Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Annals of Biological Research, 2017, 8 (3): 24-29
(http://scholarsresearchlibrary.com/archive.html)

ISSN 0976-1233
CODEN (USA): ABRNBW

Variability, Character Associations and Path Analysis in Ashwagandha (*Withania somnifera* (L). Dunal) with Respect to Root Yield and Biochemical Aspects

G Mahendra Singh[1]*, NS Dodiya[1], Arunabh Joshi[2], Champa Lal Khatik[3]

[1]*Department of Plant Breeding and Genetics, Rajasthan College of Agriculture, MPUAT, Udaipur, India*

[2]*Department of Molecular Biology and Biotechnology, Rajasthan College of Agriculture, MPUAT, Udaipur, India*

[3]*All India Coordinated Research Project (AICRP) on Medicinal and Aromatic plants, Rajasthan College of Agriculture, MPUAT, Udaipur, India*

ABSTRACT

Background: *The experimental study was carried out to know the genetic variability, characters association, interrelationship and cause and effect of various characters in ashwagandha with respect to dry root yield and biochemical aspects.*

Methods: *Twenty genotypes (including 3 checks) were evaluated for 13 traits (10 quantitative and 3 qualitative) and analysis of variance, correlation and path analysis were performed for the mean data.*

Results: *All characters were found to be differing significantly among genotypes. Estimates of variability parameters revealed that a high genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) were found for total antioxidants content in root, dry root weight per plant, dry plant weight per plant and harvest index (%). High heritability (h_2) was found for total crude fiber content in root, total antioxidants content in root, plant height and number of secondary branches per plant. Total antioxidants content in root, dry plant weight and total crude fiber content in root were recorded with high genetic advance (GA). A high heritability coupled with high genetic advance was found for total antioxidants and total crude fiber content in root. Root diameter, plant height, dry plant weight and days to 75% maturity were shown significant positive correlation with dry root yield per plant. Path coefficient analysis revealed that root diameter, days to75% maturity and plant height had shown high positive and direct effect on dry root yield per plant.*

Conclusion: *The heritable variability and estimates of variability can be used for crop improvement. Root diameter, days to 75% maturity, plant height and dry plant weight could be used to select high dry root yielding genotypes. Biochemical data (alkaloid and antioxidants content) will be useful to select genotypes of high medicinal value.*

Keywords: Variability, Correlation, Path analysis, Heritability (h_2), Genetic advance (GA), Biochemical aspects

---

INTRODUCTION

Ashwagandha (*Withania somnifera* (L.) Dunal) is a medicinal herb, belongs to the family of Solanaceae. It is usually an annual plant but perennial type available in wild. It is believed that it was originated in northwestern part of India. It is commonly called as winter cherry, Indian ginseng etc. Major ashwagandha growing states in India are Madhya Pradesh, Rajasthan, Gujarat, Haryana, Uttar Pradesh and Maharashtra. Madhya Pradesh ranks first in ashwagandha production. The herb can be grown in low fertile soil also. Being a hardy crop, it can withstand in various climates. The roots of ashwagandha smells like horse (*ashwa*), therefore it is called as ashwagandha. Being a powerful adaptogen, it enhances the body's resilience to stress. Ashwagandha improves the body's defense against disease by

improving the cell-mediated immunity. It also possesses potent antioxidant properties that help protect against cellular damage caused by free radicals [1]. Ashwagandha is taken for treating cold and coughs, ulcers, emaciation, diabetes, conjunctivitis, epilepsy, insomnia, senile dementia, leprosy, Parkinson's disease, nervous disorders, rheumatism, arthriti s, intestinal infections, bronchitis, asthma, impotence [2]. It is being widely used in ayurvedic system, as a Rasayana (it is an herbal preparation that promotes physical and mental health) [1]. Therefore there is a lot of scope to improve the productivity of ashwagandha through genetic selection and crop improvement. Variability is pre requisite for most of the crop improvement programs. Only the portion of genetic variability will be inherited. Knowledge about variability, interrelationship between yield and its contributing characters, their association and influences of independent variables on dependent variable, will help in selection of desirable trait and in understanding the hidden potentiality of the crop. Hence the present investigation was mainly concentrated on analysis of variance, correlation and path analysis in diverse genotypes of ashwagandha with respect to dry root yield and biochemical aspects.

