



## Field screening for drought tolerance in *Setaria italica* and *Panicum miliaceum* millet germplasm from Iran

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### Abstract

The present study aims at field screening of Proso and Foxtail millet ecotypes for drought tolerance. Accordingly, 96 promising millet ecotypes along with four checks were evaluated under field conditions in Yazd province of Iran. Field experiment was conducted using an incomplete block design (LATTICE) with two replications under drought stress and control conditions in a period of two years. Multivariate analyses showed variance significant genetic variation ( $P < 0.01$ ) among millet ecotypes of Iranian origin. Drought stress tremendously affected grain yield of all genotypes. The interaction between genotype and drought was significant for panicle weight, panicle length and days to flowering. Based on the results of multivariate analyses we identified the effective traits which are the foremost factor responsible for grain yield and dry weight of fodder under drought stress. Therefore, the selection based on these traits would be preferable to identify genotypes with high yield. Eventually, eight ecotypes with the higher grain yield and 8 with the higher dry weight fodder were found highly adoptive under moisture stress conditions. Such ecotypes can be recommended as promising genotypes which may eventually be released as new cultivars for drought-affected areas.

**Key words:** Drought stress, foxtail millet, multivariate approaches, proso millet

### Introduction

Drought is one of the most significant environmental global phenomena affecting crop production. Yield improvement under drought is a major goal of plant breeding (Cattivelli et al. 2008; Tuberosa 2012; Mir et al. 2012). In this context, water scarcity in arid and

semi-arid regions is a major concern for agricultural authorities around the world (Amini 2012). Millet is a broad term used for a diverse group of cereal crops that characteristically produce small seeds and include several annual food and fodder grasses such as foxtail millet, (or common millet or bromocorn millet) (*Setaria italica*), pearl millet (*Pennisetum glaucum*), finger millet (*Eleusine coracana*), proso millet (*Panicum miliaceum*), etc. (Dwivedi et al. 2012). About 90% of global millet production is utilized in the developing countries and 43.85% of total world millet is produced by India alone (FAO 2013). Distinguishing features of the millets are their adaptability to unfavorable agro-ecological conditions, requisite of least inputs, and excellent nutritional properties. They represent indispensable plant genetic resources for the agriculture and food security of poor farmers that reside in arid, uncultivable, and marginal lands (Charu et al. 2012). Millet grain contains 5-6% oil and among all cereals, it is the cheapest source of energy, protein, iron, and zinc. Foxtail millet is considered as a remarkably drought tolerant crop and its water use efficiency (WUE) has also been found to be higher than maize, wheat and sorghum (Zhang et al. 2007). Its drought tolerance ability has also been accredited to the association between increased WUE and its several morphological characteristics such as dense root system, thick cell walls, epidermal cell arrangements and minuscule leaf areas (Li 1997). Further for 1 g of dry biomass, foxtail millet requires only 257 g of water which is much lower than maize

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and wheat requiring 470 and 510 g of water, respectively (Diao 2007; Li and Brutnell 2011). Therefore, its short life cycle and high WUE makes foxtail millet a suitable crop for cultivation in semi-arid, dry and marginal lands. Iran has a large area falling under semi-arid with marginal lands. Seghatoleslami et al. (2008) evaluated yield and its components along with some morphological attributes in three millets namely, foxtail millet, pearl millet and proso millet under moisture stress. Foxtail millet produced highest seed yield and highest number of seeds per ear under deficit irrigation. It was also found to have highest WUE and harvest index (HI) and therefore greatest yield in both stress and non-stress conditions as compared to other two millets (Seghatoleslami et al. 2008). One of the greatest challenges in drought is to show a seed type that has the capacity to produce abundant biomass and ground cover in a short period of time (Van den Berg 2002). Millet is one of those cereal grasses which has strong development of roots and tends to have efficient adaptive mechanisms to cope with drought (Winkel and Do 1992).

Breeding progress pointed out that selection for high yield in stress free conditions has, to a certain extent, indirectly improved yield in many water-limiting conditions (Cattivelli et al. 2008). However, the breeding strategy requires the improvement of traits that reduce the gap between yield potential and actual yield in drought prone environments. Despite a positive correlation between grain yield under drought stress and non-stressed conditions (Golabadi et al. 2006; Dadbakhsh 2011; Mohammadi et al. 2011; Tester and Langridge 2010), indirect selection based on yield potential and mean yield under non-stress conditions may not give the best results for the selection of drought-tolerant genotypes (Abdolshahi et al. 2013). To date, cereal breeding has been based principally on empirical selection for yield (Evans 1993; Araus et al. 2002). However, this approach is far from being optimal, since drought prone environments are notably variable from year to year, and variability for yield is low (Ludlow and Muchow 1990; Dhanda et al. 2004). In addition, yield is characterized by a low heritability and a high genotype  $\times$  environment interaction (Jackson et al. 1996; Araus et al. 2002). Breeding for drought tolerance using secondary traits associated with yield under stress can provide additional information for breeders in selective processes (Fischer et al. 2003). Multivariate analysis in the field screening of genotypes had been suggested as a useful screening tool which was used by various

researchers (Sardouie-Nasab et al. 2014)

The main objective of this study was to identify the best millet genotypes for drought prone environments and improve genetic gain for grain yield and fodder grasses in Iran.

