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Multivariate analysis to assess abscisic acid content association with different physiological and plant growth related traits of *Petunia*

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ABSTRACT

Petunia is an important and beautiful ornamental flowering plant, grown throughout the world for its beauty and attraction. Different *Petunia* hybrids have been developed by petunia growing countries of the world. The prescribed study was conducted to investigate the association of abscisic acid with seed yield and its contributing traits of petunia line. Data for different physiological, morphological and petunia seed yield traits was recorded, analyzed and interpreted for final inferences. From results it was showed that the petunia lines IAGS-P8, IAGS-P9 and IAGS-P11 performed well for most of the studied traits. It was shown from multivariate analysis techniques that stomata conductance, chlorophyll b contents, seed area, chlorophyll a contents, flower fresh mass, flowers per plant, seed mass and abscisic acid contributed higher to seed yield per plant in petunia. The abscisic acid contents showed positive and significant association and contribution towards seed yield of petunia genotypes. It was suggested that selection on the basis of abscisic acid may be useful to develop good seed yield per plant and large number of flowers per plant in petunia under stressful environmental conditions.

Key words: petunia; multivariate analysis; heritability; genetic advantage; abscisic acid content; seed yield

IZVLEČEK

UPORABA MULTIVARIATNE ANALIZE ZA OCENITEV POVEZAVE MED VSEBNOSTJO ABSCIZINSKE KISLINE IN RAZLIČNIMI Z RASTJO POVEZANIMI FIZIOLOŠKIMI ZNAKI PRI PETUNIJI

Petunija je pomembna in lepa okrasna rastlina, ki se goji širom po svetu zaradi lepote in privlačnosti. V številnih državah, kjer jo gojijo, so bili vzgojeni različni križanci. Pričujoča raziskava je bila opravljena z namenom preučiti povezavo med vsebnostjo abscizinske kisline in lastnostmi, povezanimi s pridelkom semena preučevanih linij petunij. Izmerjene so bile različne fiziološke in morfološke lastnosti, ki vplivajo na pridelek semen, analizirana in pojasnjena je bila njihova povezava. Izsledki so pokazali, da so se linije petunij IAGS-P8, IAGS-P9 in IAGS-P11 izkazale kot primerne za večino analiziranih lastnosti. Multivariatna analiza je pokazala, da so parametri kot so stomatarna prevodnost, vsebnost klorofila b in a, površina semen, sveža masa cvetov, število cvetov na rastlino, masa semen in vsebnost abscizinske kisline prispevali največ k večjemu pridelku semena na rastlino. Vsebnost abscizinske kisline je imela značilen pozitiven vpliv na pridelek semena vseh genotipov petunij. Zaradi tega se priporoča, da je izbor genotipov petunije na osnovi večje vsebnosti abscizinske kisline primeren za vzgojo rastlin z velikim pridelkom semena na rastlino in velikim številom cvetov v stresnih okoljskih razmerah.

Ključne besede: petunija; multivariatna anliza; dednost; genetska prednost; vsebnost abscizinske kisline; pridelek semena

1 INTRODUCTION

The genus *Petunia* is an important ornamental plant of high economic imperativeness on the whole agriculture. It gives dominant qualities to serve as model plant for contemplating plant improvement. The history of *Petunia* development as a crop is accent with implications for an advanced array of added ornamental crops (Cantor et al., 2015; Gerats and Vandenbussche, 2005). This generally developed genus of flowering plants fits in with Solanaceae gang. It is an important ornamental plant in landscape because of colour diversity. A large number of hybrids and varieties have been developed with diverse color and patterns (Ganga,

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2011). This plant is native to Brazil, Argentina or Uruguay and included over 35 species (Dole and Wilkins, 1999). Petunias are considered an annual ornamental plant but may be perennial in warmer climates (Berenschot et al., 2008). Petunia genus includes about 20 species of South American origin, mostly perennials but developed as annuals (Mallona et al., 2010), 14 annual species (Toma, 2009), but *Petunia hybrida* Hort. (*P. axillaris* Lam. × *P. violacea* Lindl.) is a species which presents the biggest decorative value (Berenschot et al., 2008; Vandenbussche et al., 2016). Presently the researches have also confirmed that the genus *Petunia* is consisted of 14 closely related species (Stuurman et al., 2004). The modern petunias have been developed through hybrid breeding like the violet petunia (*P. violacea*, and *P. integrifolia* (Hook.) Schinz & Thell.) and ambrosial agrarian or white petunia (*Petunia axillaris*). A large number of researchers are also working on finding the real ancestors because their ancestors are rarely cultivated today. Petunia is a small sprawling plant with large number of flowers, grown throughout the world for its beauty and interactive colors (Anderson, 2006; Băla, 2007). Petunias blossom abundantly even in hot summers and new varieties even in seasons with top clamminess (Băla, 2007; Florin et al., 2012). The petunia is long day plant due to which it is used for landscape proposes (Anderson, 2006; Currey and Lopez, 2013). The present study was conducted to develop inbred lines of petunia through selfing for growing seasons. The data of various morphological, physiological and seed yield traits was recorded to access the performance of inbred lines under development. The identification of promising inbred lines for the development of petunia hybrids.