MATERIALS AND METHODS

The experiment was comprised of 20 promising genotypes of ashwagandha including three standard checks namely JA-20 (Jawahar Ashwagandha-20), JA-134 (Jawahar Ashwagandha-134) and RVA-100 (Raj Vijay Ashwagandha) was laid out in the fields of AICRP on Medicinal and Aromatic Plants during late kharif, 2016 at instructional farm, Rajasthan college of Agriculture, Maharana Pratap University of Agriculture and Technology, Udaipur, India situated at an elevation of 582.17 m above the mean sea level on latitude of 24°34'N and longitude of 73°42'E. The genotypes were procured from the genetic stock of available with AICRP on M and AP, Department of Plant Breeding and Genetics. Details of genotypes are given in Table 1. The experiment was laid out in randomized block design with three replications. Each genotype was sown in row of 4 m length spaced at 30 cm from row to row and 10 cm from plant to plant. The recommended package of practices was followed to raise the healthy crop. Observations were recorded from five randomly selected competitive plants of each genotype in each replication for various characters. But days to 75% flowering and days to 75% maturity which were recorded on plot basis. Characters studied were days to 75% flowering, plant height (cm), number of primary and secondary branches per plant, days to 75% maturity, root length (cm), root diameter at collar region (mm), total alkaloid content in root (%), total antioxidants content in root (µg/ml), dry plant weight (g/plant), total crude fiber content in root (%), dry root yield per plant (g/plant) and harvest index (%).

Table 1: List of genotypes used in present study

Sl. No.	Name of genotype	Source	IC number
1.	UWS 10	AICRP on M & AP, RCA, MPUAT	IC-0615336
2.	UWS 11	AICRP on M & AP, RCA, MPUAT	IC-0615337
3.	UWS20	AICRP on M & AP, RCA, MPUAT	---
4.	UWS22	AICRP on M & AP, RCA, MPUAT	IC-0615338
5.	UWS23	AICRP on M & AP, RCA, MPUAT	IC-0615339
6.	UWS37	AICRP on M & AP, RCA, MPUAT	IC-0615343
7.	UWS65	AICRP on M & AP, RCA, MPUAT	---
8.	UWS66	AICRP on M & AP, RCA, MPUAT	---
9.	UWS71	AICRP on M & AP, RCA, MPUAT	IC-0615352
10.	UWS72	AICRP on M & AP, RCA, MPUAT	---
11.	UWS75	AICRP on M & AP, RCA, MPUAT	IC-0615353
12.	UWS77	AICRP on M & AP, RCA, MPUAT	---
13.	UWS92	AICRP on M & AP, RCA, MPUAT	---
14.	UWS93	AICRP on M & AP, RCA, MPUAT	---
15.	UWS96	AICRP on M & AP, RCA, MPUAT	---
16.	UWS100	AICRP on M & AP, RCA, MPUAT	---
17.	UWS144	AICRP on M & AP, RCA, MPUAT	---
18.	JA20	RVS KVV, Mandsaur, M.P.	---
19.	JA134	RVS KVV, Mandsaur, M.P.	---
20.	RVA100	RVS KVV, Mandsaur, M.P.	---

Statistical analysis

Mean values of each thirteen characters were subjected to ANOVA as per the standard procedure given by Panse

and Sukhatme [3]. All variability parameters like GCV, PCV, h2 and GA were estimated. The correlation coefficients were calculated according to standard procedure given by Fisher [4], Al-Jibouri et al. [5] and Singh and Choudhary [6]. Path analysis was carried out according to the method given by Dewey and Lu [7].