## Materials and methods

The genetic materials used in this study consisted of 96 ecotypes including 48 proso millet (*Panicum miliaceum*) and 48 foxtail millet (*Setaria italica*) along with four check cultivars, namely, Pishahang, Bastan, Golbaf and Rabor. Pishahang is improved cultivar of proso millet and Bastan of foxtail millet, while Golbaf and Rabor are local ecotypes of proso millet. Table 1 presents the details of 100 Iranian millet genotypes with their place of origin. Two field experiments were conducted in two consecutive growing seasons (2013 and 2014). The genotypes were evaluated in a field experiment using an incomplete block design (LATTICE) with two replications under drought stress and control conditions in Meybod-Yazd, located in the South-Eastern part of Iran (1034 m amsl, 54°N, 32°E) with a hot and arid climate. Sowing time was in mid-May in both the experiments with a density of 350 plants per square meter. Irrigated plots were watered prior to planting, tillering, jointing, flowering and grain filling stages. The total amount of water used for irrigation treatments was estimated using FAO Penman-Monteith (Allen 1998). Both stress and control levels got irrigated till the flowering stage. Irrigation was done after flowering in control levels whereas soil moisture was equivalent to field capacity but stress levels were irrigated once the soil moisture was close to permanent wilting point. Ten plants were randomly chosen from each plot to measure morphological and phenological traits, namely, plant height (PH), panicle length (PL), panicle diameter (PD), flag leaf length and width (FLL, FLW), days to flowering (DF), grain yield (GY), fresh and dry weight fodder (FWF, DWF) and panicle weight (PW).

## Statistical analysis

The analysis of traits was done based on a LATTICE design as per SAS procedure (SAS Institute, 2004), and the efficiency of LATTICE was not higher than the randomized complete block design (RCBD), therefore, the analysis of variance was a combined analysis over the drought levels (stress and normal) from 2013 to 2014 according to RCBD. Duncan's multiple range tests was employed for the mean

**Table 1.** A list of 100 millets (*Panicum miliaceum* and *Setaria italica*) genotypes from Iran

			Species				
Proso millet ( <i>Panicum miliaceum</i> )			Foxtail millet ( <i>Setaria italica</i> )				
1	Pishahang (C)	27	Shahrekord-	53	Yazd-1	79	Mashhad-2
2	Bastan* (C)	28	Shahrekord-3	54	Yazd-2	80	Mashhad-3
3	Rabor (C)	29	Shahrekord-4	55	Yazd-3	81	Mashhad-4
4	Golbaf (C)	30	Mashhad-1	56	Yazd-4	82	Tabriz-1
5	Yazd-1	31	Mashhad-2	57	Kerman-1	83	Tabriz-2
6	Yazd-2	32	Mashhad-3	58	Kerman-2	84	Tabriz-3
7	Yazd-3	33	Mashhad-4	59	Kerman-3	85	Tabriz-4
8	Yazd-4	34	Tabriz-1	60	Kerman-4	86	Shiraz-1
9	Kerman-1	35	Tabriz-2	61	Esfahan-1	87	Shiraz-2
10	Kerman-2	36	Tabriz-3	62	Esfahan-2	88	Shiraz-3
11	Kerman-3	37	Tabriz-4	63	Esfahan-3	89	Ilam-1
12	Kerman-4	38	Shiraz-1	64	Esfahan-4	90	Ilam-2
13	Esfahan-1	39	Shiraz-2	65	Khozestan-1	91	Ilam-3
14	Esfahan-2	40	Shiraz-3	66	Khozestan-2	92	Gilan-1
15	Esfahan-3	41	Ilam-1	67	Khozestan-3	93	Gilan-2
16	Esfahan-4	42	Ilam-2	68	Khozestan-4	94	Gilan-3
17	Khozestan-1	43	Ilam-3	69	Birjand-1	95	Zabol-1
18	Khozestan-2	44	Gilan-1	70	Birjand-2	96	Zabol-2
19	Khozestan-3	45	Gilan-2	71	Birjand-3	97	Zabol-3
20	Khozestan-4	46	Gilan-3	72	Tabas-1	98	Mazanderan-1
21	Birjand-1	47	Zabol-1	73	Tabas-2	99	Mazanderan-2
22	Birjand-2	48	Zabol-2	74	Shahrekord-1	100	Mazanderan-3
23	Birjand-3	49	Zabol-3	75	Shahrekord-2		
24	Tabas-1	50	Mazanderan-1	76	Shahrekord-3		
25	Tabas-2	51	Mazanderan-2	77	Shahrekord-4		
26	Shahrekord-1	52	Mazanderan-3	78	Mashhad-1		

