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Genetic variability and association analysis of morphological and biochemical traits in Ashwagandha [*Withania somnifera* (L.) Dunal]

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Abstract
Ashwagandha is one of the important medicinal plant commercially cultivated for long period. The investigation was performed in Randomized Complete Block design for all the twenty one traits of sixty seven ashwagandha genotypes on *Rabi* 2016-2017, 2017-2018 and 2018-2019 were studied to assess the genetic variability and associations of morphological and biochemical traits to root yield. The analysis of variance showed a wide range of variation and significant differences for all the traits under study, indicating the presence of sufficient amount of variability among the ashwagandha genotype. From the analysis, high heritability coupled with high genetic advance as percent of mean was recorded in fiber content in root (96.00%; 70.63%), carbohydrate content in root (75.00%; 39.35%), dry plant weight/plant (68.00%; 61.56%), fresh root weight/plant (63.00%; 44.03%), fresh plant weight/plant (61.00%; 69.48%). This suggested that it was due to additive gene action and selection for that particular desirable trait will be beneficial and effective in phenotypic selection. Positive and significant association of dry root weight/plant with fresh plant weight/plant and no. of berries/plant at phenotypic and genotypic level respectively. Path analysis revealed that fresh root weight/plant, dry plant weight/plant, fresh plant weight/plant, no. of berries/plant, root diameter, root branches, should be considered for direct selection in ashwagandha as these traits must bring an improvement in the dry root weight of the plant.

Keywords: Genetic variability, association analysis, path analysis, ashwagandha

Introduction
Ashwagandha is one of the most important medicinal herbs and have a recognized medicinal properties for crude drugs and extracts. Ashwagandha belongs to family of "Solanaceae" and genus of "*Withania*" and botanically known as *Withania somnifera* (L.) Dunal. It is a cross pollinated crop having the chromosome number 2n=48 (Nigam and Kandalkar 1995) [13]. It is originated from north-western and central India as well as Mediterranean region of Africa (Kumar *et al.*, 2020) [10]. In view of extremely rich biodiversity in the state, the government has declared Chhattisgarh as "Herbal state" on July 2001. It is found in Baster district of Chhattisgarh (Handbook on Medicinal and Aromatic Plant 3rd edition). It is known as "Indian Ginseng or "winter cherry" or "poison gooseberry". Ashwagandha is a "royal herb" because of its numerous rejuvenative effects on the human body possesses antioxidant, anxiolytic, adaptatgen, memory enhancing, antiparkinsdomia, antivenom, antinflamatory properties (Gupta and Rana 2007) [8]. Ashwagandha root have an ancient medicine systems due to presence of bioactive molecules known as withanolides. One of the well-known withanolides, withaferin A has an important anticancer therapeutic properties (Koduru *et al.*, 2010; Lee *et al*., 2010; yang *et al*., 2011) [9, 11, 19]. The medicinal value of ashwagandha root is due to presence of various alkaloids which varied from 0.16-0.66%. The main alkaloid found in ashwagandha are withanolide, somniferine, somniferinine, somine, withanine, pseudowithanolides, withanonine and withasomine (Covello and Ciampa (1960) [3] and Patel and Desai (2017) [14].

Medicinal properties of ashwagandha root are attributed to the chemical quality *i.e* presence of total alkaloids (Singh and Kumar 1998) [16]. Market value of the root is based on physical quality *i.e.* root texture and root morphology brittle, robust length roots have high market value (Mishra *et al.*, 1998) [12].

The important factors restricting the large scale production and development of better cultivar is because of less information available about wider genetic variability besides knowledge of variability, a detailed knowledge of the association analysis with the root yield is necessary. Using correlation analysis we can find genetic relationship among traits of *W. somnifera* which

provides the opportunity of mutual improvement of desirable traits. Path analysis help to evaluate the relative contribution of each traits, both direct and indirect to with the yield. Therefore, attempts are made to analyse genetic variability for the improvement of the medicinal plant *W. somnifera* (Bhat *et al.*, 2012) [1] and to examine the magnitude of genetic association between various traits. The present study was conducted to get an understanding on genetic architecture of root on the basis root yield, morphological, quality traits, which will facilitate genetic upgradation to develop superior cultivars benefitting both cultivators and consumers.