2 MATERIAL AND METHODS

Prescribed research work was conducted in the research area of Institute of Agricultural Sciences, University of the Punjab Lahore, Pakistan. Twelve petunia lines, IAGS-P1, IAGS-P2, IAGS-P3, IAGS-P4, IAGS-P5, IAGS-P6, IAGS-P7, IAGS-P8, IAGS-P9, IAGS-P10, IAGS-P11 and IAGS-P12 were selected and grown in the field during 2015. Selfing of all the lines was carried out for 4 successive growing seasons (2011-14) to develop inbred lines. The selfed seed was collected to develop next generation, grown in 2015 and for obtaining data for various traits, such as leaf temperature (LT), photosynthetic rate (A), stomata conductance (gs), water use efficiency (WUE), sub-stomata CO_2 concentration (Ci) and transpiration rate (E) (by using IRGA-LI-6262 (Infrared Gas Analyzer, LI-COR Biosciences designs, USA), chlorophyll a content (Chl. a), chlorophyll b content (Chl. b) in fresh matter (measured through the dimethyl sulfoxide extraction method (Hiscox and Israelstam, 1979), plant height (PH), leaves per plant (LPP), flowers per plant (FPP), leaf area (LA), stem diameter (SD), leaf length (LL), fresh leaf mass (FLM), seeds per fruit (SPF), leaf width (LW), flower mass (FM), seed mass (SM), fresh stem mass (FSM), seed area (SA, measusered by using Digital Micrometer Screw Guage, Model: 1658DGT/25), 100-seed mass (HSM), abscisic acid (ABA) contnets (using HPLC method (Seo and Koshiba, 2002)), and seed yield per plant (SYP). The data were statistically analyzed by using analysis of variance technique (Steel et al., 1997).

3 RESULTS AND DISCUSSION

The results from Table 1 persuaded that significant differences among all the studied traits were found. The highest heritability (h^2bs) was found for photosynthetic rate, sub-stomata CO_2 concentration, stomata conductance, water use efficiency, plant height, leaves per plant, flowers per plant, seeds per fruit, leaf area, seed yield per plant and abscisic acid contents. The genetic advantage was found higher for all studied traits except sub-stomata CO_2 concentration, flowers per plant, seeds per fruit, seed mass while moderate for abscisic acid content and leaves per plant. The higher broad sense heritability referred the dominance type of gene action and suggested that the selection for such traits may be helpful to develop petunia hybrids with much of vigor and ability to tolerate harsh environmental conditions as higher value of h^2bs was recorded for abscisic acid content. Higher concentration of abscisic acid contnets in plant body gives an extra advantage to grow in drought conditions with higher and well performance. Various researchers while working on different crop plants have described about higher heritability for these traits as reported in our study (Aaliya et al., 2016; Ali et al., 2015; Ali et al., 2013; Ali et al., 2014a; Mahmood and Haider, 2016). Genetic advance indicated the presence of additive type of gene action hence the traits with higher genetic advance suggested that the selection of lines may also be helpful to develop synthetic varieties. The similar findings for different crops have been reported by various researchers (Ali and Ahsan, 2015; Ali et al., 2014b; Ali et al., 2014c; Khorasani et al., 2011; Mahmood and Haider, 2016). The results

(Supplementary Material Table S1) indicated that the lines IAGS-P8, IAGS-P9 and IAGS-P11 were better performing than all of the other lines also seen form figure 1a (principal component analysis) that the lines IAGS-P2, IAGS-P8, IAGS-P9 and IAGS-P11 fall in quadrant I which indicates the highest and best performance for respective traits. The lines which showed the best performance may be used for the development of good quality cultivars, with large number of flowers, stress tolerant and multicolor petunia hybrids and varieties (Ali et al., 2013; Florin et al., 2012; Mahmood and Haider, 2016).