Chemical analysis

Total alkaloid content in root was estimated according to the method given by Harborne [8] and total crude fiber content (%) was calculated according to the standard procedure given by Marnyard [9]. Total antioxidant content in root was extracted by DPPH assay [10]. 10 ml of water was added to 200 mg of fine grinded powder of sun dried roots of ashwagandha and stirred well. Solution was filtered through Whatmann filter paper. Transferred 1 ml of clear filtered supernatant into test tube. Added 1 ml of distilled water and 0.1 ml of DPPH solution (DPPH solution prepared by adding 10 mg of DPPH in100 ml of 80% ethanol) to it. Incubated for 30 min in dark. Readings were taken in mass spectroscopy at 510 nm. Prepared standard curve by taking querecetin (0.1 µg/1 ml) concentration (0.05 ml, 0.1 ml, 0.2 ml, 0.4 ml and 0.6 ml) on X axis and absorbance value at 510 nm at Y axis. The amount of antioxidants was calculated by using standard curve of querecetin (0.1 µg/1 ml) using spectrophotometer at 510 nm.

RESULTS AND DISCUSSION

The analysis of variance results revealed that the genotypes differ significantly for all the characters under the study. Mean sum of squares for all the characters is furnished in the Table 2. The mean sum of squares (mss) for genotypes revealed that there is a considerable amount of variation between the genotypes. Genotypic variation is the result of genetic differences among individuals within a population, which is a heritable variation and is the main concern of plant breeders. Hence variability parameters *viz*, phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability (broad sense), and genetic advance (as % of mean) were estimated for all characters and furnished in the Table 3.

Table 2: Analysis of variance (mean sum of squares) for various characters in ashwagandha

SL. No.	Characters	Source of variation		
		Replication	Treatments	Error
	Degrees of freedom (df)	2	19	38
1	Plant height (cm)	37.98	63.90**	8.28
2	Number of primary branches per plant	1.20	0.58*	0.21
3	Days to 75% flowering	12.64	20.13**	5.59
4	Days to 75% maturity	14.70	20.57**	9.53
5	Root length (cm)	34.52	6.56**	2.97
6	Root diameter at collar region (mm)	1.12	2.09**	0.68
7	Dry root yield per plant (g/plant)	1.39	0.60**	0.27
8	Dry plant weight (g/plant)	35.19	32.48**	9.21
9	Harvest Index (%)	46.43	39.47**	13.50
10	Number of secondary branches per plant	0.51	2.22**	0.50
11	Total crude fiber content in root (%)	0.25	55.47**	0.09
12	Total alkaloid content in root (%)	0.0056	0.010**	0.003
13	Total antioxidant content in root (µg/ml)	0.000735	0.009**	0.0001

*, ** Significant at 5% and 1% level of significance, respectively

Table 3: Estimates of genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), heritability in broad sense (h2), genetic advance (GA) as % of mean for different characters in ashwagandha

S. No.	Character	Mean	PCV	GCV	h_2 (broad sense) (%)	GA as % of mean
1	Days to 75% flowering	76	4.26	2.99	46.43	4.07
2	Plant height (cm)	39.03	13.27	11.03	69.12	18.90
3	Number of primary branches per plant	3.37	17.24	10.26	35.42	12.58
4	Number of secondary branches per plant	7.14	14.48	10.61	53.72	16.02
5	Days to 75% maturity	150	2.42	1.28	27.85	1.39

6	Root length (cm)	20.53	9.94	5.32	28.71	5.88
7	Root diameter (mm)	6.73	15.92	10.19	40.98	13.43
8	Total alkaloid content in root (%)	0.3562	20.98	13.99	44.46	19.21
9	Total antioxidants content in root (µg/ml)	0.103	53.55	52.58	96.40	106.34
10	Dry plant weight (g/plant)	11.91	34.57	23.37	45.72	32.55
11	Total crude fiber content of root (%)	26.57	16.21	16.17	99.54	33.24
12	Dry root weight per plant (g/plant)	1.52	40.62	21.88	29.00	24.27
13	Harvest index (%)	13.00	36.31	22.69	39.06	29.22