Materials and Methods

Sixty seven ashwagandha genotypes including three checks JA-20, RVA-100 JA-134 collected from different states of India were considered for the study. The experiment was carried out during *Rabi* 2016-17, 2017-18 and 2018-19 at research cum instructional farm, Department of Genetics and Plant Breeding, College of Agriculture, Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G.). These germplasm was planted with a spacing 30cmX10Cm row to row and Plant to plant. Randomly five plants were selected from each treatment for recording data for following traits; days to flowering, plant height (cm), no. of main branches/plant, no. of secondary branches/plant, leaf length (cm), leaf width (cm), no. of plant/plot, no. of berries/plant, seed yield (g), fresh plant weight/plant (g), dry plant weight/plant (g), main root length (cm), root diameter (cm), root branches/plant, fresh root weight/plant (g), dry root weight/plant (g), fibre content in root (%), carbohydrate content in root (%), protein content in root (%), harvest index (%) and dry matter content (%).

Statistical analysis

In statistical analysis, selected data obtained from the individual plant observations from randomized block design experiment were analyzed statistically as per the procedure given by Cochran and Cox (1957) [2]. The three years of pooled mean data of all the traits were used for the analysis of correlation and path. The genetic association and path analysis were examined using the method described by Dewey and Lu (1959) [5]. And significance of correlation was tested following the method of Fisher and Yates (1938) [7].

Result and Discussion

For all of the traits studied, the analysis of variance revealed a wide range of variation and major variations, suggesting that there is enough amount of variability among the ashwagandha genotype and that the genotype are genetically distinct. Table 1 shows the results of the study of variance. Genotypic and environmental factors combine to produce total variance in a population. The evaluation of genotypic and phenotypic coefficients of variation provides us with a true relative amount of variance among various traits. For the purpose of starting a breeding programme, information on the origin and magnitude of genetic variability is crucial. The estimates of phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability and genetic advance are presented in Table 2.

For all of the traits, the phenotypic coefficient of variation (PCV) was greater than the genotypic coefficient of variation (GCV), implying that the environment has a masking effect on genetic variability expression. The differences was due to the fact that certain characteristics were influenced by the climate. In this study, the highest phenotypic coefficient of variation recorded in no. of berries/plant (61.32%) and genotypic coefficient of variation was recorded for fresh plant weight/plant (g) (43.12%) indicating the presence of considerable genetic variability. Similarly, Sangwan *et al.* (2013) [15] observed the high PCV and GCV for the no. of berries/plant. In this study, the lowest phenotypic coefficient of variation (3.43) and genotypic coefficient of variation (1.04) was recorded for days to 50% flowering. A relatively high difference between PCV and GCV, It indicating that some traits were highly influenced by the environment, whereas low difference indicating that less influence on expression of those characteristics.

Heritability estimates' main purpose is to provide information on character transfer from parents to offspring. The high heritability was observed for fiber content in root (96.00%), carbohydrate content in root (75.00%), dry plant weight/plant (68.00%), fresh root weight/plant (63.00%), fresh plant weight/plant (61.00%). Similarly, Singh *et al.* (2017) [17] reported high heritability (h^2) for total crude fiber content in root. Heritability was due to additive gene action in the expression of that particular trait, and selection for that desirable trait would be advantageous and effective. Although heritability alone does not offer a straightforward picture of trait inheritance, genetic advancement aids in the improvement of the selected families' mean genotypic value over that of the base population. Heritability combined with genetic advance over mean can help in studying the exact nature of inheritance of particular traits.

Consideration of heritability and genetic advance together would be more useful in predicting the effect of selection on phenotypic expression of that particular traits. From the analysis, high heritability coupled with high genetic advance as percent of mean was recorded in fiber content in root (96.00%; 70.63%), carbohydrate content in root (75.00%; 39.35%), dry plant weight/plant (68.00%; 61.56%), fresh root weight/plant (63.00%; 44.03%), fresh plant weight/plant (61.00%; 69.48%). This suggested that it was due to additive gene action and selection for that particular desirable trait will be beneficial and effective in phenotypic selection. Similarly, Dubey (2010) [6] reported higher magnitude of genotypic, phenotypic coefficient of variation was recorded for dry root yield. Sangwan *et al.* (2013) [15] also reported high heritability and high genetic advance for fresh root yield/plant, biomass yield. Srivastava *et al.* (2018) [18] reported high heritability and high genetic advance for fresh root weight, plant height and 12 deoxy-withastramonolide.

In a breeding programme, association analysis is a important step. It specifies the components characters, on which selection can be based for genetics essential in root yield, and gives an idea of the relationship between the various characters. The success of the selection process is often influenced by the degree of association. Correlation coefficient between different characters at phenotypic level and genotypic level in ashwagandha is presented in Table 3 and Table 4 respectively.