Table 1: Genetic components for morpho-physiology and yield traits of petunia

Traits	M.S	G.M	GV	GCV %	PV	PCV %	EV	ECV %	h^2bs%	GA%
Photosynthetic rate (μg CO_2 s^{-1})	123.533*	14.104	39.803	167.991	43.927	176.480	4.124	54.074	90.612	74.728
Leaf tTemperature (^oC)	137.453*	21.194	38.440	134.674	60.574	169.058	22.134	102.194	63.459	40.898
Chlorphyll a (mg g^{-1} fr. mass.)	14.245*	3.199	4.238	115.095	5.770	134.298	1.532	69.203	73.447	96.786
Chlorphyll b (mg g^{-1} fr. mass)	17.345*	1.578	3.037	138.730	11.271	267.256	8.234	228.429	26.945	100.608
Stomata conductance (mmol m^{-2} s^{-1})	1.323*	0.031	0.417	366.764	0.489	397.167	0.072	152.400	85.276	337.958
Transpiration rate (mm day^{-1})	1.025*	0.884	0.258	54.013	0.509	75.873	0.251	53.286	50.678	71.773
sub-stomata CO_2 concentration (μmol mol^{-1} CO_2)	234.534*	148.889	77.000	71.914	80.533	73.546	3.533	15.404	95.613	10.114
Water use efficiency (%)	36.345*	6.792	11.700	131.248	12.945	138.055	1.245	42.814	90.382	84.026
Leaves per plant	233.342*	86.750	77.452	94.489	78.438	95.089	0.986	10.661	98.743	17.692
Plant height (cm)	219.245*	55.818	70.571	112.441	78.104	118.290	7.533	36.736	90.355	25.107
Stem diameter (cm)	1.026*	0.511	0.308	77.594	0.411	89.647	0.103	44.896	74.919	164.889
Flowers per plant	287.345*	141.000	92.791	81.123	101.764	84.955	8.973	25.227	91.183	11.449
Leaf length (cm)	36.124*	6.324	11.333	133.868	13.458	145.880	2.125	57.967	84.210	85.731
Leaf width (cm)	4.897*	1.358	1.017	86.539	2.863	145.198	1.846	116.591	35.522	77.676
Leaf area (cm^2)	41.255*	6.353	12.934	142.685	15.387	155.628	2.453	62.138	84.058	91.087
Fresh leaf mass (g)	3.522*	0.654	0.629	98.096	2.263	186.031	1.634	158.066	27.806	112.255
Fresh stem mass (g)	214.255*	49.224	67.574	117.166	79.107	126.771	11.533	48.404	85.421	27.088
Flower mass (g)	2.148*	0.601	0.391	0.806	1.366	1.945	0.975	127.396	28.608	97.648
Seeds per fruit	996.357*	866.167	326.331	61.380	343.694	62.992	17.363	14.158	94.948	3.567
100-seed mass (mg)	64.235*	12.049	16.357	116.515	31.520	161.741	15.163	112.180	51.895	42.437
Seed area (mm)	2.087*	0.357	0.434	110.317	1.218	184.745	0.784	148.192	35.657	193.490
Seed yield per plant	1.024*	0.117	0.313	163.648	0.397	184.283	0.084	84.732	78.859	745.625
Seed mass (mg)	97.573*	50.140	24.671	70.146	48.231	98.078	23.560	68.548	51.152	12.434
Abscisic Acid contents (mg/100g fresh leaf mass)	524.156*	115.124	162.969	118.979	198.219	131.217	35.250	55.335	82.217	17.646

* = significant at 5 % probability level, mean sum of squares (M.S), grand mean (G.M), genotypic variance (GV), genotypic coefficient of variance (GCV %), phenotypic variance (PV), phenotypic coefficient of variance (PCV %), environmental variance (EV), environmental coefficient of variance (ECV %), broad sense heritability (h^2bs %), genetic advance (GA)

The correlation analysis provides best opportunity to the researchers for selecting genotypes of crop plant to improve crop plant growth and production (Ali et al., 2016; Ali et al., 2014c). The results from table 2 indicated that significant correlation was found for photosynthetic rate with chlorophyll a contents, plant height, sub-stomata CO_2 concentration, leaf width, abscisic acid and seeds per fruit. Abscisic acid contents was found to be significantly correlated with most of the studied traits including photosynthetic rate, chlorophyll a contents, chlorophyll b contents, plant height, sub-stomata CO_2 concentration, transpiration rate, water use efficiency, leaf temperature, leaf area, leaf width, fresh shoot mass, seeds per fruit, 100-seed mass, seed mass and seed yield per plant. Seed yield per plant was significantly correlated with photosynthetic rate, transpiration rate, leaf temperature, leaves per plant, fresh leaf mass, stem diameter, seed area, seed mass, seeds per fruit, flowers per plant and abscisic acid contents. The positive and significant correlation revealed that the selection of lines to develop hybrids and synthetic varieties may be helpful to improve the growth and development of petunia. The significant correlation of abscisic acid content with morphological traits, seed yield and physiological traits indicated that the selection of petunia lines on the basis of good abscisic acid production may be fruitful to improve drought tolerance in petunia (Aaliya et al., 2016; Abbas et al., 2016; Filipović et al., 2014).

The growth of petunia was adversely affected by changing the environmental optimum temperature of 25 °C with minimum circadian light intensity to be 13 Wm^{-2} (Kaczperski et al., 1991). It has been observed that temperature and light caused major effects on growth and development of petunia. Therefore, new petunia varities and hybrids should be developed which can tolerate varying environmental conditions to continue optimal plant growth and development. However, the holdup of plant growth, development, the access of CO_2 by stomata in an optimized environmental condition has shown not any extensive adverse effect on petunia plants (Blanchard and Runkle, 2009).

