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Effect of water deficit on grain yield, yield components of Narrow-leaved plantain (*Plantago lanceolata* L.)

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ABSTRACT

Aims: Drought is the primary factor limiting plant growth and productivity in many world regions. Narrow-leaved plantain (*Plantago lanceolata* L.) is widely used as a medicinal plant to treat some diseases. This study examined the response of four local ecotypes of P. *lanceolata* to different drought stress levels in a field experiment.

Materials & Methods: A split-plot design was conducted based on a randomized complete block design (RCBD) with three replications at the Research Institute of Forests and Rangelands farm, Karaj, Iran, in 2018. The main factor was drought stress at three levels (D1 is regular irrigation, D2 is drought stress after the flowering stage with supplemental irrigation at the filling stage, and D3 is stop irrigation after the flowering. The second factor contained four ecotypes: G1-Arak, G2-Khoramabad, G3-Meshkin1, and G4-Meshkin2. Data were collected and statistically analyzed for grain yield and yield components.

Findings: Results showed a significant effect of drought stress and ecotype for all traits except for root lengths ($p<0.05, 0.01$). The ecotype × drought stress interaction effects were significant for spikes number per plant, leaves number per plant, leaf width, and plant height. The mean values of grain yield in D1, D2, and D3 were 729.41, 660.81, and 595.95 $kg.h^{-1}$, respectively.

Conclusion: The highest grain yield with a value of 670.92 $kg.h^{-1}$ was obtained in G1-Arak. This ecotype also had higher grain yields in mild and severe stress than the other ecotypes and was recommended for breeding improved varieties.

Keywords: Plantago lanceolate; Drought stress; Regression; Path analysis.

CITATION LINKS

[1] Daneshian, J., Ahmadi, M., Kalantarahmadi, S.A. Stability ... [2] Mahajan S., Tuteja N. Cold, salinity and drought stresses: ... [3] Wang W.X., Vinocur B., Shoseyov O., Altman A. Biotechnolog ... [4] Farooq M., Wahid A., Kobayashi N., Fujita D., Basra S.M.A. ... [5] Barnabas B., Jäger K., Fehér A. The effect of drought and ... [6] Belayet H.M., Rahman W., Rahman M.N., Noorul Anwar A.H.M., ... [7] Xiaolong J., Chunyan H. and Xudan G. Physicochemical prope ... [8] Goncalves S., Romano A. The medicinal potential of plants ... [9] Janighorban, M. Plantaginaceae. In: Assadi M. (Ed.), Flora ... [10] Jankovi T., Zduni G., Beara I. Comparative study of some p ... [11] Kreitschitz, A., Kovalev, A., Gorb, S.N., 2016. "'Sticky I ... [12] Amininasab, S.S, Mahmoudi Otaghvari, A. and Nazifi, E. 2 ... [13] Temur C., Uslu, S. Effects of Plantain (*Plantago anceolata* ... [14] Oloumi M.M., Vosough D., Derakhshanfar A., Nematollahi M.H ... [15] Foster, L. Herbs in pastures. Development research in Brit ... [16] Bahadori, M. B., Sarikurkcu, C., Kocak, M. S., Calapoglu, ... [17] Pol M., Schmidtke K., Lewandowska, S. *Plantago lanceolata* ... [18] Kaswan, V., Kaushik, A., Devi, J., Joshi, A. and Ratan Mal ... [19] Steel R.G.D., Torrie J.H. Principles and procedures of sta ... [20] Levene, H. 1960. Robust tests for equality of variances in ... [21] Akhzari D., Ghasemi Aghbash F. Effect of Salinity and Drou ... [22] Saberi M., Shahriari A., Niknahad-Gharmakher H., Heshmati ... . [23] Najafi F., Rezvani Moghaddam P. Effects of irrigation regi ... [24] Tabrizi L.The effect of water stress and manure on yield, ... [25] Koocheki A., Tabrizi L., Nassiri Mahallati, M. Organic cul ... [26] Pirasteh-Anosheh H., Emam Y., Ashraf M., Foolad M.R. Exoge ... . [27] Blum A. 2005. Drought resistance, water-use efficiency, an ... [28] Neumann, P. M. Cropping mechanisms for crop plants in drou ... [29] Xue Q., Zhu Z., Musick J.T., Stewart, B.A., Dusek D.A. Roo ... [30] Koocheki A., Mokhtari V., Taherabadi Sh., Kalantari S. The ... [31] Patra D.D., Anwar M., Singh S., Prasad A., Singh D.V. Arom ... [32] Mousavinick M. Effect of drought stress and Sulphur fertil ... [33] Bernal M., Estiarte M., Penuelas, J. Drought advances spri ... [34] Roumani A., Biabani A., Rahemi Karizaki A., Gholamalipour ... [35] Ramroudi M., Galavi M., Siahsar B.A., Allahdo M. Effect of ... [36] Asgharipour, M., Rafiei, M. Intercropping of Isabgol (Plan ... [37] Toghraei A., Mirshekari B., Daneshian J., KazemiArbat H., ... [38] Pouryousef M., Mazaheri D., Chaiechi M.R., Rahimi A., Jafa ... [39] Rahimi A., Jahansoz M.R., Rahimian Mashhadi H. Effect of D ... [40] Bhagat N.R. Studies on Variation and association among see ... [41] Wright S. Correlation and causation. J. Agric. Res. 1921; ... [42] Dewey D.R., Lu K.H. A correlation and path coefficient ana ...