At Phenotypic level

Positive and significant association of dry root weight with fresh plant weight (0.418), dry matter content (0.418), fresh root weight (0.417), dry plant weight (0.405), no. of berries per plant (0.363), plant height (0.294), root diameter (0.28), root branches (0.253), seed yield (0.247), no. of main branches (0.189), main root length (0.184), leaf length

(0.174), no. of sec. branches/plant (0.131) and plant/plot (0.127). Similarly, Das *et al.* (2011) reported positive and significant association of dry root weight with plant height. Negative and significant association of dry root weight with harvest index (-0.239) was observed. The characteristics which showed positive and significant association on dry root weight at phenotypic level was described below:

Main branches/plant: plant height (0.196).
Leaf length: plant height (0.106), main branches/plant (0.102).

Leaf width: plant height (0.143), leaf length (0.298).
No. of Berries/plant: plant height (0.419), main branches/plant (0.295), sec. branches/plant (0.162), leaf length (0.203) and leaf width (0.175).

Seed yield/plant: plant height (0.412), main branches/plant (0.226), sec. branches/plant (0.214), leaf length (0.276), leaf width (0.228), no. of berries/plant (0.599).

Fresh plant weight/plant: plant height (0.438), main branches/plant (0.257), sec. branches /plant (0.175), leaf length (0.236), leaf width (0.177), no. of berries/plant (0.712), seed yield/plant (0.578).

Dry plant weight/plant: plant height (0.461), main branches/plant (0.188), sec. branches/plant (0.172), leaf length (0.158), leaf width (0.186), no. of berries/plant (0.668), seed yield/plant (0.509), fresh plant weight/plant (0.825).

Root length: plant height (0.382), main branches/plant (0.114), leaf width (0.261), no. of berries/plant (0.310), seed yield/plant (0.240), fresh plant weight/plant (0.285), dry plant weight/plant (0.294).

Root diameter/plant: plant height (0.314), main branches/plant (0.188), leaf width (0.139), no. of berries/plant (0.576), seed yield/plant (0.337), fresh plant weight/plant (0.585), dry plant weight/plant (0.558), root length (0.264).

Root branches/plant: plant height (0.231), main branches/plant (0.219), leaf length (0.150), no. of berries/plant (0.413), seed yield/plant (0.278), fresh plant weight/plant (0.471), dry plant weight/plant (0.456), root length (0.151), root diameter (0.382).

Fresh root weight/plant: plant height (0.304), sec. branches/plant (0.227), leaf length (0.129), leaf width (0.187), no. of berries/plant (0.426), seed yield/plant (0.315), fresh plant weight/plant (0.599), dry plant weight/plant (0.651), root length (0.237), root diameter (0.418), root branches/plant (0.399).

Protein content in root: fiber content in root (0.197), carbohydrate content in root (0.335)

Harvest index: fresh root weight/plant (0.116)
Dry matter content: plant/plot (0.104), protein content in root (0.252).
The characteristic dry plant weight showed a high and strong positive significant correlation with fresh plant weight/plant (0.825) followed by fresh plant weight with no. of berries (0.712), dry plant weight with no. of berries (0.668), fresh root weight with dry plant weight (0.651) and fresh root weight with fresh plant weight (0.599) and seed yield with no. of berries (0.599).

At Genotypic level
Positive and significant association of dry root weight with no. of berries/plant (0.676), fresh plant weight (0.602), dry plant weight (0.550), seed yield (0.546), fresh root weight (0.546), root branches (0.497), leaf length (0.494), root diameter (0.478), plant/plot (0.461), plant height (0.426), no. of sec. branches (0.357), leaf width (0.340), no. of main branches (0.339), main root length (0.269), and dry matter content (0.142). Similarly, Singh *et al.* (2017) [17] reported for dry plant weight, plant height and root diameter positive and significant association with dry root weight. Negative and significant association of dry root weight with harvest index (-0.516) and days to 50% flowering (-0259). The characteristics which showed positive and significant association on dry root weight at genotypic level was described below:

Plant height: days to 50% flowering (0.249).
Secondary branches/plant: plant height (0.107), main branches per plant (0.182)

Leaf length: sec. branches per plant (0.289)
Leaf width: sec. branches per plant (0.321)
Plant/plot: main branches per plant (0.216), sec. branches per plant (0.150)

No. of Berries/plant: plant height (0.564), main branches per plant (0.448), sec. branches per plant (0.473), leaf length (0.490), leaf width (0.471), plant/plot (0.351).

Seed yield/plant: plant height (0.423), main branches/plant (0.184), sec. branches/plant (0.538), leaf length (0.561), leaf width (0.533), plant/plot (0.291), no. of berries/plant (0.819).

Fresh plant weight/plant: plant height (0.752), main branches/plant (0.256), sec. branches/plant (0.344), leaf length (0.404), leaf width (0.477), plant/plot (0.243), seed yield (0.811).