\* = *Setaria italica*; C= Check cultivars

comparisons of grain yield and dry fodder yield in stress and non-stress conditions for both the years. Statistical parameters such as minimum, maximum and mean of each trait were calculated using Excel Microsoft. To have a predictive model of grain yield and dry fodder yield, a multiple linear regression was performed in stress conditions over two years. Regression coefficient for each trait and explained proportion of variance were calculated. In order to study direct and indirect effects of traits on GY and DWF and to find the most important effective trait on GY and DWF path coefficient analysis was performed. Traits entered in the regression model were used as independent variables and GY and DWF

considered as dependent variable in the path model. Calculations were carried out with Path 2 software's.

## Results

Combined analysis of variance (ANOVA) over the drought stress treatments (drought and control) from 2013 to 2014 based on RCBD was carried out (Table 2). Highly significant differences were observed among genotypes. The interaction between genotypes and drought stress were significant for PW, PLW, DF and GY. Given that proso and foxtail millet are two different species, SS separation was done based on this two groups (within P, within S and S vs P). The results showed all of the measured traits had highly significant

**Table 2.** Combined analysis of investigated traits of 100 millet ecotypes under drought stress and well watered

Source of variation	df	DF	FWF	DWF	GY	PH	FLL	FLW	PL	PD	PW
Year	1	4050*	646.7 <sup>ns</sup>	135.1 <sup>ns</sup>	56.6**	13028.9*	963.4**	5.7**	2987**	76**	27.2*
Stress	1	58.3 <sup>ns</sup>	484.4 <sup>ns</sup>	152 <sup>ns</sup>	20.1*	5614.4 <sup>ns</sup>	658.5**	1.1 <sup>ns</sup>	957**	1.0*	1.3 <sup>ns</sup>
year*Stress	1	43.3 <sup>ns</sup>	51.1 <sup>ns</sup>	0.33 <sup>ns</sup>	0.8 <sup>ns</sup>	2616.3 <sup>ns</sup>	961.8**	5.7**	604.4**	0.4 <sup>ns</sup>	5.5 <sup>ns</sup>
block (year*Stress)	4	270.7	363.2	48.9	2.0	1481.7**	31.6	0.2	23.42	0.1	1.3
Genotype	99	454.5**	347.3**	75.2**	1.8**	2391.5**	43.8**	0.6**	109.2**	0.6**	7.6**
S vs P	1	13714 **	2035.5**	4.4**	4.2**	129773 **	88.9**	22.4**	6315**	1.9**	148.4**
within P	50	381.3**	408.4**	81.5**	1.6**	931.4**	43.3**	0.1**	31.9**	0.6**	1.6**
within S	48	254.4**	248.6**	70.2**	2.0**	1258.5**	43.4**	0.7**	60.3**	0.6**	11.0**
Stress	99	79.22**	79.7 <sup>ns</sup>	21.1 <sup>ns</sup>	0.2 <sup>ns</sup>	179.9 <sup>ns</sup>	10.2 <sup>ns</sup>	0.1 <sup>ns</sup>	12.5*	0.12 <sup>ns</sup>	0.6**
*Genotype											
Stress*s vs P	1	64.8 <sup>ns</sup>	142.6 <sup>ns</sup>	32.3 <sup>ns</sup>	1.2**	293.7 <sup>ns</sup>	24.8 <sup>ns</sup>	0.02 <sup>ns</sup>	421.8**	0.6 <sup>ns</sup>	8.3**
Stress * P	50	41.9**	71.4 <sup>ns</sup>	17.6 <sup>ns</sup>	0.2*	186.2 <sup>ns</sup>	12.1 <sup>ns</sup>	0.1*	8.3 <sup>ns</sup>	0.1 <sup>ns</sup>	0.3**
Stress * S	48	118.4**	870 <sup>ns</sup>	24.4 <sup>ns</sup>	0.2 <sup>ns</sup>	171.0 <sup>ns</sup>	7.9 <sup>ns</sup>	0.1 <sup>ns</sup>	8.5 <sup>ns</sup>	0.1 <sup>ns</sup>	0.7**
year	99	0.8 <sup>ns</sup>	94.7 <sup>ns</sup>	27.7**	0.2 <sup>ns</sup>	531.5**	19.4**	0.2**	18.5**	0.4**	0.01 <sup>ns</sup>
*Genotype											
year* S vs P	1	0.5 <sup>ns</sup>	337.3 <sup>ns</sup>	355.1**	0.7 <sup>ns</sup>	12041.8**	61.2**	5.9**	5.3 <sup>ns</sup>	0.21 <sup>ns</sup>	0.02 <sup>ns</sup>
year* P	50	1.3 <sup>ns</sup>	84.9 <sup>ns</sup>	24.7**	0.1 <sup>ns</sup>	418.7**	23.2**	0.1*	21.6**	0.6**	0.01 <sup>ns</sup>
year* S	48	0.3 <sup>ns</sup>	99.77 <sup>ns</sup>	24.0 <sup>ns</sup>	0.2 <sup>ns</sup>	409.2**	14.6**	0.21**	15.6*	0.2**	0.01 <sup>ns</sup>
year*Stress	99	0.8 <sup>ns</sup>	94.35 <sup>ns</sup>	17.7 <sup>ns</sup>	0.1 <sup>ns</sup>	198.1 <sup>ns</sup>	7.8 <sup>ns</sup>	0.1 <sup>ns</sup>	8.6 <sup>ns</sup>	0.11	0.01 <sup>ns</sup>
*Genotype											
<b>Error</b>	<b>396</b>	<b>35.2</b>	<b>81.63</b>	<b>18.8</b>	<b>0.2</b>	<b>190.7</b>	<b>8.4</b>	<b>0.1</b>	<b>9.6</b>	<b>0.133</b>	<b>0.12</b>