Introduction

Severe climate change has been a critical problem in recent years, and the forecast of drought and increasing temperature in many regions of Iran. It is necessary to achieve a strategy based on the Knowledge of practical methods to determine the increase in the yield and adaptability of crop species. One of the ways to deal with drought is to use drought-resistant cultivars [1].

Water scarcity is a major problem that drastically reduces crop production in the world's arable lands [2], and the crisis is severe in Iran. Drought stress can be defined as the lack of sufficient water required for normal growth and completion of the plant life cycle. The plant adapts to drought stress by inducing various physiological, biochemical, and morphological responses [3, 4]. The response of plants to drought stress depends on the severity and duration of stress, plant species, and the stage of stress occurrence [5]. Drought stress harms crops' emergence, growth, and production [6].

Plantago L. is a genus of medicinal plants in the Plantaginaceae family [7]. This family has around 275 species that grow annually and permanently [8]. In Iran, 25 species of *the Plantago* genus grow in many parts of the country [9]. *Plantago* has many uses, including raw materials for salads, soups, baking, and animal feed, to improve health and reduce antibiotic use [8]. Phytochemicals derived from the genus Plantago's root, leaf, and stem have shown medicinal potential [10].

Narrow-leaved plantain (*Plantago lanceolata* L.) is a perennial herb widely distributed worldwide in the rangeland and roadsides of temperate regions. It grows on a wide range of soils and is drought-resistant [11]. It grows abundantly in many parts of Iran [12]. Its seeds contain up to 30% mucilage, which swells up in the gut, acting as a bulk laxative [11].
It has been used for medicinal purposes to treat diseases such as wound healing, inflammation, cancer, respiratory system disorders, blood circulation, reproductive system, and digestive organs [13]. Phytochemicals in the root, leaf, and grain of *P. lanceolata* include iridoid glycosides, polyphenols, polysaccharides, and flavonoids, which have therapeutic potential [10]. It also treats upper respiratory tract, mouth, throat, and skin diseases [14]. Moreover, this herb is widely used as forage in Britain [15]. Bahadori *et al*., 2020 showed the potential of *P. lanceolata* for producing new food products and pharmaceuticals, especially for preventing and treating oxidative stress-induced damage [16]. Pol *et al*., 2021 in the cultivation of *P. lanceolate* in grasslands of central Europe, found its positive health effects on grazing animals [17].

The Knowledge of variability, and relationships between yield and yield components is essential for yield improvement through a selection program. Kaswan et al. 2018, found significant and positive correlations between seed yield with biological yield/plant, number of spikes/plant, number of effective tillers/plant, harvest index, seed weight/spike, and spike weight in *P. ovata* [18]

The available information about *P. lanceolata*'s response to drought stress is less than that of other species of *Plantago* in Iran. Although *P. lanceolata* is widely used as a medicinal plant, more information about its response to drought stress must be needed. Nowadays, drought stress is a severe problem in Iran. Domestication and cultivation of promising ecotypes in the dry land farming system are priorities. We hypothesized that there was a potential for domestication of wild ecotypes of *P. lanceolata* for cultivation in dryland farming or low irrigation areas. We tested the hypothesis by investigating the response of four local ecotypes of *P. lanceolata* to drought stress in a field experiment. Also, we estimated the correlation between seed yield and yield components in *P. lanceolata* to work out a regression of grain yield on its components.

Materials & Methods

This study was carried out at the experimental farm of the Research Institute of Forests and Rangelands, Karaj, Iran (latitude, 35°48′ N, longitude, 51°00′ E, Elevation, 1320 m above sea level) in 2018. The soil of the experimental site is sandy. Climate records (20 years) from a Karaj meteorological station indicate a mean annual total precipitation of 235 mm with relative humidity (68%). The mean annual temperature was 16°C.

A split plot design was conducted based on a randomized complete block design (RCBD) with three replications. The main factor was drought stress at three levels (D1=control-normal irrigation, D2= drought stress after the flowering stage with supplemental irrigation at the grain filling stage, and D3= stop irrigation at the flowering stage (severe stress). The second treatment was four ecotypes of *P. lanceolate*, namely: G1=(13561-Arak), G2=(15803-Khoramabad), G3=(27804-Meshkin1), and G4=(30196-Meshkin2). Data were collected for grain yield and 12 yield component traits. The experimental plot was 6 m^2. Seeds were sown manually at a row distance of 50 cm in 1 cm depth in mid-April 2018. The distance between the plants in the row was 30 cm. irrigation was applied every seven days intervals up to the flowering stage, then the drought stress was applied. During the growing season, weeds were controlled manually. In this experiment, nitrogen and phosphorus fertilizers were added to 100 $kg.h^{-1}$ urea (46% N) and 150 $kg.h^{-1}$ triple superphosphate (19.8% P), based on soil test and fertilizer recommendations before seed sowing, respectively.

At the biological maturity stage, 10 plants were randomly sampled from each plot to measure 12 traits as plant height (cm), stem lengths (cm), root lengths (cm), spike lengths (cm), spike number per plant, flower number per spike, grains number per spike, leaves number per plant, leaf lengths (cm), leaf width (cm), stem diameter (mm), 1000 grains weight (g) and grain yield ($kg.h^{-1}$) and averaged as a mean of the plot. Grain yield was measured from each plot after removing two side rows as border effects.

