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CORRELATION AND PATH COEFFICIENT ANALYSES OF DRY WEIGHT YIELD COMPONENTS IN THE COMMON SAINFOIN (*ONOBRYCHIS VICIIFOLIA* SCOP.)

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Abstract: Sainfoin (*Onobrychis viciifolia* Scop.) is a perennial forage crop with desirable forage properties adapted to temperate climate conditions. The purpose of this research was to study the phenotypic correlation coefficients between dry forage yield and some morphological traits, and to identify the direct and indirect effects of the associated traits. Thus, 32 ecotypes (landraces) were assessed in the randomized complete block design layout with four replications. Positive and statistically significant correlations were determined between total dry weight (TDW) and all measured traits except for internode length (IL) [$r=0.29$, $P>0.05$]. Regarding the variance inflation factor (VIF) as a multicollinearity statistic, number of nodes per main stem (VIF=1407.4) and number of internodes per main stem (VIF=1371.6) were removed from the analysis. Path coefficient analyses indicated that number of leaflets per leaf (NLL) [0.59 direct effect], height of the longest stem (HLS) [0.42 direct effect], and dry weight/fresh weight ratio (DFR) [0.27 direct effect] were influenced by TDW as a first-order trait. Five traits considered secondary or tertiary traits affected TDW – number of stems per area (NPA), number of stems per plant (NSP), number of leaves per stem (LS), length of inflorescence (LI) and stem weight/leaf weight ratio (SLR). The importance of main stem properties such as length or height, number of leaves, and number of leaflets can be used for selection in breeding programs aimed at improving common sainfoin forage yield under semi-arid conditions.

Key words: bootstrapping, dry forage yield, morphological traits, multicollinearity.

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Introduction

Common sainfoin (*Onobrychis viciifolia* Scop.) (Figure 1) is a perennial legume crop with acceptable productivity potential as a forage crop, with lower soil requirements and yields even in the poorest soils due to its high tolerance to abiotic stresses such as cold and drought (Radovic et al., 2019). It has erect or suberect hollow stems with a height of 70 cm, which can grow up to one meter tall as a cross-pollinated plant with many pink or purple flowers. It is a relatively little known forage crop compared to famous forage crops such as alfalfa or clovers, but it has attracted new interest in recent years in the world because it does not cause bloat in grazing animals, and can provide high-quality forage (Bhattarai et al., 2018). The regrowth of common sainfoin is relatively slow and it is important to give it sufficient time to replenish its root reserves to maintain its persistence and longevity (Carbonero et al., 2011). In addition, its blossoms produce large amounts of nectar, which is also attractive to pollinating insects such as honey bees.

Nowadays, common sainfoin cultivation is not widespread because its production is dependent on old, low-yielding cultivars or on local ecotypes whose future is not clear. Further expansion of common sainfoin production requires the specification of genetic resources, both for their protection and for their potential use. Common sainfoin is native to south-central Asia as a cross-pollinated crop (Burton and Curley, 1968), and the eastern Mediterranean and western Asia, especially Iran and Turkey, are considered to be the center of diversity of this species. In Iran, there are some landraces and ecotypes of common sainfoin that have been cultivated for centuries. In the northwestern and western Iran, farmers still cultivate old common sainfoin ecotypes that have good tolerance to abiotic stresses and could be an important genetic resource. Therefore, collecting and evaluating these promising ecotypes could be useful for the future breeding program.

Determining dry weight yield components will provide important benefits in common sainfoin breeding research in the future. The correlation coefficients are important in plant breeding programs because they quantify the degree of genetic and non-genetic association between two or more traits, allowing the indirect selection. However, pairwise correlation coefficients between dry weight yield and its components may not provide satisfactory results because the direct and indirect effects are not identified. In practice, selection indices based on the most important direct effects are used instead of correlation coefficients (Sharifi and Ebadi, 2018). Path analysis provides useful coefficients to construct selection indices that have been used successfully in several crops. This statistical tool is useful to identify the direct impact of one trait on another and it also separates the simple correlation coefficient into its direct and indirect effects (Takele et al., 2022). The correlation of yield performance with the other traits in common sainfoin and its partitioning into direct and indirect effects have been studied (Binek, 1983). The study of associations among traits is important for the early selection or the simultaneous selection when more than one trait is desired. However, the indirect selection using fewer complex traits with high heritability is simple and practical and may result in higher genetic progress compared to direct selection.

Figure 1. The experimental field of *Onobrychis viciifolia* Scop.

Ditterline (1973) has reported that traits such as number of stems per plant, number of branches per stem, number of florets per branch, number of seeds per branch, hundred seed weight, and seed yield are the main components in yield performance of common sainfoin. Binek (1983) has found that the number of stems has affected directly the number of florets and the number of pods in common sainfoin. Baghainiya et al. (2012) showed that dry forage yield of common sainfoin had a significant positive correlation with stem percentage, number of plants per area, and number of nodes per main stem. The utilization of such information will be of benefit in future plant breeding programs, as it may contribute to the development of genetically improved cultivars with a wide genetic base. Therefore, the aim of this study was to evaluate the association among different morphological traits of local ecotypes of common sainfoin in order to find the important traits for selection of forage yield performance useful for future breeding projects.