Stepwise regression analysis was performed to predict the trait(s) that were highly contributing towards the petunia seed yield per plant. Stepwise regression analysis provides an opportunity to select crop plant genotypes with higher contribution traits to improve crop yield and production (Aaliya et al., 2016; Abbas et al., 2016). The results from Table 3 showed that stomata conductance, chlorophyll a contents, flowers per plant, leaf area, flower fresh mass, seed area, seeds per fruit, seed mass and abscisic acid contenta contributed more to seed yield per plant but it could be biased as preceding literature has also been reported the error effects of stepwise regression (El-Badawy and Mehasen, 2011) while handling a large number of independent variables. The Intercept = 145.754, R^2 = 0.863, Adjust R^2 = 0.336 and Standard Error = 0.812 was found with expected regression equation as fellow:

$$Y = 145.754 + (7.144X_1) + (-1.254\ X_2) + (3.898\ X_3) + (-6.651X_4) + (121.14X_5) + (-4.582X_6) + (0.018X_7) + (-0.042X_8) + (1.145X_9) + (0.163X_{10}) + (-0.063X_{11}) + (2.463X_{12}) + (-0.125X_{13}) + (-17.215X_{14}) + (41.006X_{15}) + (11.267X_{16}) + (-11.175X_{17}) + (2.825X_{18}) + (8.982X_{19}) + (0.256X_{20}) + (6.902X_{21}) + (21.267X_{22}) + (25.926X_{23})$$

The use of PCA (principal component analysis) to overcome the error effect of large number of independent variables in breeding experiments and find overall attributed variation in dependent structure (Ali et al., 2015; Filipović et al., 2014; Goodarzi et al., 2015; Marjanović-Jeromela et al., 2011). It has also been reported that the eigenvalues (in PCA) showed primary significance for numerical diagnostics to evaluate variation endorsed by a large number variables on the dependent structure and their data matrix in a graphical display (Greenacre, 2010). Therefore, we have also performed principle component analysis (PCA) to inspect the traits which were contributing higher towards petunia seed yield per plant. Our data generated four PCA as shown in Table 4 with diverse variation among all of the studied traits. It was found that the PC1, PC2, PC3 and PC4 contributed variation of 35.60 %, 24.60 %, 17.90 % and 11.3 % while their cumulative proportion was 25.2 %, 43.20 %, 57.20 % and 73.10 % respectively. PC1 and PC2 contributed higher variation for respective studied traits (Fig. 1a) the eigenvalues of these four PCs was higher than 1 (Fig. 1b). The Figure 1a also showed that the petunia lines IAGS-P2, IAGS-P8, IAGS-P9 and IAGS-P11 showed better performance for most of the studied traits.

Table 2: Correlation among various morpho-physiological and yield traits of petunia

Traits	A	LT	Chl. a	Chl. b	gs	E	Ci	WUE	LPP	PH	SD	FPP	LL	LW	LA	FLM	FSM	FM	SPF	HSW	SA	SM	ABA
LT	0.2024																						
Chl. a	0.8228*	0.0852																					
Chl. b	0.0197	-0.2476	0.0282																				
gs	-0.2867	-0.2754	-0.2040	0.4668*																			
E	0.0079	0.4140*	-0.0727	0.8246*	0.4179*																		
Ci	0.4950*	-0.2424	0.4296*	0.0152	0.4287*	0.0697																	
WUE	0.1222	-0.2718	0.4619*	0.7206*	0.4996*	0.8442*	0.2587																
LPP	-0.2284	0.2949	-0.2064	-0.2864	-0.2048	0.6049*	-0.2224	0.6517*															
PH	0.4889*	0.0884	0.1840	0.5719*	0.2407	0.4948*	0.1474	-0.0464	-0.2226														
SD	-0.0772	0.2017	0.2204	-0.2140	-0.2642	0.4087*	0.2221	-0.2726	0.0482	0.1820													
FPP	0.1809	-0.4272*	0.4499*	0.4462*	0.6668*	-0.2286	-0.2616	0.4426*	-0.2204	0.2420	0.2846												
LL	-0.0172	0.1487	-0.0152	0.4146*	0.2267	0.5144*	-0.0225	0.4184*	0.2298	0.6272*	0.7846*	-0.4044*											
LW	0.4454*	-0.2097	0.2184	0.2649	0.4011*	0.2166	0.1872	0.4426*	0.4649*	0.4250*	0.2819	0.4487*	-0.0998										
LA	-0.0988	0.1709	0.0484	0.4282*	0.4186*	0.6729*	0.2268	0.4761*	-0.0258	0.4480*	0.6828*	-0.4292*	0.9210*	0.2848									
FLM	-0.2756	-0.0711	0.2048	0.4242*	0.2044	0.0602	0.2902	0.2690	0.4792*	-0.0848	-0.2441	0.4787*	0.2478	-0.2162	0.2826								
FSM	0.2586	0.0874	-0.0951	0.8227*	0.7284*	0.7286*	-0.2976	0.6617*	0.2229	0.4228*	0.4016*	0.7602*	0.1291	0.4848*	0.6144*	0.4962*							
FM	0.4268*	0.4892*	0.2216	0.5122*	-0.2800	0.8444*	0.0062	-0.0209	0.2824	-0.0691	-0.2471	-0.0049	0.4442*	0.2229	-0.4496*	-0.2210	0.4468*						
SPF	0.4417*	-0.2624	-0.2290	0.0262	0.6072*	0.2427	-0.4290*	0.0702	-0.1222	-0.0821	-0.2815	0.6472*	-0.0104	-0.2489	-0.2488	-0.2559	0.4417*	0.0109					
HSM	0.2220	0.0852	0.0289	0.2801	0.2698	0.2241	-0.2069	0.2428	0.2402	0.4799*	-0.2092	-0.4772*	0.4224*	0.4472*	0.4400*	-0.0049	0.1089	0.2227	-0.2910				
SA	0.2224	-0.0276	0.4487*	-0.4649*	-0.4144*	-0.4196*	-0.2204	-0.2422	0.0090	0.4724*	0.1642	0.2487	0.4104*	-0.2822	-0.4246*	0.4184*	0.1262	0.4474*	-0.0584	-0.4251			
SM	-0.2780	0.4446*	0.2201	0.2084	0.2294	-0.0742	-0.0224	0.2144	-0.2874	0.2422	0.4928*	-0.2649	0.4114*	-0.4291	-0.1608	0.0275	-0.0047	-0.2452	-0.2056	-0.4778*	-0.2148		
ABA	0.4874*	0.5678*	0.4291*	0.6019*	0.6201*	0.5211*	0.2949	0.5404*	0.7011*	0.0150	-0.2548	0.5052*	0.2578	0.4029*	0.4122*	-0.2109	0.4672*	-0.0241	0.4402*	0.4122*	-0.0252	0.5046*	
SYP	0.4686*	0.4744*	-0.2714	-0.2221	0.4062*	0.4179*	-0.2422	0.4978*	0.4494*	-0.0664	0.4180*	0.4444*	-0.2852	-0.2274	-0.2542	0.4222*	0.2217	0.0212	0.4284*	0.0642	0.4851*	0.4295*	0.5642*