Collected data were analyzed for variance using a split-plot design with drought stress as the main plot and ecotypes as sub-plots [19]. Mean comparisons were made using LSD tests $\alpha=0.05$. Before performing the variance analysis, the variance's uniformity was tested using the method of Levene [20]. Phenotypic correlations among characteristics were determined for all pairwise combinations. Stepwise regression analysis was used for grain yield as a dependent variable. Using path analysis, the correlation of those traits entered in the final regression model was partitioned into direct and indirect effects. SAS9 conducted all statistical analyses.

Findings and Discussion

Effect of drought stress

Analysis of variance showed that the main effect of drought stress was significant ($p<0.05$ and 0.01) for all the traits (Table 1). Moreover, the main effect of ecotypes was also significant for all the traits except root lengths ($p<0.05$). The ecotype × drought stress interaction effect was significant for spikes number per plant, leaves number per plant, leaf width, and plant height ($p<0.05$) (Table 1). In all *P. lanceolata* ecotypes, drought stress reduced the mean of plant height with average values of 41.83, 36.54, and 34.67 cm for control, mild and severe drought stress, respectively (Table 2). The highest and lowest plant height values of 40 and 34 cm were obtained in G3 and

Table 1) Analysis of variance (mean squares) of grain yield and yield components in four ecotypes of *P. lanceolata* in three irrigation levels in the field condition.

Source	DF	MS												
		Plant height	Stem length	Spike length	Root length	Spikes no per plant	Flowers no per spike	Grains no per spike	Leaves no per plant	Leaf length	Leaf width	Stem diameter	1000 grains weight	Grain yield
Replication	2	5.67	8.04	0.40	1.21	810.4*	1485.2	1987.4	40.4	1.16	0.19*	0. 11	0.06	2988
Drought stress (D)	2	165.76**	38.01*	1.12*	115.75**	14326**	4484.2**	8726.9**	3494.4**	44.64**	0.25**	0. 53*	1.6**	4726*
Error1	4	11.69	2.79	0.16	1.53	223.1	1177.4	1610.4	605.7	1.46	0.00	0. 22	0.08	1963.6
Ecotypes (G)	3	59.67**	30.07*	1.62**	9.68	216.3*	1794.4**	5925.0**	1934.9**	18.35**	1.46**	0. 85*	0.14*	36628**
G× D	6	29.98*	2.64	0.46	4.97	828.0**	396.0	1963.6	685.1**	1.91	0.17**	0. 03	0.11	2372
Error2	18	10.20	8.39	0.29	6.89	73.5	319.1	980.9	109.1	3.97	0.03	0. 21	0.06	2368
Total	35													
CV%		8.48	12.16	15.24	21.45	14.73	18.78	25.03	15.53	14.63	11.17	11.97	6.01	7.34

* and ** are, respectively, significant at 5 and 1 % probability levels

G4 ecotypes, respectively (Table 3). The ecotype × drought stress interaction effect was significant for plant height ($p<0.05$), indicating that the responses of ecotypes to drought stress were not similar. The highest value of plant height, 48.7 cm, was obtained in G3 in regular irrigation, significantly higher than other ecotypes in the same irrigation. However, its value was sharply decreased by 37% in severe drought (Table 4). Drought stress significantly affected germination and enhanced drought levels, which strongly reduced seedling growth and could decrease both shoot and root lengths [21, 22]. Najafi and Rezvani Moghaddam (2002), in the study of the effect of four irrigation regimes 7, 14, 21, and 28 interval days on *P. ovata*, found the highest and lowest plant heights with values of 20.8 and 16.3 cm in control and severe stress (25% decreases) that were following our study [23]. A similar trend was observed for stem lengths. The highest and lowest stem lengths, with 25.19 and 21.81 cm values, were observed in D1 (control) and D3 (severe stress), respectively. Similarly, the longest and shortest stem lengths with values of 25.94 and 21.49 cm were obtained in G2 and G4, respectively (Tables 2 & 3). The ecotype × drought stress interaction effect was insignificant for stem length. However, the highest values of 27.07 cm were obtained in G2 in regular irrigation, and the lowest of 18.78 cm was obtained in G4 in severe drought stress (Table 4).

For spike length, the effect of both drought stress and ecotypes was significant. The highest and lowest spike lengths with values of 3.89 and 3.30 cm were observed for D1 (control) and D3 (severe drought stress), respectively (Table 2). The longest and the shortest spike lengths with average values of 4.18 and 3.26 cm were observed in G1 and G3, respectively (Table 3). A decrease in the spike length of *P. psyllium* in response to the more extended irrigation period has been reported [24, 25]. Similarly, Najafi and Rezvani Moghaddam (2002), in the study of the effect of drought stress on *P. ovata*, found the highest and lowest spike heights with average values of 2.37 and 1.97 cm, in 7 and 28 interval days irrigation, respectively [23]. The reduction of spike length under

Table 2) Overall means of grain yield and yield components of *P. lanceolata* in three irrigation levels in the field condition.