Estimates of genetic variability parameters revealed that high GCV and PCV were recorded for total antioxidants content in root, dry root weight per plant, dry plant weight and harvest index (%). High h_2 was found for total crude fiber content of root 99.54%, total antioxidants content in root 96.40%, plant height 69.12% and number of secondary branches per plant 53.72%. High GA was recorded for total antioxidants content in root 106.34%, dry plant weight 32.55% and total crude fiber content of root 33.24%. A high repeatability coupled with high genetic advance was exhibited by total antioxidants and total crude fiber content in root, which may be due to additive gene action.

In general the genotypic correlation coefficients values were found to be higher than the phenotypic correlation coefficients in most of the cases. Dry plant weight 0.6148, days to 75% maturity 0.7511, root diameter 0.6704 and plant height 0.6346 were shown significant positive correlation with dry root yield per plant. This positive correlation could be due to, as biomass (dry plant weight and plant height) increases, the demand for water, nutrients and other minerals increases, so it increases the horizontal and vertical growth of root. As days to maturity increases there would be increase in whole vegetative growth of plant. Secondary growth of root (increase in root diameter) directly adds to root yield. However negative correlation of dry root yield both at genotypic and phenotypic level was seen with days to 75% flowering, total alkaloid and total anti-oxidants content in root. Correlation among other characters revealed that plant height was significantly and positively correlated with number of primary branches per plant, number of secondary branches per plant, days to 75% maturity, root diameter, dry plant weight per plant and total crude fiber content in root. Number of primary branches per plant was found to be strongly and positively correlated with number of secondary branches per plant, days to 75% maturity, root diameter and total crude fiber content in root. While days to 75% maturity was positively correlated with root diameter, dry plant weight per plant both at genotypic and phenotypic level. Root diameter was found to be positively correlated with total antioxidants content in root, dry plant and total crude fiber content in root. Root length found to be negatively correlated with dry root yield. Kubsad et al. [11] also recorded a negative correlation between dry root yield and root length. Total antioxidants content in root were positively correlated with days to 75% flowering, root diameter, total alkaloid and total crude fiber content of root. And dry plant weight was found to be positively correlated with total crude fiber content of root both at genotypic and phenotypic level. The results of correlation coefficients furnished in the Table 4.

Table 4: Genotypic and phenotypic correlation coefficients between different characters in ashwagandha

S.no	Days to 75 % flowering	Plant height (cm)	Number of primary branches per plant	No. of seconday branches / plant	Days to 75 % maturity	Root length (cm)	Root-diameter (mm)	Total alkaloid content in root (%)	Total antioxidants content in root (µg /ml)	Dry plant weight / Plant (g)	Total crude fiber content of root (%)	r^g
1	-0.3561	-0.1258	-0.1248	-0.0290	0.1518	-0.0778	-0.1030	0.0421	-0.1383	-0.0095	-0.2377	-0.4363
2	0.0859	0.2431	0.2267	0.1944	0.1198	-0.0341	0.2285	-0.1725	-0.0025	0.1221	0.1024	0.6346
3	-0.1908	-0.5078	-0.5443	-0.6066	-0.1702	0.3020	-0.3267	0.3616	0.0190	0.0245	-0.1965	0.4168
4	-0.0501	-0.4919	-0.6853	-0.6149	-0.5215	0.1893	-0.4337	0.2981	0.0099	0.0151	-0.0745	0.5597
5	-0.2662	0.3076	0.1952	0.5296	0.6244	0.0640	0.5358	-0.0847	-0.2407	0.2933	-0.1171	0.7511
6	-0.1298	0.0835	0.3298	0.1830	-0.0609	-0.5944	-0.1452	0.2407	0.0128	0.0167	0.2391	0.1215
7	0.2397	0.7792	0.4974	0.5845	0.7111	0.2024	0.8287	-0.3880	0.3026	0.4692	0.3020	0.6704
8	0.0626	0.3763	0.3521	0.2570	0.0719	0.2146	0.2481	-0.5301	-0.0883	0.0119	-0.0809	-0.2005
9	-0.0874	0.0023	0.0079	0.0036	0.0868	0.0049	-0.0822	-0.0375	-0.2251	0.0194	-0.0530	-0.2224
10	-0.0106	-0.2001	0.0179	0.0098	-0.1872	0.0112	-0.2256	0.0089	0.0343	-0.3985	-0.0505	0.6148
11	0.2666	0.1682	0.1442	0.0484	-0.0749	-0.1607	0.1455	0.0610	0.0940	0.0506	0.3994	0.2327