*= Significant at 5 % probability level, A = photosynthetic rate, LT = leaf temperature, Chl. a = chlorophyll a content, Chl. b = chlorophyll b content, E = transpiration rate, gs = stomata conductance, Ci = sub-stomata CO_2 concentration, WUE = water use efficiency, LPP = leaves per plant, PH = plant height, SD = stem diameter, FPP = flowers per plant, LL = leaf length, LW = leaf width, LA = leaf area, FLM = fresh leaf mass, FSM = fresh stem mass, FM = flower mass, SPF = seeds per fruit, HSM = 100-seed mass, SA = seed area, SM = seed mass, ABA = abscisic acid content, SYP = seed yield per plant

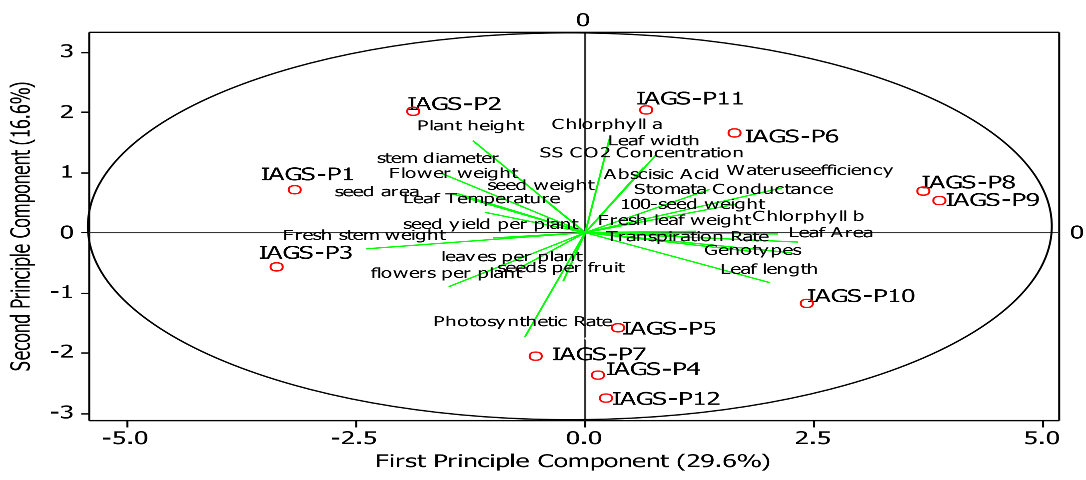
Table 3: Stepwise regression analysis for various traits of petunia for seed yield