Treatment#	Plant height (cm)	Stem length (cm)	Spike length (cm)	Root length (cm)	Spikes No per plant	Flower No per Spike	Grains No per Spike	Leaves No per plant	Leaf length (cm)	Leaf width (cm)	Stem diameter (mm)	1000 grains weight g	Grain Yield Kg.h^{-1}
D1	41.83 a	25.19 a	3.89 a	14.88 a	93.25 a	116.25 a	151.67 a	85.33 a	15.28 a	1.72 a	1.27 a	1.35 a	729.41 a
D2	36.54 b	24.46 a	3.47 b	13.03 a	57.25 b	90.75 b	125.92 b	65.08 b	14.08 a	1.68 a	1.20 b	1.28 b	660.81 b
D3	34.67 b	21.81 b	3.30 c	8.82 b	24.17 c	78.33 c	97.75 c	51.42 c	11.50 b	1.45 b	1.13 c	1.13 c	595.95 c

Means with a different letter in each column are not significantly different (LSD=5%).
D1 = control, D2 = drought stress after the flowering stage with supplemental irrigation in the filling stage, and D3 = stop irrigation at flowering (severe stress).

stress conditions can result from reduced photosynthesis, and reduced production and transfer of assimilates for plant growth [26].

For root length, only the effect of drought stress was significant, and long and short root lengths with average values of 14.88 and 8.82 cm were obtained in D1 (control) and D3 (severe drought stress), respectively. The ecotype × drought stress interaction effect was insignificant for root length; however, the long and short root lengths with average values of 16.67 and 7.17 cm were obtained in G2 in regular irrigation and drought stress, respectively. The roots are the most critical parts of the plants that control the water and nutritional status of the plants and are the most essential organs of adaptation to water stress in most crops [27]. The first sign of drought stress in the plant is the rapid inhibition of shoot growth and root growth to a lesser extent [28]. Tolerance to drought stress in some crops can be caused by differences in their root growth [29].

For spike number per plant, the effect of ecotypes and drought stress was significant ($p<0.0$) (Table 1). Drought stress reduced the number of spikes number. The highest and lowest spike numbers, with values of 93.25 and 24.17, were obtained in D1 and D3, respectively. The highest and lowest spikes number with values of 64.22 and 54.0 were recorded in G4 and G3, respectively. The ecotype × drought stress interaction effect was significant for spikes number, indicating that the responses of ecotypes to drought stress were not similar. The highest spikes number, with a value of 124 spikes per plant, was obtained in G4 in regular irrigation, which was significantly higher than other ecotypes. However, it sharply dropped to 19 spikes per plant in severe drought stress (Table 4). The spikes number is one of the grain yield components that determine the yield potential because the spikes contain the grain numbers and, on the other hand, provide the photosynthetic material needed by the grains [30]. Like our research, drought stress reduces the intensity of spikes by decreasing the irrigation frequency in *P. ovata* [31, 32] and shortening the flowering period [33].

Both flower number and grain number per spike had the same trend. The main effect of drought stress and ecotypes was significant for both traits, but the effect of the ecotype × drought stress interaction was not significant (Table 1). The highest and lowest flower numbers, with values of (133.67 and 88.33) and grain numbers, with values of (168.33 and 143.33) were observed in the regular irrigation (D1) and severe drought stress (D3), respectively (Table 2). The highest and lowest flower numbers, with values of 116.0

Table 3) Overall means of grain yield and yield components in four ecotypes of *P. lanceolata* averaged over three irrigation levels in the field condition.

Ecotype code	Plant height (cm)	Stem length (cm)	Spike length (cm)	Root length (cm)	Spikes No per plant	Flowers No per Spike	Grains No per Spike	Leaves No per plant	Leaf length (cm)	Leaf width (cm)	Stem diameter (mm)	1000 grains weight g	Grain Yield $Kg.h^{-1}$
G1=13561 (Arak)	38.22^{a}	23.76^{ab}	4.18^{a}	13.66^{a}	60.22^{a}	116.00^{a}	163.00^{a}	63.78^{b}	15.58^{a}	2.04^{a}	1.33^{a}	1.28^{a}	670.92^{a}
G2=15803 (Khoram)	38.50^{a}	25.94^{a}	3.32^{b}	12.28^{a}	54.44^{b}	89.33^{b}	111.44^{c}	56.22^{c}	13.04^{b}	1.43^{c}	1.18^{a}b	1.29^{a}	657.38^{b}
G3=27804 (Meshkin1)	40.00^{a}	24.09^{a}b	3.26^{b}	11.22^{a}	54.00^{b}	90.22^{b}	118.44^{b}	60.33^{bc}	12.22^{c}	1.83^{b}	1.10^{b}	1.22^{b}	661.83^{b}
G4=30196 (Meshkin2)	34.00^{b}	21.49^{b}	3.46^{b}	11.81^{a}	64.22^{a}	84.89^{c}	107.56^{c}	88.78^{a}	13.62^{b}	1.14 d	1.19^{a}b	1.21^{b}	658.10^{b}

Means with a different letter in each column are not significantly different (LSD=5%).

Table 4) Mean grain yield ^{a}nd yield ^{c}omponents in four ecotypes of *P. lanceolata* in three irrigation levels in the field ^{c}ondition.