\*\* ,\* and ns: significant at 0.05, 0.01 probability levels and no-significant, respectively. Plant height (PH), Days to flowering (DF), Grain yield (GY), Fresh and dry weight fodder (FWF, DWF), Panicle length (PL), Panicle diameter (PD), Panicle weight (PW), Flag leaf length and width (FLL, FLW)

**Table 3.** Means, maximum, minimum, and phenotypic coefficient variance of some morpho-physiological traits in proso millet and foxtail millet ecotypes

Traits	%PVC				Max				Min				Mean*			
	Proso		Foxtail		Proso		Foxtail		Proso		Foxtail		Proso		Foxtail	
	N	S	N	S	N	S	N	S	N	S	N	S	N	S	N	S
DF	5.69	22.8	9.8	7.1	71.0	67	77.0	76	35.0	35.0	37.0	35.0	61.6 <sup>a</sup>	51.8 <sup>a</sup>	58.4 <sup>a</sup>	58.4 <sup>a</sup>
FWF	19.9	21.1	29.8	22.3	38.0	25	37.6	24.9	4.9	4.3	2.25	2.1	29.9 <sup>a</sup>	27.5 <sup>a</sup>	25.8 <sup>a</sup>	22.1 <sup>a</sup>
DWF	19.4	21.7	9.6	24.5	25.0	22.4	24.0	20.0	2.0	2.0	4.0	3.6	14.0 <sup>a</sup>	11.8 <sup>a</sup>	13.5 <sup>a</sup>	12.0 <sup>a</sup>
GY	25.2	29.1	20.1	9.13	2.7	2.5	2.7	1.9	0.3	0.21	0.3	0.21	1.2 <sup>a</sup>	1.0 <sup>a</sup>	1.4 <sup>a</sup>	1.0 <sup>a</sup>
PH	10.8	7.7	9.9	8.23	170.0	136.0	180.6	163.8	50.0	45.0	70.0	40.8	97.2 <sup>a</sup>	92.8 <sup>a</sup>	121.6 <sup>a</sup>	110.6 <sup>a</sup>
FLL	7.43	6.3	61.1	20.9	42.0	40.0	42.0	39.0	18.0	18.0	18.0	17.0	30.5 <sup>a</sup>	28.4 <sup>a</sup>	30.8 <sup>a</sup>	29.4 <sup>a</sup>
FLW	10.3	8.3	11.3	8.8	3.0	2.5	3.5	3.2	1.0	0.8	0.87	0.7	1.5 <sup>a</sup>	1.3 <sup>a</sup>	1.9 <sup>a</sup>	1.4 <sup>a</sup>
PL	8.85	9.7	14.6	10.0	35.0	31.0	35.0	29.7	16.8	12.0	9.0	4.08	25.0 <sup>a</sup>	21.4 <sup>a</sup>	18.0 <sup>a</sup>	17.2 <sup>a</sup>
PD	16.1	11.1	13.8	13.4	3.5	3.0	3.3	2.9	1.0	0.92	0.85	0.73	1.92 <sup>a</sup>	1.8 <sup>a</sup>	1.7 <sup>a</sup>	1.5 <sup>a</sup>
PW	7.7	9.4	9.28	7.7	5.1	4.1	8.78	5.21	0.86	0.44	1.43	1.02	2.35 <sup>a</sup>	2.1 <sup>a</sup>	3.3 <sup>a</sup>	2.6 <sup>a</sup>

N = Normal condition; S = Stress condition; PVC = phenotypic coefficient variance; \* = Means followed by same letters were not significantly different at P = 0.05; Plant height (PH); Days to flowering (DF), Grain yield (GY), Fresh and dry weight fodder (FWF, DWF), Panicle length (PL), Panicle diameter (PD), Panicle weight (PW), Flag leaf length and width (FLL, FLW)

**Table 4.** Mean comparison of grain yield and dry weight fodder of millet ecotypes in two stress and non-stress environments over two years

