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# Phenotypic variation of *Vicia pannonica* Crantz (var. *pannonica* and var. *purpurascens*) in central Turkey

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## Abstract

Hungarian vetch (*Vicia pannonica* Crantz.) is an important forage crop in both central Turkey (CT) and central European countries. It can be grown in CT as winter crop, but frequent spring droughts cause yield losses. Our objectives were (I) to investigate agro-biological variation among populations, (II) to identify useful plant characters and (III) to develop selection strategies. Forty-five accessions of both var. *pannonica* and var. *purpurascens* were evaluated for the 11 plant characters in 2002/03 cropping season. Var. *pannonica* was late flowering, had more stems and seeds per pod, and longer stems and pods. Var. *purpurascens* had earlier flowering, more days to physiological maturity, more pods, greater plant biomass and heavier seeds. As var. *purpurascens* was early flowering, its longer seed-filling period is likely to contribute to greater seed size and weight. However, in var. *pannonica*, late flowering and ensuing delayed maturity caused strong negative associations between days-to-harvest and other characters. If the earliness, higher biomass and seed size from var. *purpurascens*, and the many long stems and long pods from var. *pannonica* could be incorporated into one or more genotypes through Mendelian crossings, seed and hay yields would be substantially increased.

**Keywords:** Hungarian vetch, precocity, physiological maturity, biomass, seed yield, seed size

## INTRODUCTION

*Vicia pannonica* Crantz, one of the sixty *Vicia* species identified in the natural flora of Turkey, is an annual legume species, has decumbent or climbing stems (20-110 cm) and leaflets (4-10 pairs), and is geographically distributed in north, central, west and north-west parts of Turkey, in south and central Europe, north-west Africa, and Caucasia, and is subdivided into the two varieties; var. *pannonica* (yellowish or whitish-brown corolla) and var. *purpurascens* (dusky violet corolla) [5].

Provision of high quality feed supply for an ever-growing livestock population is indispensable in Turkish agriculture. Hungarian vetch (*Vicia pannonica* var. *pannonica*) with a high ability to adapt to the climatic and edaphic conditions is a valuable crop in both Turkey and central European countries. Because of its cold tolerance, it is planted in autumn in the central Turkey (CT), and has great potential for hay, straw and seed yield [2, 7]. Hungarian vetch was first introduced to the CT from Europe through the Çorum-Çankırı rural development project in early 1980's,

and its cultivation has substantially increased since then. Though it is not cultivated in considerably large quantities yet, pink vetch (*Vicia pannonica* var. *purpurascens*) has shown great potential particularly for higher seed in the CT [7]. Several vetch cultivars from both var. *pannonica* and var. *purpurascens* have been registered over the past fifteen years, but there is still a considerable scope for yield improvement.

Because the CT is characterized by cold winters and dry summers, plant growth greatly depends on weather conditions. For current cultivars, in general freezing temperatures in winter are not problematic. However, uneven distribution and shortages of rainfall and thus spring droughts cause yield losses. Andrews and McKenzie (2007) [3] suggested two strategies to combat the drought effects: (1) match the crop development with the period of soil moisture availability and (2) select the genotypes with rapid biomass development and early flowering and maturity. Phenotypic variability is the observable variation present in a diverse plant population [13, 16].

Hay or grain yield in a vetch crop is the result of the combination of many ecological, morphological and physiological characters that influence plant growth and development. Gaining information on their variability and relations, and developing proper breeding strategies will improve the efficiency of a vetch-breeding program. In order to achieve higher yield, Wallace et al. (1993) [15] suggested the spontaneous selection on three physiological components: (1) a higher biomass production, (2) a greater rate of seed yield accumulation so as to possess a high harvest index and (3) a well-synchronization of the plant growth duration (i.e., vegetative and reproductive stages) with the cropping season longevity in production environment. Moreover, vetch populations have development plasticity to environmental variation, selecting for more rapid crop development is likely to increase the yield in the CT [8]. Fırıncioğlu et al (2010) [9] concluded that in common vetch when selecting for improved seed yield under cold and drought stress conditions, earliness with faster seed filling and high biomass with higher biomass growth rate are the most pivotal characters in the CT.