Dry plant weight/plant: plant height (0.826), main branches/plant (0.172), sec. branches/plant (0.320), leaf length (0.311), leaf width (0.477), plant/plot (0.144), no. of berries/plant (0.991), seed yield/plant (0.793), fresh plant weight/plant (0.972).

Root length: plant height (0.657), sec. branches per plant (0.232), leaf width (0.558), no. of berries/plant (0.284), seed yield/plant (0.369), fresh plant weight/plant (0.341), dry plant weight/plant (0.392).

Root diameter: plant height (0.673), main branches/plant (0.305), sec. branches/plant (0.247), leaf width (0.477), no. of berries/plant (0.874), seed yield/plant (0.727), fresh plant weight/plant (0.856), dry plant weight/plant (0.892), root length (0.354).

Root branches/plant: plant height (0.328), main branches/plant (0.129), sec. branches/plant (0.110), leaf length (0.421), leaf width (0.354), no. of berries (0.715), seed yield (0.638), fresh plant weight/plant (0.675), dry plant weight/plant (0.721), root diameter (0.581).

Fresh root weight/plant: plant height (0.734), sec. branches/plant (0.357), leaf length (0.335), leaf width (0.642), no. of berries/plant (0.875), seed yield/plant (0.703), fresh plant weight/plant (0.824), dry plant weight/plant (0.868), root length (0.40), root diameter (0.847), root branches (0.709).

Fiber content in root: days to 50% flowering (0.127), main branches/plant (0.180), seed yield (0.165), fresh plant weight/plant (0.117),

Protein content in root: main branches/plant (0.288), fiber content in root (0.207), carbohydrate content in root (0.374)

Harvest index: days to 50% flowering (0.368), carbohydrate content in root (0.123)

Dry matter content: main branches/plant (0.254), plant/plot (0.394), protein content in root (0.386).

The characteristic showed a high and strong positive significant correlation with dry plant weight with no. of berries (0.991) followed by dry plant weight with fresh plant weight (0.972), root diameter with dry plant weight (0.892), fresh root weight with no. of berries (0.875), and root diameter with no. of berries (0.874).

Thus, the correlation between dry root weight and other traits and inter-correlation between all traits were taken into account.

The correlation coefficient between different quantitative characters was used in path coefficient analysis to obtain direct and indirect effects of different characteristics on dry root weight. The dry root weight was considered as dependent variable ant all other traits were considered as independent variable. The direct and indirect effects of different characters on dry root yield at phenotypic level and genotypic level in ashwagandha is presented in Table 5 and Table 6 respectively.

At phenotypic level

The traits plant height, main branches per plant, secondary branches/plant, leaf length, plant/plot, number of berries, seed yield, fresh plant weight, dry plant weight, root length, root diameter, root branches/plant, fresh root weight and dry matter content showed positive and significant genetic association on dry root weight. The highest positive direct effect on dry root weight/plant was exerted by fresh root weight/plant (g)(0.9963), followed by dry matter content % (0.974), number of berries/plant (0.1323), carbohydrate content in root%(0.053), plant height (0.051), root length (0.0434), plant per plot (0.0264), protein content in root% (0.0195), day to 50% flowering (0.0141), fiber content in root (0.0051), number of main branches/plant (0.003), While highest negative direct effect was imposed by fresh plant weight (-0.0772), harvest index (-0.0431), root diameter (-0.0348), leaf length (-0.0284), number of sec. branches/plant (-0.0236), leaf width (-0.0163), dry plant weight (-0.0131), seed yield/plant (-0.0126), root branches/plant (-0.003). The fresh root weight/plant (0.996) was found to be direct main contributor to the dry root weight.

The characteristic fresh root weight which showed highest indirect effect on dry root weight via dry plant weight (0.649), followed by fresh plant weight (0.597), no. of berries (0.424), root diameter (0.416), and root branches per plant. Thus, dry plant weight to be the highest indirect contributor of dry root yield in ashwagandha. Residual effects (0.389) suggested that there are still some more traits to be included in further study. Similarly, Srivastava *et al.,* (2017) reported high residual effect (0.49). Thus, current analysis revealed that fresh root weight, dry plant weight, fresh plant weight, no. of berry, root diameter, root branches, should be considered for direct selection in ashwagandha as these traits must bring an improvement in the dry root weight of the plant.