Ecotype code	Drought stress	Plant height (cm)	Stem length (cm)	Spike length (cm)	Root length (cm)	Spikes no per plant	Flowers no per spike	Grains no per spike	Leaves no per plant	Leaf length (cm)	Leaf width (cm)	Stem diameter (cm)	1000 grains Weight g	Grain Yield $Kg.h^{-1}$
G1	D1#	39.33^{b}	25.37^{ab}	4.60^{a}	16.00^{a}	83.0^{b}	133.67^{a}	168.33^{a}	77.33^{b}	17.73^{a}	1.87^{b}	1.40^{a}	1.34^{b}	736.92^{a}
	D2	37.83^{bc}	25.23^{ab}	4.20^{a}	13.97^{b}	66.0^{c}	126.00^{a}	177.33^{a}	65.33^{c}	16.00^{ab}	2.07^{a}	1.33^{a}	1.33^{b}	668.83^{b}
	D3	37.50^{bc}	20.67^{c}	3.73^{b}	11.00^{bc}	37.33^{e}	88.33^{b}	143.33^{b}	48.67^{d}	13.00^{b}	2.20^{a}	1.27^{ab}	1.17^{c}	607.02^{c}
G2	D1	41.00^{b}	27.07^{a}	3.57^{b}	16.67^{a}	88.33^{b}	111.33^{ab}	140.67^{b}	79.67^{b}	15.10^{b}	1.70^{b}	1.27^{ab}	1.47^{a}	727.41^{a}
	D2	37.50^{bc}	26.40^{a}	3.50^{b}	13.00^{b}	63.33b^{c}	80.67^{b}	114.00^{d}	65.33^{c}	12.80^{c}	1.50^{c}	1.13^{b}	1.30^{b}	659.62^{b}
	D3	37.00^{bc}	24.37^{b}	2.90^{c}	7.17^{c}	11.67^{f}	76.00^{bc}	68.00^{f}	23.67^{e}	11.23^{c}	1.10^{e}	1.13^{b}	1.10^{c}d	585.11^{d}
G3	D1	48.67^{a}	25.10^{ab}	3.33^{b}	12.50^{bc}	77.00^{b}	102.00^{ab}	123.00^{c}	65.00^{c}	12.63^{c}	2.07^{a}	1.17^{ab}	1.30^{b}	722.90^{a}
	D2	36.00^{bc}	23.83^{b}	2.80^{c}	12.33^{bc}	53.33^{d}	86.67^{b}	118.67^{c}	63.33^{c}	13.43^{c}	2.00^{a}	1.13^{b}	1.30^{b}	659.40^{b}
	D3	35.33^{bc}	23.33^{b}	3.63^{b}	8.83^{c}	28.67^{e}	82.00^{b}	113.67^{d}	52.67^{d}	10.60^{d}	1.43^{c}	1.00^{c}	1.07^{d}	603.19^{c}
G4	D1	38.33^{b}	23.23^{b}	4.07^{a}	14.33^{ab}	124.67^{a}	118.00^{ab}	174.67^{a}	119.33^{a}	15.63^{ab}	1.23^{d}	1.23^{ab}	1.30^{b}	730.42^{a}
	D2	34.83^{c}	22.37^{bc}	3.37^{b}	12.83^{bc}	52.67^{d}	69.67^{c}	93.67^{e}	66.33^{c}	14.07^{b}	1.13^{d}e	1.20^{ab}	1.17^{c}	655.38^{b}
	D3	28.83d	18.87^{c}	2.93^{c}	8.27^{c}	19.0^{ef}	67.00^{c}	66.00^{f}	80.67^{b}	11.17^{c}	1.07^{e}	1.13^{b}	1.17^{c}	588.51^{d}

Means with a different letter in each column are not significantly different (LSD=5%).
D1=Control, D2=drought stress after the flowering stage with supplemental irrigation in the filling stage, and D3 = Stop irrigation at flowering (severe stress)

and 84.89, and grains numbers, with values (of 163 and 107 per spike), were obtained in G1 and G4 ecotypes, respectively (Table 3).
The number of grains per spike determines sink capacity. The higher number of grains represents the more oversized sink for receiving photosynthetic material, and by increasing this trait, the grain yield is increased [30]. In addition, water stress at the flowering stage reduces the number of fertile flowers and consequently reduces the number of grains, thus significantly reducing grain yield [30, 34]. Many publications indicate the negative effect of drought stress on grain yield in *P. ovata* [23, 31, 35].
For leaves number, the effect of drought stress, ecotype, and their interaction was significant (Table 1). In all *P. lanceolata* ecotypes, drought stress reduced means leaves number per plant with values of 85.33, 65.08, and 51.42 for control, mild, and severe drought stress, respectively (Table 2). The higher and lower leaves number with average values of 88.78 and 56.22 were obtained in G4 and G2 ecotypes, respectively (Table 3). The ecotype × drought stress interaction effect was significant for leaves number, indicating that ecotypes' responses to drought stress were not similar. The result showed that the highest leaf number (119) value was obtained in G4 in regular irrigation. Its value sharply dropped to 66 leaves in mild stress, whereas, for other ecotypes, the lower values were consistently

Table 5) Phenotypic correlation between grain yield and yield components of *P. lanceolata* ecotypes grown under drought stress.

Traits	Spike length	Stem length	Flower per spike	Grains per spike	Spikes per plant	Stem Diameter	Root Length	Leaves Number	Leaf lengths	Leaf width	Plant height	1000 grains
Stem length	0.27											
Flowers per spike	0.83**	0.52*										
Grains per spike	0.87**	0.39	0.92**									
Spikes per plant	0.56**	0.48*	0.73**	0.74**								
Stem diameter	0.76**	0.23	0.78**	0.73**	0.51*							
Root lengths	0.65**	0.60**	0.77**	0.77**	0.86**	0.72**						
Leaves number	0.40	-0.04	0.44*	0.48*	0.79**	0.33	0.58**					
Leaf length	0.79**	0.40	0.86**	0.83**	0.75**	0.90**	0.89**	0.54*				
Leaf width	0.38	0.32	0.55*	0.63**	0.22	0.44*	0.42	-0.15	0.37			
Plant height	0.28	0.67**	0.56**	0.46*	0.55**	0.27	0.50*	0.03	0.31	0.57**		
1000 grains	0.40	0.67**	0.71**	0.63**	0.78**	0.61**	0.90**	0.51*	0.74**	0.44*	0.54*	
Grain yield	0.44*	0.66**	0.71**	0.59**	0.90**	0.50*	0.85**	0.58**	0.70**	0.28	0.71**	0.84**

* and ** =significant at 5 and 1 % probability levels, respectively,

Table 6) Results of stepwise regression analysis (b values) for grain yield as dependent variables and other traits as independent variables of *P. lanceolata* ecotypes.