Material and Methods

During the 2021 regular cropping season, 32 local ecotypes of common sainfoin were studied in field experiments at the field station of University of Maragheh (37°23′21″N 46°14′15″E) under lowland conditions. Some geographical properties and annual rainfall patterns of the collection areas are shown in Table 1. The Maragheh region in northwestern Iran has a typical cool, sub-humid, temperate climate and a sandy loam soil type texture which is classified as Regosols with 1.8% organic matter (Roozitalab et al., 2018). In autumn, 30 kg ha^{-1} nitrogen and 50 kg ha^{-1} P_2O_5 fertilizers were incorporated into the soil by tillage. The experimental design was a randomized block design with four replicates, and the plots consisted of four rows, 2-m long, with a spacing of 0.25 m between rows and 0.20 m between plants. The two central rows were considered as useful harvesting area for forage yield in terms of total dry weight (TDW). In addition, number of plants per area (NPA) and stem weight/leaf weight ratio (SLR) were measured in the two central rows. Each replication was considered an observation, making up ten sample observations randomly selected from the middle row of each plot. The following traits were observed and measured: number of stems per plant (NSP), number of nodes per main stem (NMS), height of the longest stem (HLS), peduncle length (PL), length of inflorescence (LI), number of internodes per main stem (IMS), internode length (IL), number of leaves per main stem (LMS), number of leaves per stem (LS), and number of leaflets per leaf (NLL).

The dataset was tested for normality and was subjected to analysis of variance using an appropriate model with randomized block design. Simple phenotypic correlation coefficients were computed and were separated into direct and indirect effects using path analysis. A stepwise multiple regression model was performed to detect the predictor traits in first-, second- and third-order paths according to their respective contributions to the total variation in dry forage yield as well as the minimal multicollinearity. The magnitude of multicollinearity in each pathway was evaluated using the tolerance and the variance inflation factor (VIF) statistics. The tolerance statistic is the amount of variation in the selected independent trait that is not described by other independent traits ($1-R^2$), where R^2 is the coefficient of determination. The VIF statistic shows the magnitude of effects of other independent traits on the variance of the selected independent traits [$VIF = 1/(1/R^2)$]. Thus, small tolerance values (much lower than 0.1) or high VIF values (> 10) show high multicollinearity. Partial R^2 values were calculated from the path coefficients for all predictor traits. We were interested in obtaining not only a point estimate of a path coefficient, but also an estimate of its variation and a confidence interval.

Table 1. Geographical properties of collection areas of sainfoin ecotypes.

Code	Name	Coordinates	Altitude	Rainfall
G1	Bonab	37°20′N 46°03′E	1290	370
G2	Sarab	37°56′N 47°32′E	1650	285
G3	Marand	38°25′N 45°46′E	1344	526
G4	Zonuz	38°35′N 45°49′E	1700	480
G5	Varzaqan	38°30′N 46°39′E	1670	350
G6	Ahar	38°28′N 47°04′E	1341	340
G7	Azarshahr	37°45′N 45°58′E	1384	250
G8	Tabriz	38°04′N 46°18′E	1348	310
G9	Heris	38°14′N 47°06′E	1900	315
G10	Miandoab	36°58′N 46°06′E	1314	289
G11	Urmia	37°32′N 45°04′E	1332	338
G12	Silvaneh	37°25′N 44°51′E	1587	300
G13	Oshnavieh	37°02′N 45°05′E	1411	450
G14	Azna	33°27′N 49°27′E	1871	496
G15	Khorramabad	33°29′N 48°21′E	1147	412
G16	Aligudarz	33°24′N 49°41′E	2022	390
G17	Khalkhal	37°37′N 48°31′E	2243	320
G18	Garjan	38°18′N 48°12′E	1500	295
G19	Kahlaran	38°18′N 48°12′E	1500	295
G20	Meshginshahr	38°23′N 47°40′E	1400	380
G21	Sanandaj	35°18′N 46°59′E	1450	500
G22	Divandarreh	35°54′N 47°01′E	1850	275
G23	Khomeyn	33°38′N 50°04′E	1830	296
G24	Arak	34°05′N 49°41′E	1743	341
G25	Saqqez	36°14′N 46°15′E	1476	500
G26	Asadabad	34°46′N 48°07′E	1607	403
G27	Zanjan	36°40′N 48°29′E	1663	300
G28	Damavand	35°43′N 52°03′E	2051	385
G29	Faridan	32°59′N 50°24′E	2390	350
G30	Khansar	33°13′N 50°18′E	2215	453
G31	Fereydunshahr	32°56′N 50°07′E	2530	450
G32	Kabutarabad	32°29′N 51°49′E	1545	110

Resampling methods such as the bootstrap procedure provide estimates of the standard error values. Thus, the mean direct effects estimated from a set of 1,000 bootstrap samples agreed well with the observed direct effects of the various traits. To estimate the standard error of the path coefficients, the bootstrap procedure was performed. All statistical analyses were carried out using S-Plus Version 2000 (MathSoft, 1999), IBM SPSS AMOS Version 20.0 (Arbuckle, 2011), and SPSS Version 14.0 (SPSS, 2004).