Hence, to improve plant characters in *Vicia pannonica* varieties, it is imperative to gain insights into the magnitude of their variability, as this will provide the basis for effectual selection. However, there has been little research results available about *V. pannonica* var. *pannonica* and *V. pannonica* var. *purpurascens* so far. Our objectives, therefore, were to explore and quantify variation in var. *pannonica* and var. *purpurascens*, to identify pivotal plant characters for the yield improvement, and to develop selection strategies.

## MATERIALS AND METHODS

The trial was established in the first half of October in 2002 at the Research Farm of the Central Research Institute for Field Crops, located 44 km south-west of Ankara. A total of forty-five accessions of *V. pannonica*, obtained from both the local farmers and the ICARDA's germplasm development program, formed the experimental

material, of which *V. pannonica* var. *pannonica* (white flower color) and *V. pannonica* var. *purpurascens* (pink flower color) consisted of 20 and 25 entries, respectively. The experiment was laid out in completely randomized design with three replicates. Each line comprised of 30 spaced plants in three rows, 50 cm apart. In autumn 300 seeds of each line were sown into the drills, and in early spring seedlings were rarefied by hand to 50 cm spaced individual plants. Winter was cold in the experimental year; monthly average minimum temperatures were -9°C in December, -5°C in February and -2°C in March, and rainfall was 67% less in May and nil in June, compared to long term-average, 1982 to 2006 (54 and 49 mm), respectively. Therefore, late vegetative and reproductive stages of vetch crop became quite dry, then the trial was irrigated once at field capacity. Weeds were controlled by hoeing in spring.

Eleven plant characters were measured on individual spaced plants: (1) days-to-flowering (DF), number of days from sowing to the first flower appearance; (2) days-to-harvest (DH), number of days from sowing to harvest; (3) days-to-physiological maturity (DPM) were calculated by subtracting number of days to harvest by number of days to flowering; (4) stem length (cm) (SL), length of the main stem from crown to stem tip; (5) stem number (SN), number of stems arising from the crown; (6) pod number (NP), number of pods on a plant; (7) seed number per pod (NSP), number of seeds counted on the five randomly selected pods; (8) pod length (cm) (PL) is measured in randomly selected five pods; (9) plant biomass (g) (PB), at full maturity plants were harvested and weighed; (10) seed weight (g) (PSW), individual plants were threshed and the acquired seed was weighed; (11) 1000-seed weight (TSW) was calculated by dividing seed weight by seed number and multiplying by a thousand. For each plant character, data collected from thirty plants of each accession were combined by averaging, and descriptive statistics (i.e. mean, standard error of means, range and coefficient of variation), and t-test were performed to determine the differences between the two varieties for the plant characters studied. Relations among plant characters were investigated separately for each variety using simple Correlation Analysis (SCA) and Principle Component Analysis (PCA) with data standardization. MINITAB version 14.0 was used for all analysis.

## RESULTS

### Comparison of plant characters

The results of descriptive statistics and t-test, based on eleven metric characters of *V. pannonica* showed that there were significant differences between two varieties (Table 1). Though they had the same value of DH (268), var. *purpurascens* flowered 11 days earlier than var. *pannonica*, but reverse was true for DPM ( $P < 0.001$ ). Var. *pannonica* possessed significantly greater SN and longer SL (4.9 and 44.5 cm), compared to var. *purpurascens* (4.4 and 32.5 cm) ( $P < 0.05$  and  $P < 0.001$ ), respectively. Var. *purpurascens* had considerably greater NP (64.1) than that of var. *pannonica* (33.4) ( $P < 0.001$ ), whereas var. *pannonica* possessed significantly greater

NSP (4.3) and longer PL (2.9 cm) than those of var. *purpurascens* (1.9 and 2.1 cm), in that order ( $P<0.001$ ). Var. *purpurascens* acquired considerably heavier PB (14.9 and 10.5 g) and TSW (52.2 and 34.6 g) than those of var. *pannonica* ( $P<0.01$  and  $P<0.001$ ), correspondingly (Table 1). However, PSW did not differ between both varieties, and had the highest variation (47.1 and 45.1%), was followed by PB (30.2 and 38.4%), NP (37.9 and 36.5%) and SN (17.1 and 20.9%), respectively.

Table 1 Phenological and morphological characters of *Vicia pannonica* var. *pannonica* (PAN) and var. *purpurascens* (PUR.) and descriptive statistics (mean, standard error of mean (SEM), range and coefficient of variation (CV)) and significance of t-test.