	Traits	*Coefficients B*	*Standard Error*	*t Stat*	*Cumulative R^2*	*Partial R^2 %*
X_1	Photosynthetic rate	7.144	0.553	2.013	0.1673	16.73
X_2	Leaf temperature	-1.254	0.127	-2.320	0.2151	21.51
X_3	Chlorphyll a	3.898	2.531	1.114	0.2573	25.73
X_4	Chlorphyll b	-6.651	1.632	1.052	0.2661	26.61
X_5	Stomata conductance	121.14	45.125	2.525	-0.235	23.50
X_6	Transpiration rate	-4.582	3.153	-1.172	0.266	26.60
X_7	Sub-stomata CO_2 concentration	0.018	0.053	-1.512	-0.263	26.30
X_8	Water use efficiency	-0.042	0.351	-0.153	0.5631	56.31
X_9	Leaves per plant	1.145	0.121	1.522	0.2634	26/34
X_{10}	Plant height	0.163	0.086	-1.315	0.2534	25.34
X_{11}	Stem diameter	-0.063	0.015	0.063	0.4743	47.43
X_{12}	Flowers per plant	2.463	8.535	0.279	0.5386	53.86
X_{13}	Leaf length	0.125	0.015	2.233	-0.2157	21.57
X_{14}	Leaf width	-17.215	16.815	-1.037	0.2353	23.53
X_{15}	Leaf area	41.006	24.150	-0.137	-0.2327	23.27
X_{16}	Fresh leaf mass	11.267	8.759	0.521	0.0333	3.33
X_{17}	Fresh stem mass	-11.175	5.115	-1.255	-0.0847	8.47
X_{18}	Flower mass	2.825	0.131	0.522	0.3562	35.62
X_{19}	Seeds per fruit	8.982	10.525	-1.248	-0.2237	22.37
X_{20}	100-seed mass	0.256	0.052	-0.113	0.3644	36.44
X_{21}	Seed area	6.902	4.315	1.522	-0.1245	12.45
X_{22}	Seed mass	21.267	15.517	1.535	0.3252	32.52
X_{23}	Abscisic acid contents	25.926	11.258	0.463	0.5437	54.37

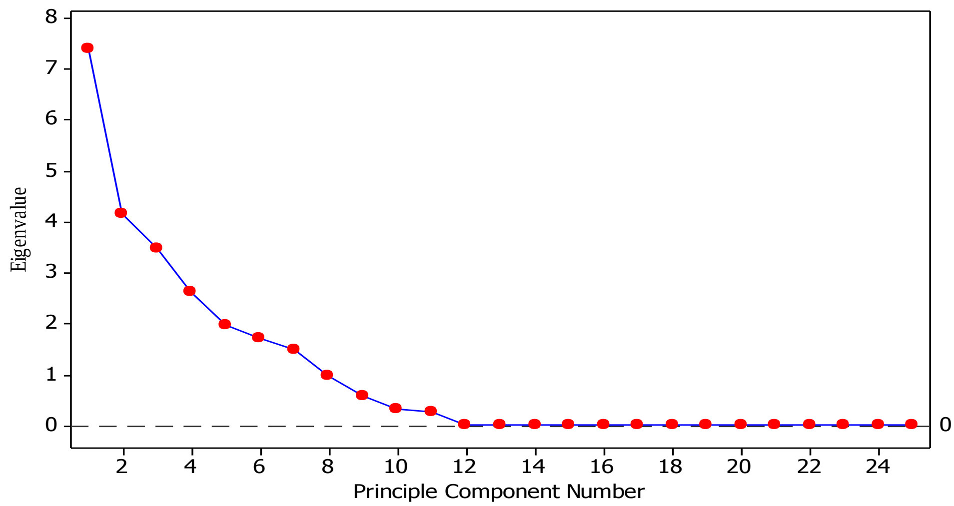
Intercept = 145.754, R^2 = 0.863, Adjust R^2 = 0.336, Standard Error = 0.812

Table 4: Principal component analysis

Eigen value	6.147	3.9536	4.-0717	3.0313
Proportion	0.356	0.246	0.179	0.113
Cumulative	0.252	0.432	0.572	0.731
Traits	**PC2**	**PC2**	**PC3**	**PC4**
Photosynthetic rate	0.689	0.425	0.026	0.055
Leaf temperature	-0.25	0.083	-0.336	-0.093
Chlorphyll a	0.035	0.376	0.07	0.036
Chlorphyll b	0.286	-0.005	-0.025	-0.227
Stomata conductance	0.222	0.228	-0.297	-0.047
Transpiration rate	0.323	-0.037	0.252	-0.262
Sub-stomata CO_2 concentration	0.087	0.163	0.029	0.002
Water use efficiency	0.291	0.183	0.223	-0.202
leaves per plant	-0.204	-0.125	-0.34	0.257
Plant height	-0.166	0.372	0.237	-0.033
Stem diameter	-0.113	0.143	-0.044	-0.224
flowers per plant	-0.413	-0.216	0.325	-0.208
Leaf length	0.114	-0.202	-0.02	0.258
Leaf width	0.214	0.323	0.076	0.248
Leaf area	0.301	-0.017	0.028	0.242
Fresh leaf mass	0.231	-0.007	-0.245	-0.042
Fresh stem mass	-0.314	-0.064	0.216	0.002
Flower mass	-0.291	0.216	0.018	0.337
Seeds per fruit	-0.031	-0.216	0.414	-0.234
200-seed mass	0.262	0.109	-0.074	0.372
Seed area	-0.291	0.212	0.012	-0.206
Seed yield per plant	-0.231	-0.012	-0.126	-0.056
Seed mass	-0.061	0.135	-0.105	-0.499
Abscisic acid contents	0.284	0.218	0.136	0.075



