/plant	3.10	0.732	**
2	Survival rare	1.60	0.830	**	TSW	6.40	0.964	**
3	TSW	3.61	0.908	**	Survival rate	1.25	0.972	**
4	Harvest index	2.19	0.925	**	Branch no/plant	4.42	0.979	**
5	Plant height	0.55	0.942	**	Harvest index	1.47	0.988	**
6	Branches no/plant	4.01	0.948	**	Plant height	0.21	0.988	ns
	Intercept	-260.28		**	Intercept	-245.80		**

ns and **: non-significant and significant in the probability levels of 1%, respectively.

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تحقیقات کشاورزی ایران (۱۴۰۰) ۴۰(۱) ۷۱-۸۲

مقاله علمی- پژوهشی

انتخاب توده‌های شنبلیله (*Trigonella foenum-graecum* L.) برای کشت پاییزه و تحمل به تنش یخزدگی

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اطلاعات مقاله

تاریخچه مقاله:

تاریخ دریافت: ۱۴۰۰/۱/۳۰

تاریخ پذیرش: ۱۴۰۰/۴/۱۳

تاریخ دسترسی:۱۴۰۰/ ۶/۳۰

واژه‌های کلیدی:

خوسرمایی

عملکرد دانه

رگرسیون گام به گام

درصد بقا

چکیده- کشت گیاهان دارویی از جمله شنبلیله جهت تأمین غذای سالم و تولید مواد اولیه تولید داروهای گیاهی حائظ اهمیت است. تعیین تاریخ کاشت مناسب و میزان تحمل به دماهای پایین در شنبلیله می‌تواند به افزایش عملکرد اقتصادی منجر شود. در این پژوهش اثر تنش سرما در شرایط کنترل شده و اثر کشت پاییزه بر توده‌های شنبلیله ایران در شرایط مزرعه طی در دو سال (۹۶-۱۳۹۴) مورد ارزیابی قرار گرفت. در سال نخست ده توده شنبلیله شامل توده های آذری، اردستان، پابلند، پاکوتاه، شیراز، شیروان، مشهد، نیشابور، همدان و هندی در پنج تاریخ (۲۳ شهریور، ۲۳ مهر، ۲۳ آبان، ۱۵ اسفند و ۱۵ فروردین) در مزرعه کاشت شدند. این ده توده همچنین در چهار تاریخ (۲۳ شهریور، ۲۳ مهر، ۱۵ اسفند و ۱۵ فروردین) در گلدان کشت شدند. سپس برای اعمال دماهای یخزدگی (۶-، ۹-، ۱۲- و ۱۵- درجه سلسیوس) در فریزر ترموگرادیان قرار گرفتند. در سال دوم چهار توده شامل توده های پاکوتاه، شیروان، مشهد، و نیشابو) انتخاب شدند و در همان پنج تاریخ پیشین در مزرعه کشت شدند. نتایج نشان داد که توده شیروان بیشترین تحمل را به دماهای یخزدگی داشت. ارزیابی میزان بقا در هر دو شرایط مزرعه و کنترل شده نشان داد که توده‌هایی که در شرایط کنترل شده بقای بالایی داشتند، در شرایط مزرعه نیز بقای بالایی نشان دادند. بیشترین و کمترین درصد کاهش عملکرد دانه در اثر تأخیر در کشت از شهریور به فروردین به ترتیب در توده مشهد (۵۵ درصد) و توده شیروان (۴۴ درصد) وجود داشت. نتایج تجزیه رگرسیون گام به گام نشان داد که تعداد شاخه‌های فرعی در سال اول و وزن هزار دانه در سال دوم بیشترین تأثیر را در تغییرات عملکرد دانه داشتند. همچنین نتایج نشان داد توده شیروان در همه تاریخ‌های کشت و در هر دو سال انجام آزمایش در مزرعه، بیشترین عملکرد دانه را به خود اختصاص داد و از این رو برای کشت در شرایط مشابه قابل توصیه می‌باشد.