Characters	Subspecies	Mean±SEM	Range	CV (%)	Significance of subspecies differences
Days to flowering (DF)	PUR	219.32±0.44	8.88	1.00	***
	PAN	230.31±0.88	12.97	1.70	
Days to harvest (DH)	PUR	267.92±0.23	4.62	0.42	ns
	PAN	267.68±0.42	6.81	0.71	
Days to physiological maturity (DPM)	PUR	48.59±0.41	7.45	4.20	***
	PAN	37.37±0.50	8.17	6.03	
Stem/plant (SN)	PUR	4.35±0.15	2.97	17.07	*
	PAN	4.90±0.23	4.27	20.89	
Stem length (cm) (SL)	PUR	32.45±0.78	15.48	12.01	***
	PAN	44.49±1.05	21.38	10.51	
Pod/plant (NP)	PUR	64.08±4.86	91.86	37.90	***
	PAN	33.41±2.73	53.22	36.53	
Seed/pod (NSP)	PUR	1.89±0.04	0.88	9.73	***
	PAN	4.32±0.07	1.12	7.58	
Pod length (cm) (PL)	PUR	2.08±0.01	0.27	3.33	***
	PAN	2.89±0.04	0.64	6.47	
Plant biomass/plant (g) (PB)	PUR	14.90±0.85	18.53	30.22	**
	PAN	10.49±0.90	17.28	38.36	
Seed weight/plant (g) (PSW)	PUR	2.76±0.26	5.18	47.12	ns
	PAN	2.40±0.24	4.92	45.07	
1000-seed weight (g) (TSW)	PUR	52.17±0.81	14.62	7.79	***
	PAN	34.64±0.48	6.74	6.24	

\*, \*\*, \*\*\* indicate significance at  $P<0.05$ ,  $P<0.01$  and  $P<0.001$ , respectively, and ns=not significant

Table 2 The correlation coefficients for the eleven plant characters measured on the 20 and 25 populations of var. pannonica and var. purpurascens respectively. (days to flowering (DF); days to harvest (DH); days to physiological maturity (DPM); stem number (SN), stem length (SL), number of pod (NP), seeds pod-1 (NSP), pod length (PL), plant biomass (PB), plant seed weight (PSW) and 1000-seed weight (TSW))

	DF	DH	DPM	SN	SL	NP	NSP	PL	PB	PSW
DH	0.383									
DPM	-0.858***	0.147								
SN	0.312	-0.204	-0.446*							
SL	0.107	-0.285	-0.277	0.386						
NP	-0.097	-0.582**	-0.223	0.348	0.867***					
NSP	0.136	0.022	-0.131	0.624**	0.055	-0.110				
PL	0.140	0.261	-0.001	-0.241	-0.548**	-0.515**	0.142			
PB	-0.030	-0.449*	-0.220	0.343	0.843***	0.957***	-0.018	-0.373		
PSW	-0.069	-0.520**	-0.218	0.165	0.782***	0.936***	-0.274	-0.360	0.927***	
TSW	-0.004	0.531**	0.301	-0.253	-0.519**	-0.571**	0.055	0.389	-0.448*	-0.545**
DH	0.935***									
DPM	-0.954***	-0.787***								
SN	-0.344	-0.475*	0.203							
SL	-0.468*	-0.602**	0.306	0.757***						
NP	-0.297	-0.352	0.220	0.852***	0.818***					
NSP	-0.591**	-0.708***	0.434	0.199	0.465*	0.139				
PL	-0.911***	-0.919***	0.811***	0.333	0.494*	0.219	0.769***			
PB	-0.296	-0.372	0.201	0.865***	0.836***	0.988***	0.133	0.211		
PSW	-0.538*	-0.604**	0.429	0.868***	0.892***	0.933***	0.385	0.511*	0.935***	
TSW	0.150	0.422	0.090	-0.475*	-0.483*	-0.331	-0.419	-0.233	-0.355	-0.347

\*\*\*, significant at P<0.05. 0.01, 0.001; Other values are not significant.