a: Principle components



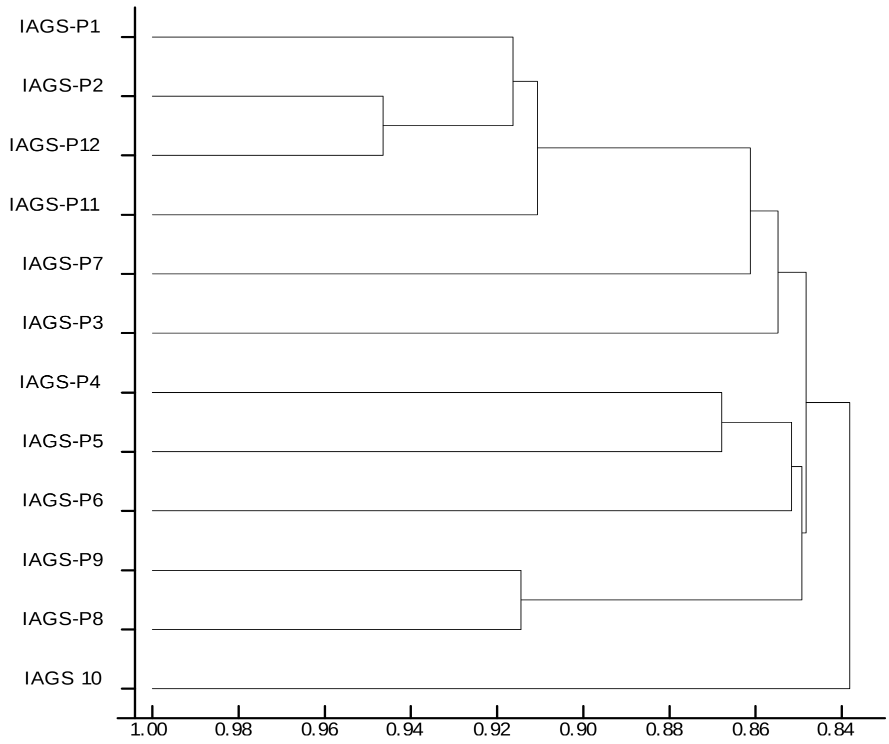
b: Scree plot

Figure 1: a. Principle component analysis of yield and its attributing traits, **b**. Scree plot and respective eigen values