At genotypic level

The traits plant height, main branches/plant, secondary branches/plant, leaf length, leaf width, plant/plot, number of berries/plant, seed yield, fresh plant weight, dry plant weight, root length, root diameter, root branches per plant, fresh root weight and dry matter content showed positive and significant genetic association on dry root weight. At the level of genotypic, the highest positive direct effect on dry root weight/plant was exerted by fresh root weight/plant (g) (2.0157), followed by number of berry/plant (1.1817), fresh plant weight (0.9275), root branches/plant (0.7514), fiber content in root (0.6911), dry matter content (0.6379), root length (0.4704), root diameter (0.4089), no. of sec. branches/plant (0.3041), leaf width (0.1737). While highest negative direct effect was imposed number of plant/plot (-0.1196), carbohydrate content in root (-0.2478), day to 50% flowering (-0.2515), plant height (-0.2918), protein content in root (-0.5785), number of main branches/plant (-0.8715), leaf length (-0.9625), seed yield/plant (-1.1591), harvest index (-1.9237) and dry plant weight (-4.0427). The fresh root weight/plant (2.015) was found to be direct main contributor to the dry root weight.

The characteristic fresh root weight which showed highest indirect effect on dry root weight via no. of berries (1.76), followed by dry plant weight (1.74), root diameter (1.70), fresh plant weight (1.66), and plant height (1.47) and harvest index showed indirect effect via no. of berries (1.49). Thus, no. of berries to be the highest indirect contributor of dry root yield in ashwagandha. Residual effects (0.229) suggested that there are still some more traits to be included in further study. Thus, current analysis revealed that fresh root weight, no. of berries, dry plant weight, root diameter, fresh plant weight, plant height and harvest index should be considered for direct selection in ashwagandha as these traits must bring an improvement in the dry root weight of the plant.

Table 1: Pooled Analysis of variance (ANOVA) for root yield and quality traits of ashwagandha

S. no.	Characteristics	Replication (*d.f.*=1)	Treatment (*d.f.*=66)	Pooled Error (*d.f.*=330)
1	Days to 50% flowering	0.3	16.764**	10.393
2	Plant height (cm)	0.001	247.351***	121.076
3	No. of main branches/plant	0.046	0.645**	0.393
4	No. of secondary branches/plant	0.022	20.661***	4.059
5	Leaf length (cm)	0.037	1.660***	0.333
6	Leaf width (cm)	0.162	0.267***	0.088
7	No. of plant/plot	0.002	17.541***	7.123
8	No. of berry/plant	427.141	28585.112***	10945.592
9	Seed yield/plant (g)	10.856	723.455***	194.522
10	Fresh plant weight/plant (g)	0.049	6621.598***	632.81
11	Dry plant weight/plant (g)	0.117	180.965***	13.444
12	Main root length (cm)	0.009	35.156***	8.838
13	Root diameter (cm)	0.086	1.232***	0.369
14	Root branches/plant	0.105	10.267***	2.967
15	Fresh root weight/plant (g)	0.003	52.971***	4.659
16	Dry root weight/plant (g)	0.075	2.136***	0.631
17	Fiber content in root (%)	0.894	190.784***	1.409
18	Carbohydrate content in root (%)	0.035	162.554***	8.7
19	Protein content in root (%)	0.243	10.178***	0.115
20	Harvest index (%)	8.218	113.646***	46.271
21	Dry matter content (%)	83.969	278.574***	55.828

& * represent significant at 1% & 0.5% respectively. *d.f.* = degree of freedom

Table 2: Genetic Parameter of variation for root yield and quality traits of ashwagandha

S. No.	Characteristics	PCV (%)	GCV (%)	h^2 (bs) (%)	GA as % of mean
1	Days to 50% flowering	3.43	1.04	09.00	0.66
2	Plant height (cm)	20.99	8.08	15.00	6.41
3	No. of main branches/plant	28.46	8.88	09.00	5.64
4	No. of secondary branches/plant	31.88	20.29	41.00	26.62
5	Leaf length (cm)	16.01	10.11	39.00	13.15
6	Leaf width (cm)	17.46	8.76	25.00	9.06
7	No. of plant/plot	27.72	12.27	19.00	11.19
8	No. of berry/plant	61.32	28.22	21.00	26.75
9	Seed yield/plant (g)	43.19	24.12	31.00	27.75
10	Fresh plant weight/plant (g)	55.11	43.12	61.00	69.48
11	Dry plant weight/plant (g)	44.27	36.37	68.00	61.56
12	Main root length (cm)	16.84	9.7	33.00	11.51
13	Root diameter (cm)	20.67	10.93	28.00	11.91
14	Root branches/plant	28.23	15.23	29.00	16.92
15	Fresh root weight/plant (g)	33.74	26.86	63.00	44.03
16	Dry root weight/plant (g)	33.97	18.12	29.00	19.91
17	Fiber content in root (%)	35.82	35.95	96.00	70.63

18	Carbohydrate content in root (%)	25.58	22.11	75.00	39.35
19	Protein content in root (%)	26.05	25.2	09.00	50.23
20	Harvest index (%)	44.49	19.66	19.00	17.9
21	Dry matter content (%)	35.45	22.4	39.00	29.16