S.No*	Grain yield (tha <sup>-1</sup> )	Forage yield (tha <sup>-1</sup> )	S.No	Grain yield (tha <sup>-1</sup> )	Forage yield (tha <sup>-1</sup> )	S.No	Grain yield (tha <sup>-1</sup> )	Forage yield (tha <sup>-1</sup> )	S.No	Grain yield (tha <sup>-1</sup> )	Forage yield (tha <sup>-1</sup> )
1	1.57 <sup>d-m</sup>	18.7 <sup>a-d</sup>	26	0.74 <sup>z</sup>	12.3 <sup>h-y</sup>	51	0.80 <sup>x-z</sup>	9.0 <sup>w-y</sup>	76	1.35 <sup>j-w</sup>	14.2 <sup>c-x</sup>
2	0.60 <sup>z</sup>	8.3 <sup>y-z</sup>	27	1.58 <sup>d-m</sup>	18.3 <sup>a-f</sup>	52	1.69 <sup>b-l</sup>	17.5 <sup>a-i</sup>	77	0.50 <sup>z</sup>	9.2 <sup>u-y</sup>
3	1.79 <sup>a-i</sup>	9.5 <sup>t-y</sup>	28	0.70 <sup>z</sup>	16.2 <sup>a-n</sup>	53	2.05 <sup>a-d</sup>	16.7 <sup>a-m</sup>	78	0.66 <sup>z</sup>	9.9 <sup>q-y</sup>
4	1.91 <sup>a-e</sup>	9.8 <sup>t-y</sup>	29	0.65 <sup>z</sup>	13.0 <sup>e-y</sup>	54	2.09 <sup>a-b</sup>	19.0 <sup>a-c</sup>	79	1.36 <sup>j-v</sup>	18.0 <sup>a-g</sup>
5	1.7 <sup>a-k</sup>	15.3 <sup>b-s</sup>	30	1.35 <sup>j-w</sup>	19.9 <sup>ab</sup>	55	0.71 <sup>z</sup>	6.1 <sup>z</sup>	80	1.06 <sup>o-z</sup>	14.2 <sup>c-x</sup>
6	0.84 <sup>w-z</sup>	15.4 <sup>b-q</sup>	31	0.66 <sup>z</sup>	12.2 <sup>h-y</sup>	56	1.99 <sup>a-e</sup>	16.3 <sup>a-n</sup>	81	1.51 <sup>e-q</sup>	16.1 <sup>a-n</sup>
7	1.41 <sup>f-s</sup>	17.6 <sup>a-h</sup>	32	1.19 <sup>l-z</sup>	11.9 <sup>i-y</sup>	57	1.88 <sup>a-h</sup>	12.1 <sup>h-y</sup>	82	1.58 <sup>c-m</sup>	14.2 <sup>c-x</sup>
8	1.03 <sup>p-z</sup>	12.2 <sup>h-y</sup>	33	0.81 <sup>x-z</sup>	13.1 <sup>e-y</sup>	58	2.21 <sup>a</sup>	16.8 <sup>a-l</sup>	83	1.65 <sup>b-m</sup>	15.4 <sup>b-r</sup>
9	0.69 <sup>z</sup>	15.6 <sup>b-p</sup>	34	1.86 <sup>a-i</sup>	15.0 <sup>b-t</sup>	59	0.79 <sup>x-z</sup>	15.0 <sup>b-t</sup>	84	1.52 <sup>e-o</sup>	15.5 <sup>b-p</sup>
10	1.50 <sup>e-q</sup>	13.7 <sup>c-y</sup>	35	0.60 <sup>z</sup>	5.8 <sup>z</sup>	60	1.11 <sup>n-z</sup>	16.0 <sup>a-n</sup>	85	2.19 <sup>a</sup>	11.5 <sup>k-y</sup>
11	0.78 <sup>x-z</sup>	14.9 <sup>b-t</sup>	36	0.61 <sup>z</sup>	14.3 <sup>c-x</sup>	61	1.19 <sup>l-z</sup>	10.0 <sup>t-y</sup>	86	0.80 <sup>x-z</sup>	12.6 <sup>g-y</sup>
12	0.66 <sup>z</sup>	15.8 <sup>b-o</sup>	37	1.98 <sup>a-e</sup>	15.0 <sup>b-t</sup>	62	0.79 <sup>x-z</sup>	8.7 <sup>w-y</sup>	87	0.50 <sup>z</sup>	15.3 <sup>b-s</sup>