Traits	Step 1	Step 2	Step 3	Step 4	Step 5	Step 6
Constant	584.10	450.50	426.20	439.10	360.90	177.00
Spikes per plant	1.36**	1.14**	1.05**	1.25**	1.03**	0.91*
Stem length		6.10**	4.00	4.10*	2.50	5.50*
Plant height			2.10	2.30*	3.60**	5.60**
Grains no per Spike				-0.24	-0.57**	-0.50**
Leaf length					8.80**	9.80**
Leaves number						1.01*
R^2	79.14	85.80	87.13	88.27	92.56	93.89

*, **= Regression coefficients are significant at 5%, 1%, respectively.

Table 7) Partitioning correlation coefficients to direct and indirect effects for grain yield in *P. lanceolata* ecotypes.

Traits	Direct	Indirect effects via						Total effects
	effects	Spikes per Plant	Stem Length	Plant Height	Grain per Spike	Leaf Length	Leaves Number	correlation
Spikes per Plant	0.10	-	0.12	0.29	-0.33	0.37	0.36	0.90**
Stem length	0.26	0.05	-	0.35	-0.17	0.19	-0.02	0.66**
Plant height	0.52	0.05	0.17	-	-0.20	0.15	0.01	0.71**
Grain per spike	-0.44	0.07	0.10	0.24	-	0.40	0.21	0.59**
Leaf length	0.49	0.07	0.10	0.16	-0.37	-	0.24	0.70**
Leaves number	0.45	0.08	-0.01	0.02	-0.21	0.26	-	0.59**

Residual effects=1.65

obtained in severe drought stress (Table 4). A similar trend was reported by Asgharipour and Rafiei, 2010, in *P. ovata* [36].
The leaf size of *P. lanceolata* was estimated using leaf length and width. For both traits, the effect of ecotype and drought stress were significant. The highest and lowest leaf lengths (15.28 and 11.50 cm) and leaf widths (1.72 and 1.45 cm) were observed in the regular irrigation (D1) and severe drought (D3), respectively. The ecotype × drought stress interaction effect was significant for leaf width ($p<0.01$), indicating that the responses of ecotypes to drought stress were not similar. The higher leaf widths (2.07 and 2.20) were obtained in G1 in mild and severe drought stress, respectively. In contrast, higher leaf widths (2.07 and 2.00) were obtained in G3 in regular irrigation and mild drought stress, respectively (Table 4). For stem diameter, the effect of both drought stress and ecotypes was significant (Table 1). Drought stress reduced the stem diameter with 1.27, 1.20, and 1.13 mm values in D1,

D2, and D3, respectively. The higher and lower stem diameters with 1.33 and 1.10 mm values were obtained in G1 and G3 ecotypes, respectively (Table 2 and 3).

For 1000 grain weight, the main effect of drought stress and ecotypes were significant (Table 1). Drought stress reduced 1000-grain weight with values of 1.35, 1.28, and 1.13 g in D1, D2, and D3, respectively (Table 2). Water stress reduced sink capacity and caused a reduction of grain weight consequently [37]. A similar trend was observed in *P. ovata* [23, 24, 32]. The highest and lowest 1000-grain weights with 120 and 121 g values were obtained in G1 and G4 ecotypes (Table 3). For grain yield, the main effect of drought stress and ecotypes was significant (Table 1). The values of grain yield in D1 (control), D2 (mild stress), and D3 (severe stress) were 719.42, 660.58, and 609.0 $kg.h^{-1}$, respectively (Table 2). The mean values of grain yield in ecotypes G1 and G2, G3, and G4 were 670.92, 657.38, 661.83, and 658.10 $kg.h^{-1}$, respectively (Table 3). The ecotype G1 (Arak) also had higher grain yields in mild and stress than other ecotypes. The decrease in grain yield in drought stress can be attributed to less vegetative growth and, consequently, more limited photosynthetic levels and less dry matter production in the plant under drought conditions. On the other hand, shortening the grain filling period and earlier ripening due to drought stress can effectively reduce grain yield [33]. Like our research, drought stress reduces grain yield by decreasing the irrigation frequency in *P. ovata* [23, 24, 30, 38, 39].

Relationship between grain yield and other traits

The result of correlation analysis showed that many of the traits had positively correlated with each other in *P. lanceolata* (Table 5). Grain yield had a significant positive correlation with all the traits except leaf width. Similarly, Bhagat (1980) found positive correlations between grain yield and some of its components in *P. ovata* [40]. Before path analysis, the essential traits were defined by a stepwise regression equation for grain yield as dependent and other traits as independent variables (Table 6). Based on the regression analysis results, six traits of spikes per plant, stem lengths, plant height, grain number per spike, leaf lengths, and leaf number were entered into the regression equation (Table 7). All these traits had strong positive correlations with grain yield (Table 6).