r^g : Genotypic correlation coefficient

Path coefficient analysis revealed that four characters out of eleven characters showed positive and direct effect on dry root yield per plant. Those four characters were plant height 0.2431, days to 75% maturity 0.6244, root diameter 0.8287 and total crude fiber content in root 0.3994. Root diameter shown highest positive and direct effect 0.8287 on dry root yield per plant. Whereas there was a negative direct effect exhibited by days to 75% flowering, number of primary branches and secondary branches per plant, root length , total alkaloid content in root, dry plant weight and total antioxidants content in root on dry root yield per plant. Root length exhibited high negative and direct effect on dry root yield per plant. However plant height and days to 75% maturity had indirect positive effect on dry root yield per plant through no. of primary branches and secondary branches per plant and root diameter.

However root diameter had positive indirect effect on dry root yield per plant through plant height, no. of primary and secondary branches per plant, days to 75% maturity, root length, dry plant weight per plant and total crude fiber content in root. Then total crude fiber content of root had indirect positive effect on dry root yield per plant through plant height, no. of primary branches per plant and root diameter. The magnitude of residual effect was found to be 0.552. Residual effects tell about other unknown unaccounted variation present in the genotypes due to independent variables which were not included in the study. As residual effect present study is high, therefore still a considerable amount of the variation for dry root yield per plant is present in the genotypes, which is contributed by other characters, which were not included in the study. Therefore still much research should be carried out in ashwagandha to know and understand the performance and inheritance of the dry root yield per plant. Results of Path analysis were depicted in Table 5 and Figure 1.

Table 5: Direct (diagonal) and indirect effects of different characters on dry root yield per plant in ashwagandha

r^p \ r^g	Days to 75 % flowering	Plant height (cm)	Number of primary branches per plant	No. of secondary branches / plant	Days to 75 % maturity	Root length (cm)	Root-diameter (mm)	Total alkaloid content in root (%)	Total antioxidants content in root (µg /ml)	Dry plant weight / Plant (g)	Total crude fiber content of root (%)	Dry root yield / plant (g)
1		0.2387	0.1203	- 0.1048	-0.0360	0.0947	0.2903	-0.1260	0.2555	0.0341	0.4452**	-0.0847
2	0.3534		0.5105**	0.4444**	0.2904	-0.0538	0.5892 **	-0.3993	-0.0351	0.3622	0.3458	0.4291
3	0.3506	0.9328**		0.7130**	0.3591	-0.1049	0.3455	-0.2084	-0.0224	0.0481	0.2064	0.2548
4	0.0815	0.800**	0.9874**		0.2706	-0.1772	0.2117	-0.3003	-0.0192	0.0210	0.0968	0.2396
5	-0.4264	0.4927*	0.3126	0.8482**		-0.0651	0.177	-0.1204	-0.2215	0.2960	-0.1139	0.4159
6	0.2184	-0.1405	-0.5549*	-0.3079	0.1025		0.1831	-0.1320	0.0039	0.0513	-0.1912	-0.0123
7	0.2892	0.9402**	0.6002**	0.7053**	0.8581**	0.2442		-0.1372	0.2756	0.3396	0.2315	0.4363*
8	-0.1182	-0.7099**	-0.6643**	-0.4848*	-0.1357	-0.4049	-0.4681*		0.1358	-0.1191	0.1015	-0.1729
9	0.3883	-0.0104	-0.0349	-0.016	-0.3855	-0.0216	0.3651	0.1667		-0.0756	0.2313	-0.1137
10	0.0265	0.5022*	-0.045	-0.0245	0.4697*	-0.0281	0.5662**	-0.0224	-0.086		0.0991	0.6215**
11	0.6675**	0.4213	0.361	0.1211	-0.1875	-0.4023	0.3644	0.1526	0.2353	0.1268		0.1315
12	-0.4363*	0.6346**	0.4168	0.5597**	0.7511**	0.1215	0.6704**	-0.2005	-0.2224	0.6148**	0.2327	