13	0.88 <sup>u-z</sup>	17.3 <sup>a-j</sup>	38	0.64 <sup>z</sup>	15.7 <sup>b-p</sup>	63	1.17 <sup>m-z</sup>	8.0 <sup>z</sup>	88	1.35 <sup>j-v</sup>	13.6 <sup>c-y</sup>
14	1.90 <sup>a-g</sup>	10.3 <sup>o-y</sup>	39	0.77 <sup>x-z</sup>	11.7 <sup>k-y</sup>	64	0.64 <sup>z</sup>	12.1 <sup>h-y</sup>	89	1.75 <sup>a-k</sup>	12.0 <sup>i-y</sup>
15	0.69 <sup>z</sup>	14.9 <sup>b-t</sup>	40	1.40 <sup>g-t</sup>	11.8 <sup>j-y</sup>	65	0.69 <sup>z</sup>	11.7 <sup>ky</sup>	90	0.88 <sup>u-z</sup>	13.3 <sup>d-y</sup>
16	0.64 <sup>z</sup>	12.1 <sup>h-y</sup>	41	0.57 <sup>z</sup>	11.2 <sup>m-y</sup>	66	1.63 <sup>b-m</sup>	16.0 <sup>a-n</sup>	91	2.08 <sup>a-b</sup>	17.0 <sup>a-k</sup>
17	1.03 <sup>p-z</sup>	11.7 <sup>k-y</sup>	42	0.61 <sup>z</sup>	10.3 <sup>o-y</sup>	67	1.00 <sup>q-z</sup>	9.0 <sup>w-y</sup>	92	1.45 <sup>f-r</sup>	14.4 <sup>c-w</sup>
18	0.87 <sup>v-z</sup>	13.7 <sup>c-y</sup>	43	0.92 <sup>s-z</sup>	14.7 <sup>c-u</sup>	68	0.80 <sup>x-z</sup>	10.3 <sup>n-y</sup>	93	0.88 <sup>u-z</sup>	14.4 <sup>c-w</sup>
19	1.21 <sup>l-z</sup>	9.1 <sup>v-y</sup>	44	0.90 <sup>t-z</sup>	9.6 <sup>t-y</sup>	69	0.92 <sup>s-z</sup>	11.1 <sup>m-y</sup>	94	1.26 <sup>k-x</sup>	12.9 <sup>f-y</sup>
20	1.55 <sup>d-o</sup>	11.1 <sup>n-y</sup>	45	0.83 <sup>x-z</sup>	12.2 <sup>h-y</sup>	70	1.58 <sup>d-m</sup>	16.6 <sup>a-n</sup>	95	0.73 <sup>z</sup>	12.3 <sup>h-y</sup>
21	0.86 <sup>v-z</sup>	14.2 <sup>c-x</sup>	46	0.53 <sup>z</sup>	11.3 <sup>l-y</sup>	71	1.19 <sup>l-z</sup>	14.6 <sup>c-u</sup>	96	0.73 <sup>z</sup>	11.6 <sup>k-y</sup>
22	0.73 <sup>z</sup>	13.6 <sup>c-y</sup>	47	0.64 <sup>z</sup>	9.7 <sup>s-y</sup>	72	1.39 <sup>h-u</sup>	13.9 <sup>c-x</sup>	97	0.50 <sup>z</sup>	9.6 <sup>t-y</sup>
23	1.80 <sup>a-i</sup>	12.8 <sup>f-y</sup>	48	0.98 <sup>t-z</sup>	12.8 <sup>f-y</sup>	73	0.69 <sup>z</sup>	11.2 <sup>l-y</sup>	98	1.53 <sup>e-o</sup>	13.6 <sup>c-y</sup>
24	0.81 <sup>x-z</sup>	10.2 <sup>p-y</sup>	49	1.43 <sup>f-r</sup>	21.2 <sup>a</sup>	74	0.76 <sup>y-z</sup>	18.5 <sup>a-e</sup>	99	1.28 <sup>j-x</sup>	14.8 <sup>c-u</sup>
25	1.24 <sup>k-z</sup>	19.0 <sup>a-c</sup>	50	0.90 <sup>t-z</sup>	9.5 <sup>t-y</sup>	75	0.75 <sup>y-z</sup>	12.7 <sup>g-y</sup>	100	1.77 <sup>a-j</sup>	14.4 <sup>c-w</sup>