Path analysis is a method to explain cause and causation among traits and identify more effective traits [41]. In this study, grain yield was used as the dependent variable, and the six traits (that were entered into the regression equation), were used as independent variables. The correlation coefficients were partitioned into direct and indirect effects using path coefficients [42]. According to Table 7, the traits of plant height, leaf lengths, and leaf number had the highest positive direct effects, and the trait of the number of grains per spike had the negative direct effect. The values of traits with the same sign as the correlation coefficient suggested that these traits had genetic relationships with grain yield. Therefore, selection for plant height, leaf length, and leaf number could improve *P. lanceolata* grain yield.

Conclusion

All the ecotypes had the highest values for many traits in regular irrigation, and their values decreased by increasing drought stress. The highest grain yield with average values of 670.92 $kg.h^{-1}$ was obtained in ecotype G1 (Arak). This ecotype always had higher yields in three irrigation systems than the other ecotypes and was recommended for breeding improved varieties. The result of correlation analysis showed that all the traits were positively correlated with grain yield. However, based on the regression and path analysis results, three traits of plant height, leaf length, and leaf number had the highest positive direct effects on grain yield. Therefore, these traits could be used as selection indices to improve the *P. lanceolata* variety.

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References

1. Daneshian, J., Ahmadi, M., Kalantarahmadi, S.A. Stability evaluation of advanced soybean lines (*Glycine max* L.) in drought conditions using GGE-Biplot analysis and Ammi. Crop Prod. J. 2022; 15(4): 119-138. (In Persian).
2. Mahajan S., Tuteja N. Cold, salinity and drought stresses: an overview. Arch. Biochem. Biophys. 2005; 444(2):139-158.
3. Wang W.X., Vinocur B., Shoseyov O., Altman A. Biotechnology of plant osmotic stress tolerance: physiological and molecular considerations. Acta Hortic. 2001; 560(1):285-292.
4. Farooq M., Wahid A., Kobayashi N., Fujita D., Basra S.M.A. Plant drought stress: effects, mechanisms, and management. Agron. Sustain. Dev. 2009; 29(1): 185- 212.
5. Barnabas B., Jäger K., Fehér A. The effect of drought and heat stress on reproductive processes in cereals. Plant Cell Environ. 2008; 31(1): 11-38.
6. Belayet H.M., Rahman W., Rahman M.N., Noorul Anwar A.H.M., Hossein A.K.M. Effects of water stress on yield attributes and yield of different mung bean genotypes.Int. J. Sustain. Crop Prod. 2010; 5(1):19-24.
7. Xiaolong J., Chunyan H. and Xudan G. Physico-chemical properties, structures, bioactivities and future prospective for polysaccharides from *Plantago* L. (Plantaginaceae): a review. Int. J. Biol. Macromol. 2019; 135(1):637-646.
8. Goncalves S., Romano A. The medicinal potential of plants from the genus *Plantago* (Plantaginaceae). Ind. Crops Prod. 2016; 83(1): 213–226.
9. Janighorban, M. Plantaginaceae. In: Assadi M. (Ed.), Flora of Iran, No. 14. Research Institute of Forests and Rangelands, Tehran, Iran. 1995; 1–53.
10. Jankovi T., Zduni G., Beara I. Comparative study of some polyphenols in *Plantago* species. Biochem. Syst. Ecol. 2012; 42(1):69-74.
11. Kreitschitz, A., Kovalev, A., Gorb, S.N., 2016. "'Sticky Invasion'-the Physical. Properties of *Plantago Lanceolata* L. Seed Mucilage". Beilstein J. Nanotechnol.7(1):1918-1927
12. Amininasab, S.S, Mahmoudi Otaghvari, A. and Nazifi, E. 2020. Palynological study of *Plantago major and P. lanceolata* in north of Iran. Rostaniha.21(1): 38-48 (In Persian).
13. Temur C., Uslu, S. Effects of Plantain (*Plantago anceolata*) containing diets of quails on growth performance, carcass characteristic, some blood parameters and mast cell. Yuzuncu Yil. Univ. J. Agric. Sci. 2019; 29(1):114–120.
14. Oloumi M.M., Vosough D., Derakhshanfar A., Nematollahi M.H. The healing potential of *Plantago lanceolata* ointment on collagenase-induced tendinitis in burros (*Equus asinus*). J. Equine Vet. Sci. 2011; 31(8):470–474.
15. Foster, L. Herbs in pastures. Development research in Britain, 1850–1984. Biol. Agric. Hortic. 1988; 5(2):97–133.
16. Bahadori, M. B., Sarikurkcu, C., Kocak, M. S., Calapoglu, M., Uren, M. C., Ceylan, O. *Plantago lanceolata* as a source of health-beneficial phytochemicals: Phenolics profile and antioxidant capacity. Food Biosci. 2020; 34(1): 100536.
17. Pol M., Schmidtke K., Lewandowska, S. *Plantago lanceolata* – An overview of its agronomically and healing valuable features. Open Agric. 2021; 6(1):479–488.
18. Kaswan, V., Kaushik, A., Devi, J., Joshi, A. and Ratan Maloo, S. Genetic association studies for yield and yield contributing traits in Plantago ovata Forsk. Electron. J. Plant Breed. 2018 ;9(1): 51-59.
19. Steel R.G.D., Torrie J.H. Principles and procedures of statistics. A biometrical approach, 2nd Edition, McGraw-Hill Book Company, New York 1980.
20. Levene, H. 1960. Robust tests for equality of variances in contribution to probability and Statistics, (Ed) 1. Olkin: Stanford University Press, Palo Alto. California, USA.