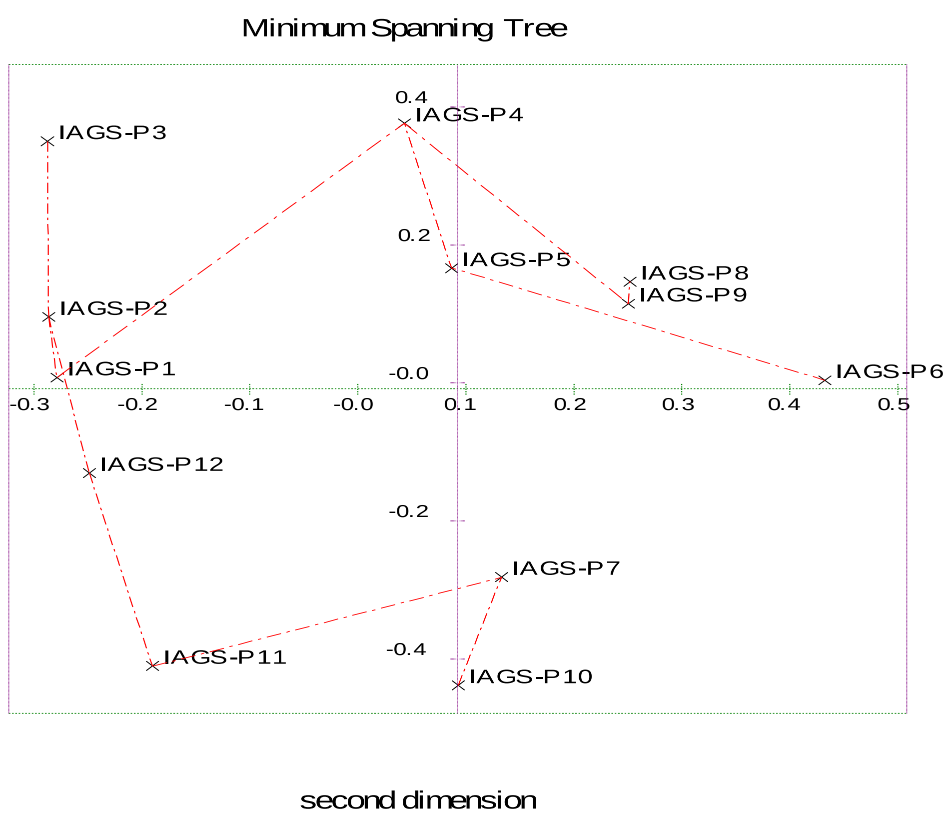
Principal factor analysis was performed by using principle component analysis values, to check that traits which were directly contributing and highly associated with petunia seed yield per plant. The factor 1 was found to be highly contributing factor trait which contributes 48.20 % in total variation were chlorophyll a, chlorophyll b, transpiration rate, stomata conductance, leaves per plant, water use efficiency, leaf area, leaf length, seed yield per plant stem diameter and abscisic acid content (Table 5). Abscisic acid content and seed yield per plant were found the most contributing traits of petunia. Various researchers have suggested that the selection of crop plant genotypes on the basis of the factor analysis (traits from factor 1) may be supportive to develop higher yield hybrids and synthetic varieties of crop plants. While the traits which fall in factor 2 (from factor loading table 5) indicated that the selection of crop plant genotypes on the basis of such traits will not be helpful as the segregation will take place in the next growing generations (Ali et al., 2016; Filipović et al., 2014; Mahmood and Haider, 2016). The better performance of petunia lines for chlorophyll a, transpiration rate, chlorophyll b, leaves per plant, stomata conductance, leaf length, water use efficiency, leaf area, stem diameter, seed yield per plant, seed mass and abscisic acid content revealed that the accumulation or assimilation of organic matter/compounds will be higher in the plant body. It has been also found the accumulation or assimilation of organic biomass in plant body is very essential for the proper enhanced growth and development of petunia plant (Hladni et al., 2011; Huang, 2007; Huang and Yeh, 2009; Mahmood and Haider, 2016). The accumulation of organic compounds generally takes place in the leaves, stem and flowering parts of a plant body. The results from our study were well supported by results which demonstrated the role of factor analysis for effective selection criteria in maize breeding program (Filipović et al., 2014). In order to understand about the genetic association among petunia lines, cluster analysis was performed (Khorasani et al., 2011; Mostafavi et al., 2011). The results from clustering showed that the petunia lines IAGS-P2 and IAGS-P12 followed by IAGS-P8 and IAGS-P9 were highly associated with each other as compared with other petunia lines (Fig. 2a) the association was verified through the development of minimum spanning tree (Fig. 2b) that showed smaller distance between petunia lines IAGS-P8 and IAGS-P9 while IAGS-P2 and IAGS-P12 were having IAGS-P1 in between them through the use of eigen values. So, from results it may be revealed that the petunia lines IAGS-P8 and IAGS-P9 were highly associated with each other and may be used as two separate male or female lines to develop petunia hybrids as also verified by mean performance and principal component analysis Figure 1a results of these lines. Also the petunia line IAGS-P11 showed better performance for almost all under studied traits, so it may also be used as male to develop good quality petunia hybrids (Mahmood and Haider, 2016). It was also suggested that in future breeding program of IAGS-P8, IAGS-P9 and IAGS-P11, these traits may be important for primary selection of synthetic petunia varieties and hybrids to increase seed yield per plant of petunia under various environmental regimes of Pakistan and other growing countries. Moreover, the hybrid seed production technology proved to be more efficient as it reduced the cost, time and increase efficacy for better selection in petunia improvement programs. Still, further studies are required which should cover different years and locations.

Table 5: Factor loadings for different traits of petunia

Factor1	Factor loadings	% Communality
Chlorophyll a	0.674	
Chlorophyll b	0.735	
Stomata conductance	0.643	
Transpiration rate	0.743	
Water use efficiency	0.879	
Leaves per plant	0.568	
Stem diameter	0.789	
Leaf length	0.678	
Leaf area	0.568	
Seed yield per plant	0.567	
Abscisic acid content	0.876	
		48.2
Factor2		
Sub-stomata CO2 concentration	-0.563	
Plant height	-0.577	
Fresh leaf mass	-0.636	
		22.1
Factor3		
Leaves per plant	0.323	
Leaf length	0.325	
Leaf area	0.327	
Fresh leaf mass	0.241	
Seed yield per plant	0.263	
		11.1
Factor4		
Photosynthetic rate	0.221	
Leaves per plant	0.135	
Plant height	0.119	
Stem diameter	0.219	
Leaf area	0.287	
Flower mass	0.153	
100-seed mass	0.206	
		8.62
Cumulative variance		90.02



a: Dendrogram



b: Minimum spanning tree

Figure 2: a. Dendrogram analysis based on hierarchal clustering. Association of petunia lines on genetic basis of all studied traits, **b**. Minimum spanning tree using eigene values for petunia lines on the basis of all studied traits

4 CONCLUSION

The present study concluded that abscisic acid contents showed positive and significant association and contribution towards seed yield of petunia genotypes. It was suggested that selection on the basis of abscisic acid content may be useful to develop good seed yield per plant and large number of flowers per plant in petunia under stressful environmental conditions.

5 CONFLICT OF INTEREST

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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