*, ** Significant at 5% and 1% level of significance respectively
Residual effect = 0.552
r_g: Genotypic correlation coefficient
r_p: Phenotypic correlation coefficient

From both correlation and path analysis it reveals that traits like root diameter, days to 75% maturity, plant height, dry plant weight per plant and total crude fiber content in root were shown high contribution to dry root yield per plant. So these characters could be used for further selection in ashwagandha for dry root yield per plant.

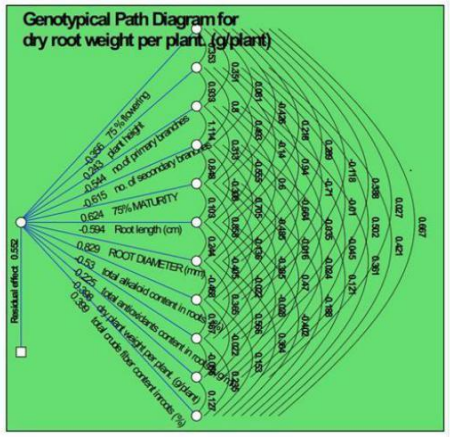

Figure 1: Genotypical path diagram depicting direct and indirect effects of various characters (independent characters) on dependent character root weight per plant (g/plant)

CONCLUSION

As per results it can be concluded that root diameter, days to 75% maturity, plant height and dry plant weight could be used to select high dry root yielding genotypes. The genetic variability present in ashwagandha can be used in crop improvement programs for both dry root yield and chemotype (alkaloid and antioxidants content in root). Characters like days to 75% flowering root diameter, total alkaloid and total crude fiber content of root would be used to select genotypes having high antioxidants content in root.

ACKNOWLEDGEMENT

We thanks and acknowledge AICRP on Medicinal and Aromatic Plants, Rajasthan College of Agriculture, MPUAT, Udaipur.

REFERENCES

[1] Singh, N., et al., *African Journal of Traditional, Complementary and Alternative Medicines,* **2011**. 8(5): p. 208-213.

[2] Umadevi, M., et al., *The Pharma Innovation.* **2012**. 1(9): p. 102-111.

[3] Panse, V.G. and Sukhatme, P.V., Statistical methods for agricultural workers, 4 ed. New Delhi. **1985**. pp. 63-66.

[4] Fisher, R.A., Statistical Methods for Research Workers. 12 ed. Edinburgh. **1954**.

[5] Al-Jibouri, H.A., Miller, P.A and Robinson, H.F., *Agron J*, **1958**. 50: p. 633-636.

[6] Singh, R.K. and Chaudhary, B.D., Biometrical methods in quantitative genetics analysis. New Delhi. **1979**.

[7] Dewey, D.R. and Lu, K.H., *Agronomy Journal*, **1959**. 51: p. 515-517.

[8] Harborne, J.B., 2 ed. New York. **1984**. p. 288.

[9] Maynard, A.J. New York. **1970**. p. 76.

[10] Blois, M. S., *Nature,* **1958**. 26: p. 1199-1200.

[11] Kubsad, V.S., et al., *Madras Agric J,* **2009.** 96(7-12): p. 314-315.