21. Akhzari D., Ghasemi Aghbash F. Effect of Salinity and Drought Stress on the Seedling Growth and Physiological Traits of Vetiver Grass (*Vetiveria zizanioides* stapf.) ECOPERSIA 2013; 1(4). 339-352.
22. Saberi M., Shahriari A., Niknahad-Gharmakher H., Heshmati G., Barani H. Effects of Different Drought and Salinity Levels on Seed Germination of *Citrullus colocynthis*. ECOPERSIA 2017, 5(3):1903-1917
23. Najafi F., Rezvani Moghaddam P. Effects of irrigation regimes and plant density on yield and agronomic characteristics of isabgol (*Plantago Ovata*). ECOPERSIA 2002; 16(2):59-65.
24. Tabrizi L.The effect of water stress and manure on yield, yield components, and quality characteristics of *Plantago psyllium*. M.Sc. Thesis, Faculty of Agriculture, Ferdowsi University of Mashhad, Iran. J. Soil Water Conserv.2004;25(3):656-664
25. Koocheki A., Tabrizi L., Nassiri Mahallati, M. Organic cultivation of P. ovata and P. psyllium in response to water stress. Iran. J. Field Crop. Res. 2004; 2(1):67-78. (In Persian).
26. Pirasteh-Anosheh H., Emam Y., Ashraf M., Foolad M.R. Exogenous application of salicylic acid and chlormequat chloride alleviates negative effects of drought stress in wheat. Adv. Stud. Biol. 2012; 4(11):501–520.
27. Blum A. 2005. Drought resistance, water-use efficiency, and yield potential, are they compatible, dissonant, or mutually exclusive? Aust. J. Agric. Resour. Econ. 56(11): 1159-1168.
28. Neumann, P. M. Cropping mechanisms for crop plants in drought-prone environments. Ann. Bot. 2008; 101(7):901-907.
29. Xue Q., Zhu Z., Musick J.T., Stewart, B.A., Dusek D.A. Root growth, and water uptake in winter wheat under deficit irrigation. Plant Soil. 2003; 257(1):151-161.
30. Koocheki A., Mokhtari V., Taherabadi Sh., Kalantari S. The effect of water stress on yield, yield components and quality characteristics of *Plantago Ovata* and *P. psyillium.* J. Soil Water Conserv. 2011; 25(3): 656-664 (In Persian).
31. Patra D.D., Anwar M., Singh S., Prasad A., Singh D.V. Aromatic and medicinal plants for salt and moisture stress condition. Recent Advances in Management of Arid Ecosystem. Proceeding of a Symposium Held in India. 1999; 347-350.
32. Mousavinick M. Effect of drought stress and Sulphur fertilizer on quantity and quality yield of psyllium (*Plantago ovata* L.) in Baluchestan. J. Agroecol. 2012; 4(2):170-182.
33. Bernal M., Estiarte M., Penuelas, J. Drought advances spring growth phenology of the Mediterranean shrub Erica multiflora. Plant Biol. 2011; 13(2):252-257.
34. Roumani A., Biabani A., Rahemi Karizaki A., Gholamalipour Alamdari E., Gholizadeh A. The response of quantitative and qualitative characteristics of Isabgol (*Plantago ovata* Forssk.) to foliar application of salicylic acid and spermine under drought stress conditions. Environ. Stresses Crop Sci. 2020; 13(2):503-517. (In Persian).
35. Ramroudi M., Galavi M., Siahsar B.A., Allahdo M. Effect of micronutrient and irrigation deficit on yield and yield components of isabgol (*Plantago ovata* Forsk) using multivariate analysis. J. Sci. Food Agric. 2011; 9(1):247–251.
36. Asgharipour, M., Rafiei, M. Intercropping of Isabgol (Plantago ovata L.) and Lentil as Influenced by Drought Stress American-Eurasian J. Agric. Environ. Sci. 2010 ;9(1): 62-69.
37. Toghraei A., Mirshekari B., Daneshian J., Kazemi-Arbat H., Mohasses Mostashari M. Effect of Fertilizers Containing Nitrogen on Yield and Mucilage of Isabgol (*Plantago ovata* L.) in Irrigation and Cutting off Irrigation. J. Crop Ecophysiol. 2017; 11(4):791-804 (In Persian).
38. Pouryousef M., Mazaheri D., Chaiechi M.R., Rahimi A., Jafari A.A. Effects of different soil fertilizing treatment (chemical, organic and integrated) on yield and yield components and seed mineral nutrient content of Isabgol (*Plantago ovata* Forsk). Agron. J. 2014; 102(27):82-91. (In Persian).
39. Rahimi A., Jahansoz M.R., Rahimian Mashhadi H. Effect of Drought Stress and Plant Density on Quantity and Quality Characteristics of Isabgol (*Plantago ovata*) and French Psyllium. J. Crop Prod. Process.2014;4(12):143-155.
40. Bhagat N.R. Studies on Variation and association among seed yield and some component traits in *Plantago ovata* Forsk. Crop Improven. 1980; 7(1):60-63.
41. Wright S. Correlation and causation. J. Agric. Res. 1921; 20(1):557-585.
42. Dewey D.R., Lu K.H. A correlation and path coefficient analysis of components of crested wheatgrass seed production. Agron. J. 1959; 51(9):515-518.