Means with similar letters in each column are not significantly different ( $P>0.05$ ); \*Sl. Nos. represent the genotypes listed in Table 1

differences among two millet species. Also significant differences were observed within groups.

In the interaction of stress × proso millet and stress × foxtail millet all the measured traits had the same reaction to drought (no significant difference) except for GY and FLW. However, the proso millet ecotypes showed a significant difference in terms of GY, whereas no significant difference was observed for GY in drought levels for foxtail millet ecotypes. There was a significant difference among years for dry weight fodder. The statistical parameters for two millet species in stress and normal conditions are shown in Table 3. In proso millet ecotypes, the mean of days to flowering at drought conditions was lower than normal conditions. The highest of phenotypic coefficient of variance (PCV) was related to GY in both conditions. The lower value was related to DF at

normal condition and FLL at stress conditions.

#### Means comparisons for yield

Mean comparison of grain yield (Table 4) indicated that, 8 proso millet ecotypes had much better performance than the tolerant cultivar, Pishahang and 29 foxtail millet ecotypes displayed much better performance than the tolerant cultivar (Bastan) under both the conditions. Among this better (foxtail millet and proso millet) ecotype from Kerman-2 of *Setaria* was the most productive one in terms of grain yield (2.2 tha<sup>-1</sup>), whereas proso millet ecotype from Zabol-3 showed higher value for DWF (21.77 tha<sup>-1</sup>).

#### Path coefficient analysis for yield

The correlation coefficients representing correlation of yield contributing traits with yield, while path



**Table 5.** Results of path analysis for grain yield in stress environment of two group milletecotypes

Grain yield	Traits	proso millet				Total effects	Traits	foxtail millet				Total effects
		Direct effects	indirect effects via					Direct effects	indirect effects via			
		PD	PL	PW		PL	PW	PD				
	PW	0.562	0.02	0.04	-	0.622	PD	0.551	0.15	-0.026	-	0.675
	PD	-0.253	-	0.011	0.02	-0.244	PL	0.319	-	-0.01	0.012	0.297
	PL	0.512	0.034	-	0.041	0.587	PW	0.486	-0.04	-	-0.03	0.416
	Residual	-	0.651	-	-	-	Residual	-	0.631	-	-	-
	Traits	Direct effects	FWF	FLL	PH	Total effects		Direct effects	FWF	FLL	PH	Total effects
Dry	PH	0.614	0.102	0.15	-	0.866	FWF	0.698	0.024	0.134	-	0.856
fodder	FWF	0.257	-	0.057	0.058	0.372	PH	0.419	-	-0.04	0.015	0.394
	FLL	-0.270	-0.04	-	-0.07	-0.38	FLL	-0.255	-0.08	-	-0.051	-0.386
	Residual	-	0.532	-	-	-	Residual	-	0.573	-	-	-

coefficient analysis depicts nature and extent of correlation whether direct or indirect towards yield. In proso millet, path coefficients under drought stress conditions revealed that PW and PL had a positive direct effect on grain yield, while panicle diameter had negative direct effect (-0.253) and indirect effects through PL and PW was positive (Table 5). In foxtail millet, the highest direct effect on GY was ascribed to PD (0.55) and its indirect effects through PL and PW was positive (0.15) and negative (-0.026), respectively.

Path coefficients in proso millet for fodder yield revealed that PH had the highest direct effect on DWF and high correlation between PH and DWF was related to direct effect of this trait while its indirect effect through other traits was not considerable. In Foxtail millet based on the results of path coefficient in drought stress, FWF had a positive direct effect equal to 0.698

and the highest indirect effect was through FLL (0.13).

#### **Regression analysis for grain yield and forage yield**

In this study a regression model was used to facilitate the interpretation of GY and DWF. The results of regression analysis including, the regression coefficients (b) and explained proportion of variance (R<sup>2</sup>) in each substrates are presented in Table 6. The results of regression on GY indicate that 55% of total GY variation explained by PW, PD and PL in proso millet. PL, PD and PW were explained in terms of 51% variation in GY in foxtail millet with regression coefficient, 0.77, -0.25 and 0.21, respectively. The results of regression on DWF indicated that 75% of variation of DWF explained by PH, FWF and FLL in proso millet. PH, FWF and FLL explained 76% of variation for DWF in foxtail millet, which had positive effect on it.

**Table 6.** Results of regression for grain yield and dry weight fodder of two millet groups under stress environment over two years

Yield	Group	Fixed variable	Grain yield			Forage yield				
			Beta	Model R-square	Partial R-square	F	Beta	Model R-square	Partial R-square	F
Proso millet	Intercept	-0.57	-	-	8.14**	Intercept	-0.66	-	-	0.56
	PW	0.74	0.48	0.48	9.46**	PH	0.37	0.71	0.71	512**
	PD	0.50	0.51	0.14	14.5**	FWF	.34	0.74	0.03	27.51**
	PL	0.41	0.55	0.06	23.5**	FLL	0.19	0.75	0.01	2.92 <sup>ns</sup>
Foxtail millet	Intercept	1.18	-	-	7.18**	Intercept	-0.19	-	-	0.03
	PD	0.77	0.32	0.32	27.4**	FWF	1.06	0.65	0.65	373**
	PL	-0.25	0.45	0.16	16.7**	PH	0.37	0.72	0.06	47.92**
	PW	0.21	0.51	0.04	10.3**	FLL	0.15	0.76	0.01	27.11**

ns,\* and \*\*: non-significant and significant at the 5% and 1% levels of probability, respectively

## Discussion

Significant improvement in adaptation of millet to stress-prone environments will increase the effectiveness of breeding programs. This success achieved through field-based empirical selection for high yielding cultivars for drought affected areas. Simultaneous analysis of multiple parameters to increase the accuracy of the genotype ranking is the most important advantage of using a multivariate analysis in screening of genotypes. Field screening in drought affected areas accelerate the identification of promising genotypes that may be eventually released as new cultivars.