Relationships among plant characters

The correlation matrix was presented in Table 2, and showed strong relations between plant characters. Var. *pannonica* had more potent associations, compared to var. *purpurascens* (Table 2). In var. *pannonica*, DF was correlated significantly and positively with DH ( $r=0.935^{***}$ ) but inversely related with DPM ( $r=-0.954^{***}$ ), SL ( $-0.468^*$ ), SN ( $-0.591^{**}$ ), PL ( $-0.911^{***}$ ), and PSW ( $-0.538^*$ ), while DH had significant and negative relations with DPM ( $r=-0.787^{***}$ ), SN ( $-0.475$ ), SL ( $-0.602^{***}$ ), NSP ( $-0.708^{***}$ ), PL( $-0.919^{***}$ ) and PSW( $-0.604^{***}$ ). On the other hand, PSW significantly increased as SN ( $0.868^{***}$ ), SL ( $0.892^{***}$ ), NP ( $0.933^{***}$ ), PL ( $0.511^*$ ) and PB ( $0.935^{***}$ ) also increased significantly. TSW had negative relations with SN ( $-0.475^*$ ) and SL ( $-0.483^*$ ). In var. *purpurascens*, while DF produced only significant and inverse relation with DPM ( $r=-0.858^{***}$ ), DH was associated significantly and negatively with NP ( $r=-0.582^{**}$ ) and PB ( $r=-0.449^*$ ), PSW ( $-0.520^{**}$ ), but positively with TSW ( $r=0.531^{**}$ ). PB significantly increased with the increase of SL ( $0.843^{***}$ ) and NP ( $0.957^{***}$ ), while PSW showed positive correlation with SL ( $0.782^{***}$ ), NP ( $0.936$ ) and PB ( $0.927^{***}$ ). TSW increased with the decreased SL ( $-0.519^{***}$ ), NP ( $-0.571^{***}$ ), PB ( $-0.448$ ) and PSW ( $-0.545^{**}$ ).

Table 3 Eigenvalue of the correlation matrix for the estimated variables using the principle component analysis for *V. pannonica* var *pannonica* and var. *purpurascens*

Variables	var. <i>purpurascens</i>				var. <i>pannonica</i>			
	PC1	PC2	PC3	PC4	PC1	PC2	PC3	PC4
Days to flowering (DF)	-0.004	-0.578	-0.418	-0.060	0.311	0.349	-0.178	-0.227
Days to harvest (DH)	0.275	-0.171	-0.230	-0.658	0.341	0.277	0.093	-0.232
Days to physiological maturity (DPM)	0.159	0.522	0.322	-0.300	-0.254	-0.375	0.386	0.210
Stems/plant (SN)	-0.201	-0.423	0.440	-0.056	-0.312	0.299	0.011	0.265
Stem length/plant (cm) (SL)	-0.401	-0.052	-0.025	-0.352	-0.347	0.188	-0.082	-0.296
Pods/plant (NP)	-0.440	0.086	-0.016	-0.087	-0.304	0.360	0.181	-0.074
Seeds/pod (NSP)	0.015	-0.377	0.656	-0.063	-0.243	-0.282	-0.433	-0.638
Pod length (cm) (PL)	0.256	-0.121	-0.082	0.323	-0.305	-0.367	-0.035	-0.082
Plant biomass/plant (g) (PB)	-0.414	0.037	-0.021	-0.197	-0.306	0.367	0.159	-0.039
Seed weight/plant (g) (PSW)	-0.410	0.131	-0.192	-0.028	-0.365	0.204	0.141	-0.166
1000-seed weight (g) (TSW)	0.313	-0.002	0.016	-0.437	0.186	-0.129	0.733	-0.495
Eigenvalue	4.8727	2.2764	1.3255	0.9258	6.3328	2.6663	1.1446	0.4212
Proportion of variation	0.443	0.207	0.120	0.084	0.576	0.242	0.104	0.038
Cumulative proportion of variation	0.443	0.650	0.770	0.855	0.576	0.818	0.922	0.960



### Classifying the plant characters into major components

For both var. *pannonica* and var. *purpurascens*, the result of the PCA, based on highly correlated 11 plant characters, showed great diversity among 45 accessions of *V. pannonica* (Table 3). In both varieties, the first three principle components (PC) had Eigen values greater than 1.0, and together accounted for 92 and 77% of the total variation (TV) in the data set, respectively. In var. *pannonica*, scores on the first PC, which explained 58% of TV, were highly correlated with DF, DH, NS, SL, NP, PL, PB, and PSW. The second PC, 24% of TV, was associated with DF, DPM, NP, PL, and PB. In var. *purpurascens*, PC1, accounted for 44% of TV, was mainly loaded by characters associated with SL, NP, PB, PSW and TSW, and while PC2, explained 21% of TV, was mainly related with DF, DPM, NS and NSP.