PCV: Phenotypic coefficient of variation, **GCV**: Genotypic coefficient of variation, **h^2 (bs):** Heritability in broad sense**, GA**: Genetic advance

Table 3: Correlation coefficient between different characters at phenotypic level in ashwagandha

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	1																			
2	-0.018	1																		
3	-0.034	0.196***	1																	
4	-0.09	0.03	0.024	1																
5	-0.06	0.106*	0.102*	0.092	1															
6	0.081	0.143**	-0.001	0.028	0.298***	1														
7	-0.012	0.001	0.061	0.019	0.05	0.057	1													
8	-0.079	0.419***	0.295 ***	0.162**	0.203***	0.175***	0.019	1												
9	-0.038	0.412 ***	0.226***	0.214***	0.276***	0.228***	-0.014	0.599***	1											
10	-0.079	0.438***	0.257***	0.175***	0.236***	0.177***	0.042	0.712***	0.578***	1										
11	-0.046	0.461***	0.188***	0.172***	0.159**	0.186***	0.077	0.668***	0.509***	0.825***	1									
12	-0.004	0.382 ***	0.114*	0.012	0.078	0.261***	0.025	0.310***	0.24***	0.285***	0.294***	1								
13	-0.112*	0.314***	0.188***	0.099*	0.069	0.139 **	0.007	0.576***	0.337***	0.585***	0.558***	0.264***	1							
14	-0.068	0.231***	0.219***	0.076	0.150**	0.029	0.044	0.413***	0.278***	0.471***	0.456***	0.151**	0.382***	1						
15	-0.108 *	0.304**	0.088	0.227***	0.129**	0.187***	0.011	0.426***	0.315***	0.599***	0.651***	0.237***	0.418***	0.399***	1					
16	0.023	-0.01	0.033	0.029	-0.024	-0.057	0.018	0.039	0.097	0.089	0.061	-0.09*	0.007	-0.012	0.064	1				
17	-0.004	-0.066	-0.028	0.039	-0.214***	-0.139**	-0.176 ***	-0.061	-0.152**	-0.089	-0.095	-0.017	0.019	-0.013	0.002	-0.022	1			
18	-0.086	-0.039	0.091	-0.094	-0.113*	-0.171***	0.016	-0.090	-0.19 ***	-0.114*	-0.183***	-0.049	-0.011	-0.155**	-0.248***	0.197 ***	0.335***	1		
19	0.021	-0.229***	-0.219***	-0.046	-0.274***	-0.089	-0.040	-0.343***	-0.385***	-0.570***	-0.332***	-0.129**	-0.311 ***	-0.206***	0.116*	0.024	0.073	-0.081	1	
20	0.007	-0.077	0.071	-0.073	0.075	-0.115*	0.104 *	-0.138 **	-0.092	-0.201***	-0.268***	-0.119 *	-0.158**	-0.159**	-0.579***	-0.082	-0.039	0.252***	-0.334***	1
21	-0.087	0.294**	0.189**	0.131**	0.174**	0.062	0.127*	0.363**	0.247**	0.418**	0.405**	0.184**	0.28**	0.253**	0.417**	-0.016	0.019	0.047	-0.239**	0.418**

* & **represent significant at 5% & 1% respectively

1. DAY_FLW: Days to flowering
2. PLT_HGT: Plant height (cm)
3. BRN_PLT: No. of main branches/plant
4. SBRN_PLT: No. of sec. branches/plant
3. LF_LT: Leaf length (cm)
4. LF_WD: Leaf width (cm)
5. PLT_PLOT: No. of plant/plot
6. BERR_PLT: No. of berries/plant
7. SED_YLD: Seed yield/plant (g)
8. PLT_WTF: Fresh plant weight/plant (g)
9. PLT_WTD: Dry plant weight/plant (g)
10 ROT_LT: Main root length (cm)
11. ROT_DIA: Root diameter (cm)
12. ROT_PLT: Root branches/plant
13. ROT_WGTF: Fresh root weight/plant (g)
14. FIB%: Fiber content in root (%)
15. CARBO%: Carbohydrate content in root (%)
15. PRO%: Protein content in root (%)
16. HI%: Harvest index (%)
16. DMC%: Dry matter content (%)
16. ROT_WGTD: Dry root weight/plant (g)

Table 4: Correlation coefficient between different characters at genotypic level in ashwagandha

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	1																			
2	0.249**	1																		
3	-0.345**	-0.324**	1																	
4	-0.167**	0.107*	0.182**	1																
5	-0.615**	0.056	-0.006	0.289**	1															
6	-0.149**	0.591**	0.056	0.321**	0.549**	1														
7	-0.087	0.047	0.216**	0.150**	0.024	-0.049	1													