Results and Discussion

The results of the analysis of variance showed significant differences in common sainfoin ecotypes for all of the measured traits (data not shown). The results of correlation coefficient analysis (Table 2) showed that there were highly positive correlations between total dry weight (TDW) and all measured traits except for internode length (IL). Therefore, to identify the most reliable pattern of the associations and to determine the magnitudes of the direct and indirect effects of the measured traits on TDW, performing a path coefficient analysis is essential. All traits were positively and significantly correlated with number of plants per area (NPA) and height of the longest stem (HLS), except for dry weight/fresh weight ratio (DFR). It is interesting that DFR had no significant positive/negative correlation with the measured traits except for TDW and stem weight/leaf weight ratio (SLR). There was a statistically significant and positive correlation between number of stems per plant (NSP) and other common sainfoin characters except for IL and DFR. The peduncle length (PL) had significant and positive correlations with length of inflorescence (LI), number of leaves per main stem (LMS), and IL. The length of inflorescence was significantly and positively correlated with number of leaves per stem (LS) and number of leaflets per leaf (NLL), IL and LMS. We also found a significant and positive correlation between IL and LMS, and between LMS with LS and NLL. The positive significant correlations were observed between LS with SLR and NLL, and between SLR and NLL.

Table 2. Correlation coefficients between 14 traits of 32 common sainfoin (*Onobrychis viciifolia* Scop.) genotypes.

	NPA*	NSP	HLS	PL	LI	IL	LMS	LS	NLL	SLR	DFR
NSP	0.679**										
HLS	0.581	0.792									
PL	0.445	0.667	0.632								
LI	0.472	0.734	0.743	0.753							
IL	0.427	0.280	0.506	0.287	0.359						
LMS	0.639	0.760	0.689	0.599	0.773	0.531					
LS	0.576	0.863	0.701	0.716	0.670	0.138	0.668				
NLL	0.468	0.505	0.484	0.198	0.372	0.169	0.466	0.498			
SLR	0.373	0.490	0.416	0.229	0.225	0.136	0.355	0.523	0.464		
DFR	0.344	0.160	0.139	-0.029	-0.004	0.135	0.206	0.078	0.195	0.635	
TDW	0.672	0.714	0.699	0.542	0.568	0.294	0.623	0.666	0.730	0.495	0.357

**Critical values of correlation $P < 0.05$ and $P < 0.01$ (D.F. 30) are 0.35 and 0.45, respectively; *Abbreviations are: number of plants per area (NPA), number of stems per plant (NSP), number of nodes per main stem (NMS), height of the longest stem (HLS), peduncle length (PL), length of inflorescence (LI), number of internodes per main stem (IMS), internode length (IL), number of leaves per main stem (LMS), number of leaves per stem (LS), number of leaflets per leaf (NLL), stem weight/leaf weight ratio (SLR) and total dry weight (TDW).

To identify the relative importance of the measured traits for the target trait (TDW), the dataset was subjected to multiple linear regression analysis, path analysis, two statistics of multicollinearity analysis, tolerance and VIF were computed (Table 3). Within the analysis, all traits were considered as first-order variables (Model I) with TDW as the response variable. The result showed high multicollinearity for number of nodes per main stem (NMS) and number of internodes per main stem (IMS). These traits show high direct effects on TDW, but their multicollinearity statistics were very high, VIF = 1407.4 for NMS and 1371.6 for IMS, thus, these traits were removed from the analysis. The estimation of direct effects by path analysis was considered by removing the NMS and IMS traits (Model II), and the analysis of multicollinearity showed a better understanding of the associations among the measured traits and their relative contribution to TDW. The results of the tolerance and VIF values for the predictor traits did not show any remarkable reduction in the VIF values in Model II compared with Model I.

Table 3. Direct effects of first-order predictor variables on the dry forage yield of 32 common sainfoin (*Onobrychis viciifolia* Scop.) genotypes and two measures of collinearity in path analysis; Model I (all predictor traits used as first-order variables) and Model II (the traits with high collinearity were removed).