## DISCUSSION

In this study, analysis of agro-biological characters revealed remarkable variation existed among 45 *V. pannonica* populations. Despite of this great variation, the results have ascertained that some characters would be useful to develop vetch cultivars. Moreover, multiple statistical analyses, based on 11 plant characters, successfully differentiated the variability that exists within and between two varieties.

Var. *pannonica* required the longer time period from sowing to flower, and possessed higher stem number per plant and seeds per pod, and longer stem and pod length, whereas var. *purpurascens* had the longer time span from flowering to harvest, greater number of pod, higher biomass and greater seed size (Table 1). Since var. *purpurascens* had less number of days to flower, its greater seed size can be attributed to longer duration for physiological maturity. Because seed size is the most stable yield component, the number of seeds per unit area is most likely to contribute to seed yield [4, 12]. Therefore, in this study it appeared that numbers of pods per plant and seeds per pod became quite important characters to improve seed yield. Low seed yield is also associated with the delayed appearance of floral buds and high abortion rates in flowers and young pods after fertilization in the onset of high temperatures in spring [1]. Selection for early flowering is likely to prolong the seed filling period and to render the larger seed mass [6, 11]. Whitehead et al. (2000) [14] suggested that early flowering allows the longer duration for seed-filling period. Siddique et al. (1996) [10] also emphasized on early flowering as an important character for vetches in short-season, Mediterranean environments. Hence, in this study, the lengthened seed-filling period through early flowering is likely to have accumulated more dry matter to seed in var. *purpurascens* .

The results of correlation analysis showed that some plant characters had stronger relations and var. *pannonica* produced more significant relations, compared to var. *purpurascens* (Table 2). Interestingly, in both varieties, seed weight per plant was strongly associated with plant biomass, and still more, these two characters were strongly correlated with others. In grain legume species, the positive relation between seed yield and plant biomass has been long-recognized [4, 6, 8, 9, 14]. While stem

length and pod number had strong associations with biomass and seed weight in both varieties, they were also potently correlated with stem number in var. *pannonica*. Thus, it appeared that var. *pannonica* is more appropriate for herbage type vetch whereas var. *purpurascens* is better for seed type vetch. Furthermore, phenological characters significantly produced more and strong inverse relations with morphological characters in var. *pannonica*, and indicated that precocity became a pivotal character for plant growth and development. The PCA revealed great diversity among the 45 accessions in *V. pannonica* Crantz. populations evaluated in this study (Table 3). The plant characters were successfully represented by two principle components, which accounted for 65.0 and 81.0% of TV in both varieties, respectively (Table 3). It is interesting to point out that there was not a single quantitative character to explain a reasonable extent of variation. Nevertheless, it appeared that morphological and phenological characters were more distinctly dispersed between the first two principle components in var. *purpurascens*, while they did relatively more equally in var. *pannonica*.

Overall, the results showed that some characters were more effective in discriminating the variability present. First, crop phenology is the most influential character in varieties. Because var. *purpurascens* was early flowering, its longer seed-filling period is likely to contribute to greater seed size. However, in var. *pannonica*, late flowering and resultant delayed maturity gave strong negative associations between days to harvest and other characters. This indicates that its growth period does not fit well with the time of soil moisture availability. Second, the variability reveals that distinctive plant attributes structurally differentiate two varieties. For example, var. *purpurascens* is a smaller plant, and has short stems, more pods with fewer larger seeds, whereas var. *pannonica* is a larger plant with many long stems, a few pods which are long and contain small seeds.

In conclusion, multiple statistical techniques used in this study are useful tools in describing the existing variation and identifying the most influential characters among vetch populations. It is possible to make crosses between these two varieties (Pers. Com. with Dr Ali Abd El Moneim). Therefore, if these pivotal plant attributes such as earliness, high biomass and great seed size from var. *purpurascens*, and many long stems and pods from var. *pannonica* could be incorporated into one or more plant genotypes through Mendelian crossings, both seed and hay yields could be substantially increased in *V. pannonica* Crantz.

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