8	-0.427**	0.564**	0.448**	0.473**	0.490**	0.471**	0.351**	1													
9	-0.469**	0.423**	0.184**	0.538**	0.561**	0.533**	0.291**	0.819**	1												
10	-0.286**	0.752**	0.256**	0.344**	0.404**	0.477**	0.243**	1.028	0.811**	1											
11	-0.184**	0.826**	0.172**	0.320**	0.311**	0.477**	0.144**	0.991**	0.793**	0.972**	1										
12	-0.074	0.657**	-0.136**	0.232**	-0.024	0.558**	0.041	0.284**	0.369**	0.341**	0.392**	1									
13	-0.327**	0.673**	0.305**	0.247**	0.095	0.477**	-0.011	0.874**	0.727**	0.856**	0.892**	0.354**	1								
14	-0.269**	0.328**	0.129**	0.110*	0.421**	0.354**	0.053	0.715**	0.638**	0.675**	0.721**	0.095	0.581**	1							
15	-0.215**	0.734**	-0.043	0.357**	0.335**	0.642**	-0.024	0.875**	0.703**	0.824**	0.868**	0.400**	0.847**	0.709**	1						
16	0.127*	-0.053	0.180**	0.039	-0.023	-0.109*	0.028	0.098	0.165**	0.117*	0.082	-0.186**	0.014	-0.071	0.073	1					
17	0.004	-0.126*	0.077	0.094	-0.312**	-0.194**	-0.438**	-0.159**	-0.272**	-0.137**	-0.156**	-0.033	0.009	-0.138**	-0.044	-0.049	1				
18	-0.232**	-0.128*	0.288**	-0.173**	-0.222**	-0.337**	0.007	-0.208**	-0.318**	-0.144**	-0.239**	-0.094	-0.021	-0.291**	-0.318**	0.207**	0.374**	1			
19	0.368**	-0.445**	-0.496**	-0.230**	-0.433**	-0.232**	-0.336**	-0.775**	-0.605**	-0.691**	-0.626**	-0.140**	-0.536**	-0.392**	-0.226**	0.052	0.123*	-0.180**	1		
20	0.017	-0.544**	0.254**	-0.125*	0.065	-0.481**	0.394**	-0.438**	-0.347**	-0.431**	-0.563**	-0.308**	-0.560**	-0.363**	-0.736**	-0.118*	-0.001	0.386**	-0.248**	1	
21	-0.259**	0.426**	0.339**	0.357**	0.494**	0.340**	0.461**	0.676**	0.546**	0.602**	0.550**	0.269**	0.478**	0.497**	0.546**	-0.038	0.010	0.055	-0.516**	0.142**	

* & **represent significant at 5% & 1% respectively

1. **DAY_FLW:** Days to flowering
2. **PLT_HGT:** Plant height (cm)
3. **BRN_PLT**: No. of main branches/plant
4. **SBRN_PLT:** No. of sec. branches/plant
5. **LF_LT:** Leaf length (cm)
6. **LF_WD:** Leaf width (cm)
7. **PLT_PLOT**: No. of plant/plot
8. **BERR_PLT:** No. of berries/plant
9. **SED_YLD**: Seed yield/plant (g)
10. **PLT_WTF**: Fresh plant weight/plant (g)
11. **PLT_WTD**: Dry plant weight/plant (g)
12. **ROT_LT**: Main root length (cm)
13. **ROT_DIA**: Root diameter (cm)
14. **ROT_PLT:** Root branches/plant
15. **ROT_WGTF**: Fresh root weight/plant (g)
16. **FIB%:** Fiber content in root (%)
17. **CARBO%:** Carbohydrate content in root (%)
18. **PRO%:** Protein content in root (%)
19. **HI%:** Harvest index (%)
20. **DMC%:** Dry matter content (%)
21. **ROT_WGTD:** Dry root weight/plant (g)

Table 5: Path coefficient analysis showing direct and indirect effects on dry root weight at phenotypic level in ashwagandha