Traits	Model I			Model II		
	Direct effect	Tolerance	VIF*	Direct effect	Tolerance	VIF
NPA	0.145	0.354	2.8	0.128	0.369	2.7
NSP	0.086	0.136	7.4	0.114	0.141	7.1
NMS	2.120	0.001	1407.4			
HLS	-0.059	0.103	9.7	0.228	0.224	4.5
PL	0.128	0.183	5.4	0.300	0.292	3.4
LI	0.028	0.154	6.5	-0.086	0.208	4.8
IMS	-1.769	0.001	1371.6			
IL	0.023	0.363	2.8	-0.046	0.409	2.4
LMS	-0.131	0.187	5.3	-0.046	0.205	4.9
LS	0.053	0.140	7.1	0.020	0.142	7.1
NLL	0.561	0.498	2.0	0.512	0.550	1.8
SLR	-0.169	0.289	3.5	-0.160	0.293	3.4
DFR	0.291	0.374	2.7	0.287	0.384	2.6

*VIF: variance inflation factor; **Abbreviations are: number of plants per area (NPA), number of stems per plant (NSP), number of nodes per main stem (NMS), height of the longest stem (HLS), peduncle length (PL), length of inflorescence (LI), number of internodes per main stem (IMS), internode length (IL), number of leaves per main stem (LMS), number of leaves per stem (LS), number of leaflets per leaf (NLL), stem weight/leaf weight ratio (SLR) and total dry weight (TDW).

The adjusted coefficient of determination (R^2 = 73.3) indicates the influence of the NLL, PL, and DFR traits as first-order traits that contribute to exploring the total variation of TDW (Table 4). Of the three traits influencing TDW, the NLL had the greater direct effect (0.594) than PL and DFR. The PL had the greater direct effect (0.418) than DFR on TDW. The indirect effect of NLL via PL and DFR was relatively low and positive (Table 5). For a better understanding of the association among the traits, a graphical representation of the results can be useful, thus, the diagram of path analysis (Figure 2) was generated. The results of the path analysis, when the first-order traits were used as response traits, showed that NSP positively influenced NLL and accounted for more than 50% of the observed variation while LI and LS positively influenced the PL and accounted for more than 62% of the observed variation (Table 4). The indirect effect of LI on PL via LS and the indirect effect of LS on PL via LI were relatively moderate. Finally, the last first-order trait (DFR) was influenced positively by SLR and NPA while it was influenced negatively by LS, and more than 53% of its total variation was explained by these second-order traits. The indirect effect of SLR on DFR via LS was moderate and negative, whereas the indirect effect of SLR on DFR via NPA was relatively low and positive. The indirect effect of LS on DFR via SLR was relatively high and positive, while the indirect effect of LS on DFR via NPA was relatively moderate and positive. The indirect effect of NPA on DFR via SLR and LS was relatively high and positive.

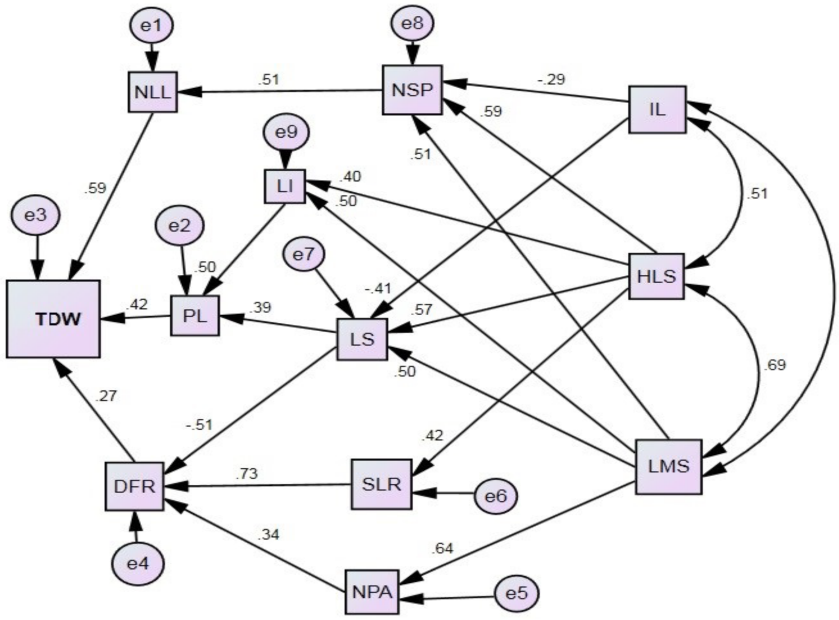

Figure 2. Path analysis diagram illustrating the associations among morphological traits contributing to dry forage yield.

Abbreviations are: number of plants per area (NPA), number of stems per plant (NSP), height of the longest stem (HLS), peduncle length (PL), length of inflorescence (LI),), internode length (IL), number of leaves per main stem (LMS), number of leaves per stem (LS), number of leaflets per leaf (NLL), stem weight/leaf weight ratio (SLR) and total dry weight (TDW).

Table 4. The estimation of coefficients of determination, direct effects, collinearity statistics and standard error values of path coefficients.