In this study, interaction between genotype and drought was significant for PW, PLW, DF and GY. This variation can be explained with the fact that traits suitable for a drought affected environment with specific climatic conditions may be unsuitable in another condition. Highly significant differences were observed among genotypes which showing considerable variation in Iranian millet ecotypes. These results were in accordance with the results of Upadhyaya et al. (2011) which reported significant differences for these traits in ICRISAT millet collection. Based on the study carried out in bread wheat, Molasadeghi and Dadbakhsh (2011) reported that there is significant difference between genotypes in terms of weight of spikelets after flowering in drought condition due to pollen sterility during stress period, which is ascribed to abnormal photosynthesis and transportation of photosynthate product in to spikelet. Resultantly, significant weight reduction in spikes was observed among genotypes as reported earlier.

Ecotypes viz., Esfahan (*Panicum*) and Yazd-1, Yazd-2, Yazd-4, Kerman-1, Kerman-2, Tabriz-4 and Elam-3 belonging to *Setaria* group gave higher yield as compared to four checks. Ecotypes belonging to *Panicum* group, viz., Kerman-4, Esfahan-1, Zabol-3 and Mazanderan-3 and Yazd-2, Mashhad-2, Tabriz-3 and Ilan-3, all from *Setaria* group produced higher dry weight fodder in comparison to four checks. Therefore, these genotypes were found suitable for stressed conditions and appeared to cope better with moisture stress. In this study, among all the ecotypes and according to means comparisons over two years under water deficit and normal conditions an ecotype, Kerman-2 was the most productive one in both years ( $2.2 \text{ t ha}^{-1}$ ). This value reported in the present study is more than the amount ( $1.118 \text{ t ha}^{-1}$ ) reported earlier by FAO and Upadhyaya et al. (2011) in a different study.

Ecotype from Zabol-3 (*Panicum*) showed the higher value ( $21.77 \text{ t ha}^{-1}$ ) for DWF. Non of the millet species studied under both the conditions showed reduction in measured traits in either normal or stress conditions except the DM which showed the significant reduction, most likely due to early maturity. In such cases escape mechanism plays an crucial role under drought stress. The number of tillers declined under water deficit conditions. Similar results have been reported in a number of studies that have shown reduction of tiller number caused by drought stress in different crops (Ludlow and Muchow 1990) and in pearl millet by Mahalakshmi and Bidinger (1985). This reduction in number of tillers affects the transpiration area and hence helps the plant to withstand against water stress.

Inducing moisture stress especially at fragile development stages (shoot elongation onwards) has resulted in reduction of plant height. As a result, reduction in and photosynthesis area has lowered sink size (product). Insufficient irrigation may also reduce the plant growth and height (Bruck et al. 2000). Therefore, an increased yield potential may be influenced by production of biomass characteristics (Natu and Ghildiyal 2005).

In the present study, foxtail millet produced more number of leaves per plant than proso millet contributing towards higher biomass. Grain filling period was also longer in foxtail millet in comparison to proso millet. In addition, the time of maturity in foxtail millet was longer than proso millet as already observed in sub-species of *Setaria* (Li et al. 1996). On the other hand foxtail millet gives higher grain yield and dry weight fodder under both stress and non-stress conditions than proso millet. Thus, the findings suggest that even though foxtail millet has higher yield potential but under drought stress proso millet complete its vegetative stage earlier than foxtail, most probably due to its earliness, which encourages escape from water stress.

Results of regression analysis in both millet species for grain yield indicated that, selection of PD, PW and PL would encourage the breeders to achieve higher grain yield under drought stress. The regression on DWF indicated 75 and 76% of variation explained by PH, FWF and FLL in proso and foxtail millet, respectively. Path analyses in proso millet indicated that, PW play a major role for determining grain yield in millet under drought. Plant height had positive and direct effect on DWF. The results of path analyses in foxtail millet showed that PH had the largest direct

effect on grain yield and FWF had the highest positive direct effect on dry weight fodder.

In conclusion, these traits are the foremost factor responsible for grain yield and dry weight fodder under drought stress. Thus selection based on these traits would be preferable. The present report is a part of a comprehensive breeding program under taken for screening the drought tolerance in millet germplasm for identifying high yielding promising genotypes in Iran. It may further be concluded that, application of all multivariate analysis simultaneously is a good approach for screening drought adapted genotypes. Eight ecotypes with the higher grain yield and another set of 8 with higher dry weight fodder in were identified from Iranian millet germplasm. These ecotypes could be recommended as promising genotypes for their eventual release as new cultivars through a national system for appropriate drought affected areas of Iran.

#### Authors' contribution

Conceptualization of research (GMN, BN, EMH, FDK); Designing of the experiments (GMN, BN, FDK); Contribution of experimental materials (GMN, BN, HV); Execution of field/lab experiments and data collection (HV, GMN); Analysis of data and interpretation (GMN, HV); Preparation of manuscript (GMN, HV, BN).

#### Declaration

The authors declare no conflict of interest.

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