Response	Predictor	Path analysis coefficients		Collinearity statistics		Bootstrap statistics		
Trait	Trait	R^2*	Direct effect	Tolerance	VIF	Mean	Bias	SE
TDW**	NLL	51.7	0.594	0.921	1.1	0.593	-0.001	0.1
	PL	67.6	0.418	0.956	1.0	0.420	0.001	0.1
	DFR	73.3	0.273	0.957	1.0	0.272	-0.001	0.1
NLL	NSP	23.0	0.505	1.000	1.0	0.506	0.001	0.2
PL	LI	55.3	0.497	0.552	1.8	0.504	0.008	0.1
	LS	62.5	0.386	0.552	1.8	0.384	-0.001	0.1
DFR	SLR	38.3	0.732	0.718	1.4	0.745	0.014	0.2
	LS	45.7	-0.511	0.558	1.8	-0.512	-0.001	0.2
	NPA	53.4	0.344	0.661	1.5	0.327	-0.017	0.2
NSP	HLS	61.5	0.588	0.497	2.0	0.654	0.066	0.2
	LMS	69.5	0.507	0.480	2.1	0.497	-0.010	0.1
	IL	74.5	-0.286	0.680	1.5	-0.436	-0.150	0.3
LI	LMS	58.4	0.497	0.525	1.9	0.502	0.005	0.1
	HLS	65.9	0.401	0.525	1.9	0.399	-0.002	0.2
LS	HLS	47.5	0.568	0.497	2.0	0.562	-0.006	0.1
	LMS	52.6	0.496	0.480	2.1	0.503	0.007	0.1
	IL	63.8	-0.414	0.680	1.5	-0.397	0.017	0.2
SLR	HLS	14.6	0.416	1.000	1.0	0.404	-0.012	0.2

*R^2, coefficient of determination; **Abbreviations are: number of plants per area (NPA), number of stems per plant (NSP), height of the longest stem (HLS), peduncle length (PL), length of inflorescence (LI), internode length (IL), number of leaves per main stem (LMS), number of leaves per stem (LS), number of leaflets per leaf (NLL), stem weight/leaf weight ratio (SLR) and total dry weight (TDW).

To identify the third-order traits as predictors, the second-order traits were adopted separately as response traits. The results showed that IL negatively influenced NSP, whereas HLS and LMS positively influenced NSP and accounted for about 75% of the observed variation. The indirect effect of HLS on NSP via LMS was relatively high and positive, whereas the indirect effect of HLS on NSP via IL was relatively low and negative (Table 5). The indirect effect of LMS on NSP via HLS was relatively high and positive, whereas the indirect effect of LMS on NSP via IL was relatively moderate and negative. The indirect effect of IL on NSP via HLS and LMS was relatively high and positive. LMS and HLS also positively influenced LI and accounted for about 66% of the variation, whereas SLR was positively influenced by HLS (Table 4). The indirect effect of LMS on LI via HLS and the indirect effect of HLS on LI via LMS were relatively high and positive (Table 5). Finally, HLS and LMS positively influenced LS, whereas IL negatively influenced LS, and together these traits accounted for about 64% of observed variability. The indirect effect of LI on PL via LS and the indirect effect of LS on PL via LI were relatively high and positive (Table 5).

Table 5. Direct and indirect effects for the predictor traits in the path analysis divided into first-, second- and third-order traits.

TDW*	NLL	PL	DFR	DFR	SLR	LS	NPA
NLL	0.594	0.083	0.053	SLR	0.732	-0.267	0.128
PL	0.117	0.418	-0.008	LS	0.383	-0.511	0.198
DFR	0.116	-0.012	0.273	NPA	0.273	-0.294	0.344
NSP	HLS	LMS	IL	LS	HLS	LMS	IL
HLS	0.588	0.349	-0.145	HLS	0.568	0.342	-0.209
LMS	0.405	0.507	-0.152	LMS	0.392	0.496	-0.220
IL	0.298	0.269	-0.286	IL	0.288	0.264	-0.414
PL	LI	LS		LI	LMS	HLS	
LI	0.497	0.258		LMS	0.497	0.276	
LS	0.333	0.386		HLS	0.342	0.401	

*Abbreviations are: number of plants per area (NPA), number of stems per plant (NSP), height of the longest stem (HLS), peduncle length (PL), length of inflorescence (LI),), internode length (IL), number of leaves per main stem (LMS), number of leaves per stem (LS), number of leaflets per leaf (NLL), stem weight/leaf weight ratio (SLR) and total dry weight (TDW).

Morphological traits have been used to explain the variability of different crop landraces, and no comprehensive studies have been conducted for common sainfoin (*Onobrychis viciifolia* Scop.), with the exception of Binek (1983). The results of these studies indicate a high variation in measured traits among accessions and ecotypes of various geographical regions (Mohajer et al., 2013; Bhattarai et al., 2018) and we found relatively high variation in our plant materials and measured traits. A total of fourteen traits were used to assess thirty-two common sainfoin landraces, highlighting the remarkable potential to improve this species by breeding programs. As for common sainfoin breeding and selection of the most favorable individuals, it is important to reveal the variation available for the crop pattern and yield components. A better understanding of how forage yield components influence forage yield formation can be determined by path analysis, which indicates the direct and indirect effects of primary, secondary, and tertiary traits on forage yield formation. In addition, the path analysis shows how traits indirectly influence the yield performance through other traits and it provides more information on the relationship between traits than simple correlation coefficients (Kozak and Kang, 2006). A significant and positive correlation between forage yield (TDW) of common sainfoin and number of stems per plant (NSP), number of branches per stem, and number of leaflets per branch was reported by Hasanzadeh-Gorttapeh et al. (2014). In addition, Bhattarai et al. (2018) found a positive and significant correlation among TDW, NSP and HLS in common sainfoin, and

Mohajer et al. (2013) found similar results in terms of correlations between NSP and PL with other important traits such as TDW, HLS, LMS and NLL in common sainfoin. Some authors have reported similar results about the highly positive and significant correlation of number of stems per plant and number of stems per area with forage yield of common sainfoin (Veisipoor et al., 2012; Hasanzadeh-Gorttapeh et al., 2014; Bhattarai et al., 2018), while in this research, these traits did not show any direct effects as primary variables on forage yield, but had indirect effects as secondary variables on forage yield formation. Veisipoor et al. (2012) reported number of stems per plant and dry matter percent as the main variables in forage yield determination of common sainfoin, while Najafipoor and Majidi (2017) found number of stems per plant and number of seeds per inflorescences as the main traits in seed yield performance. Zarabiyan et al. (2015) also found that number of leaves per stem and number of stems leaves plant played a role in the path analysis of common sainfoin.

The direct effects estimated from a set of 1,000 bootstrap samples and the results indicated that the standard error values as well as the bias amounts for all the direct effects were low (Table 3), demonstrating the good robustness of Model II in path analysis. The path analysis might result in a multicollinearity of the traits. To avoid this problem, we removed the highly multicollinearized traits similar to other researchers who have employed this strategy in different crops: Mohammadi et al. (2003) in maize, Asghari-Zakaria et al. (2007) in potato, and Sabaghnia et al. (2010) in rapeseed. Our results indicate that there were three primary traits in forage yield formation, namely, number of leaflets per leaf (NLL), peduncle length (PL) and dry weight/fresh weight ratio (DFR), whereas Binek (1983) has reported only the number of inflorescences as the most important trait for yield performance. However, the role of NLL as one of the yield components in forage yield formation could not be neglected, whereas the role of PL and DFR traits in common sainfoin yield is logical. Similarly, Veisipoor et al. (2012) found peduncle length and dry weight/fresh weight ratio as the primary traits in forage yield. In contrast, Zarabiyan et al. (2015) reported plant height, number of leaves per stem, number of stems per plant and stem diameter as the variables directly influencing path analysis of common sainfoin, while we detected these traits as secondary or tertiary traits. This method of evaluating the association of different traits and regression analysis was used by Sabaghnia et al. (2010) in rapeseed, Janmohammadi et al. (2014) in bread wheat and Nayebi-Aghbolag et al. (2019) in rye.

In the next step of path analysis, we found five traits as secondary or tertiary traits, namely, number of stems per area (NPA), number of stems per plant (NSP), number of leaves per stem (LS), length of inflorescence (LI) and stem weight/leaf weight ratio. Similarly, Veisipoor et al. (2012) and Zarabiyan et al. (2015) found NPA and NSP as the traits determining forage yield, while Dadkhah et al. (2011)

reported NPA, NSP and LI as primary traits for seed yield performance. Thus, it seems that number of stems per plant or area is an important trait in common sainfoin and must be used in selection indices for genetic improvement programs. Regarding both primary and secondary traits, NSP→NLL path had the great impact on forage yield, followed by NPA→DFR path. Also, SLR→ DFR path and LI→PL path had a relatively remarkable impact on forage yield, but LS showed no such effect, having a positive effect on PL, but a negative effect on DFR. Thus, using number of leaves per stem in breeding programs must be done with caution due to its relatively complex role on forage yield formation. Finally, the remaining traits were identified as quaternary traits, including internode length (IL), number of leaves per main stem (LMS), and height of the longest stem (HLS). Similarly, Dadkhah et al. (2011) and Zarabiyan et al. (2015) found that plant height was the variable affecting forage yield. Also, Davazdahemami et al. (2019) reported number of leaves per plant as the contributing variable in common sainfoin forage yield. Thus, despite the number of leaves per stem (LS), number of leaves per main stem (LMS) had a remarkable impact on the forage yield. Regarding all primary, secondary, and tertiary traits, the sum of all paths of HLS had the great impact on forage yield, followed by the sum of all paths of LMS, but IL showed no such effect because the sum of its positive and negative effects was not high due to relatively equal positive and negative effects. Also, the relatively low magnitudes of the standard error of all the direct effects as well as the low bias amounts in the bootstrap procedure showed the robustness of our path analysis results. The T-test of significance using standard error values obtained through bootstrap resampling showed that all the direct effects were significant (data not shown). The used path analysis method in this study minimized the collinearity statistics (tolerance and VIF) of all traits facilitating the identification of the actual contribution of each predictor traits in the different paths, with negligible and confounding effects and interference. The advantage of this method in decreasing collinearity challenges and detecting real partnerships for each trait in different paths is similar to those reported in other crop studies (maize: Mohammadi et al., 2003; potato: Asghari-Zakaria et al., 2007 and rapeseed: Sabaghnia et al., 2010), indicating that it should be very useful in obtaining reliable result.

Conclusion

Generally, the correlation coefficients, regular path coefficients, and bootstrapping procedures in this study showed very close associations between forage yield performance and other morphological traits, with NLL, PL and DFR being the first-order variables, and with NSP, LI, LS, SLR, LS and NPA being the second-order variables. Finally, HLS, LMS and IL were the third-order variables associated with forage yield performance. The importance of main stem properties such as length or height, number of leaves and number of leaflets can be used for selection in breeding programs, with the aim of improving common sainfoin forage yield under semi-arid conditions.

References

Arbuckle, J.L. (2011). IBM SPSS AMOS 20 User's Guide. IBM Corporation, Armonk.

Asghari-Zakaria, R., Fathi, M., & Hasan-Panah, D. (2007). Sequential path analysis of yield components in potato. *Potato Research, 49,* 273-279.

Baghainiya, M., Majidi, M.M., & Mirlohi, A. (2012). Evaluation of agro-morphological traits of sainfoin genotypes (*Onobrychis viciifolia*) with natural and induced genetic variability. *Iranian Journal of Field Crop Science, 42* (4), 695-704.

Bhattarai S., Coulman, B., Beattie, A.D., & Biligetu, B. (2018). Assessment of sainfoin (*Onobrychis viciifolia* Scop.) germplasm for agro-morphological traits and nutritive value. *Grass and Forage Science, 73,* 958-966.

Binek, A. (1983). Correlation and analysis of path coefficients between features determining the seed yield structure in common sainfoin (*Onobrychis viciaefolia* Scop.). Zeszyty Problemowe Postepow Nauk Rolniczych (Poland).

Burton, J.C., & Curley, R.L. (1968). Nodulation and nitrogen fixation in sainfoin (*Onobrychis sativa* LAM.) as influenced by strains of rhizobia. In: Cooper, C.S., & Carleton A.E. ed., Sainfoin Symposium. *Bulletin - Montana State College, Agricultural Experiment Station, 627,* 3-5.

Carbonero, C.H., Mueller-Harvey, I., Brown, T.A., & Smith, L. (2011). Sainfoin (*Onobrychis viciifolia*): a beneficial forage legume. *Plant Genetic Resources: Characterization and Utilization, 9* (1), 70-85.

Dadkhah, M., Majidi M.M, & Mirlohi, A. (2011). Multivariate analysis of relationships among different characters in Iranian Sainfoin populations (*Onobrichis viciifolia* Scop.). *Iranian Journal of Field Crop Science, 42,* 349-357.

Davazdahemami, S., Ma-Alizadeh, M.A., Jalali, S., & Zeinali, H. (2019). Assessment of variation of sainfoin (*Onobrychis vicifolia* Scop.) genotypes through forage yield and its components. *Iranian Journal of Rangelands and Forests Plant Breeding and Genetic Research, 27,* 15-27.

Ditterline, R.L. (1973). Yield and yield components of sainfoin (*Onobrychis viciaefolia* Scop.) seed and an evaluation of its use as a protein supplement. Doctoral dissertation, Montana State University-Bozeman, College of Agriculture.

Hasanzadeh, Gorttapeh, A., Abbasi, Holasou, H., & Abbasi, M.R. (2014). A Study of Agronomy, Morphological and Phenological Traits of Sainfoin (*Onobrychis viciifolia* Scop) Using Multivariate Analysis. *Applied Crop Breeding, 2* (2), 153-169.

Janmohammadi, M., Sabaghnia, N., & Nouraein, M. (2014). Path analysis of grain yield and yield components and some agronomic traits in bread wheat. *Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis, 62* (5), 945-952.

Kozak, M., & Kang, M.S. (2006). Note on modern path analysis in application to crop science. *Communications in Biometry and Crop Science, 1,* 32-34.

MathSoft (1999). MathSoft, S-Plus 2000. Guide to Statistics, Volume 1. Data Analysis Products Division, MathSoft Inc, Seattle, WA.

Mohajer, S., Jafari, A.A., Mat-Taha, R., Yaacob, J.S., & Saleh, A. (2013). Genetic diversity analysis of agro-morphological and quality traits in populations of sainfoin (*Onobrychis sativa*). *Australian Journal of Crop Science, 7* (7), 1024-1031.

Mohammadi, S.A., Prasanna, B.M., & Singh, N.N. (2003). Sequential path model for determining interrelationships among grain yield and related characters in maize. *Crop Science, 43,* 1690-1697.

Najafipoor, A., & Majidi, M.M. (2017). Genetic analysis of seed yield components and its association with forage production in wild and cultivated species of sainfoin. *Journal of Crop Production and Processing, 6,* 1-17

Nayebi-Aghbolag, K., Sabaghnia, N., Pasandi, Somehsofla, M., & Janmohammadi, M. (2019). Study of correlation coefficients of agronomic traits and path analysis of seed yield in Rye. *Journal of Plant Productions, 42* (1), 31-46.

Radovic, J., Lugic, Z., Sokolovic, D., Babic, S., Jevtic, G., & Andjelkovic, S. (2019). Phenotyping diversity within a collection of sainfoin (*Onobrychis sativa* L.) populations. Research Collection, 400. Improving sown grasslands through breeding and management. *Proceedings of the Joint 20th Symposium of the European Grassland Federation and the 33rd Meeting of the EUCARPIA Section Fodder Crops and Amenity Grasses*, Zürich, Switzerland.Roozitalab, M.H., Toomanian, N., Ghasemi Dehkordi, V.R., & Khormali, F. (2018). Major soils, properties, and classification. *The soils of Iran,* 93-147.

Sabaghnia, N., Dehghani, H., Alizadeh, B., & Mohghaddam, M. (2010). Interrelationships between seed yield and 20 related traits of 49 canola (*Brassica napus* L.) genotypes in non-stressed and water-stressed environments. *Spanish Journal of Agricultural Research, 2,* 356-370.

Sharifi, P., & Ebadi, A.A. (2018). Relationships of rice yield and quality based on genotype by trait (GT) biplot. *Anais da Academia Brasileira de Ciências, 90,* 343-356.

SPSS Inc, 2004. SPSS 14. SPSS User's guide. SPSS Inc, Chicago, IL, USA

Takele, E., Mekbib, F. & Mekonnen, F. (2022). Genetic variability and characters association for yield, yield attributing traits and protein content of lentil (*Lens Culinaris* Medikus) genotype in Ethiopia. *CABI Agriculture and Bioscience, 3* (1), 1-14.

Veisipoor, A., Majidi, M.M., & Mirlohi, A. (2012). Traits relationship in Sainfoin (*Onobrychis viciifolia*) under normal and water stress conditions. *Iranian Journal of Field Crop Science, 42* (4), 745-756.

Zarabiyan, M., Majidi, M.M., & Bahrami, F. (2015). Relationship of morphological and agronomic traits in Iranian and exotic sainfoin populations using multivariate statistical analysis. *Iranian Journal of Rangelands and Forests Plant Breeding and Genetic Research, 22,* 278-290.

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ANALIZE KOEFICIJENATA KORELACIJE I PAT KOEFICIJENTA KOMPONENTI PRINOSA SUVE MASE KOD ESPARZETE (*ONOBRYCHIS VICIIFOLIA* SCOP.)

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R e z i m e

Esparzeta (*Onobrychis viciifolia* Scop.) je višegodišnja krmna kultura sa poželjnim krmnim svojstvima prilagođenim uslovima umerene klime. Cilj ovog istraživanja bio je da se ispitaju koeficijenti fenotipske korelacije između prinosa suve krme i nekih morfoloških osobina, kao i da se prepoznaju direktni i indirektni uticaji pridruženih osobina. Stoga su procenjena 32 ekotipa (sorte) u potpuno slučajnom blok dizajnu sa četiri ponavljanja. Utvrđene su pozitivne i statistički značajne korelacije između ukupne suve mase (USM) i svih merenih osobina osim dužine internodije (DI) [r=0,29, P>0,05]. Uzimajući u obzir faktor inflacije varijanse (FIV) kao statistički pokazatelj multikolinearnosti, broj nodija po glavnoj stabljici (FIV=1407,4) i broj internodija po glavnoj stablljici (FIV=1371,6) uklonjeni su iz analize. Analiza koeficijenata putanje pokazala je da je na broj listića po listu (BLL) [0,59 direktni uticaj], visinu najduže stabljike (VNS) [0,42 direktni uticaj], i odnos suve mase/sveže mase (OSS) [0,27 direktni uticaj] uticala USM kao osobina prvog reda. Pet osobina koje se smatraju sekundarnim ili tercijarnim osobinama uticale su na USM – broj stabljika po površini (BSP), broj stabljika po biljci (BSB), broj listova po stabljici (LS), dužina cvasti (DC) i odnos mase stabljike/mase lista (OSL). Važnost osobina glavne stabljike kao što su dužina ili visina, broj listova i broj listića može se koristiti za selekciju u programima oplemenjivanja koji imaju za cilj poboljšanje prinosa krme esparzete u polusušnim uslovima.

Ključne reči: butstraping, prinos suve krme, morfološke osobine, multikolinearnost.

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