	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**	**19**	**20**	**21**
1	0.0141	-0.0003	-0.0005	-0.001	-0.001	0.001	-0.0002	-0.001	-0.001	-0.001	-0.001	-0.000	-0.002	-0.001	-0.002	0.0003	-0.0001	-0.001	0.0003	0.0001	-0.087
2	-0.001	0.051	0.01	0.002	0.005	0.007	0	0.021	0.021	0.022	0.024	0.020	0.016	0.012	0.016	-0.001	-0.003	-0.002	-0.012	-0.004	0.294**
3	-0.0001	0.001	0.003	0.0001	0.0003	0	0.0002	0.001	0.001	0.001	0.001	0.0003	0.001	0.001	0.0003	0.0001	-0.0001	0.0003	-0.001	0.0002	0.189**
4	0.0021	-0.001	-0.001	-0.024	-0.00	-0.001	-0.001	-0.004	-0.005	-0.004	-0.004	-0.0003	-0.002	-0.002	-0.005	-0.001	-0.001	0.002	0.001	0.002	0.131**
5	0.0017	-0.003	-0.003	-0.003	-0.029	-0.009	-0.001	-0.006	-0.008	-0.007	-0.005	-0.002	-0.002	-0.004	-0.004	0.001	0.006	0.003	0.008	-0.002	0.174**
6	-0.0013	-0.002	0	-0.001	-0.005	-0.017	--0.001	-0.003	-0.004	-0.003	-0.003	-0.004	-0.002	-0.001	-0.003	0.001	0.002	0.003	0.001	0.002	0.062
7	-0.0003	0	0.002	0.001	0.001	0.002	0.027	0.001	-0.0004	0.001	0.002	0.001	0.0002	0.001	0.0003	0.001	-0.005	0.0004	-0.001	0.003	0.127*
8	-0.011	0.056	0.039	0.021	0.027	0.023	0.003	0.132	0.079	0.094	0.088	0.041	0.076	0.055	0.056	0.005	-0.008	-0.012	-0.045	-0.018	0.363**
9	0.0005	-0.005	-0.003	-0.003	-0.004	-0.003	0.0002	-0.008	-0.013	-0.007	-0.006	-0.003	-0.004	-0.004	-0.004	-0.001	0.002	0.002	0.005	0.0012	0.247**
10	0.0061	-0.034	-0.020	-0.014	-0.018	-0.014	-0.003	-0.055	-0.045	-0.077	-0.064	-0.022	-0.045	-0.036	-0.046	-0.007	0.007	0.009	0.044	0.016	0.418**
11	0.0006	-0.006	-0.003	-0.002	-0.002	-0.003	-0.001	-0.009	-0.007	-0.011	-0.013	-0.004	-0.007	-0.006	-0.009	-0.001	0.001	0.002	0.004	0.004	0.405**
12	-0.0002	0.017	0.005	0.0005	0.0034	0.0113	0.0011	0.0135	0.0104	0.0124	0.0128	0.043	0.0114	0.0066	0.0103	-0.0043	-0.0007	-0.0021	-0.0056	-0.0051	0.184**
13	0.0039	-0.011	-0.007	-0.003	-0.002	-0.005	-0.0002	-0.020	-0.012	-0.020	-0.019	-0.009	-0.035	-0.013	-0.015	-0.0003	-0.001	0.0004	0.011	0.006	0.280**
14	0.0002	-0.001	-0.001	-0.0002	-0.001	-0.0001	-0.0001	-0.001	-0.001	-0.0014	-0.0014	-0.001	-0.001	-0.003	-0.001	0	0	0.001	0.001	0.001	0.253**
15	-0.1072	0.303	0.088	0.226	0.128	0.186	0.011	0.425	0.314	0.597	0.649	0.236	0.416	0.398	0.996	0.064	0.002	-0.247	0.116	-0.577	0.417**
16	0.0001	-0.0001	0.0002	0.0002	-0.0001	-0.0003	0.0001	0.0002	0.001	0.001	0.0003	-0.001	0	-0.0001	0.0003	0.005	-0.0001	0.001	0.0001	-0.0004	-0.016
17	-0.0002	-0.004	-0.002	0.002	-0.011	-0.007	-0.009	-0.003	-0.008	-0.005	-0.005	-0.001	0.001	-0.001	0.0001	-0.001	0.053	0.018	0.004	-0.002	0.019
18	-0.002	-0.001	0.002	-0.002	-0.002	-0.003	0.0003	-0.002	-0.004	-0.002	-0.004	-0.001	-0.0002	-0.003	-0.005	0.004	0.007	0.019	-0.002	0.005	0.047
19	-0.001	0.010	0.01	0.002	0.012	0.004	0.002	0.015	0.017	0.025	0.014	0.006	0.013	0.009	-0.005	-0.001	-0.003	0.004	-0.043	0.014	-0.239**

20	0.007	-0.075	0.069	-0.071	0.074	-0.112	0.101	-0.134	-0.090	-0.196	-0.261	-0.116	-0.154	-0.155	-0.564	-0.080	-0.039	0.246	-0.325	0.975	0.418**
21	-0.087	0.294**	0.189**	0.131**	0.174**	0.062	0.127*	0.363**	0.247**	0.418**	0.405**	0.184**	0.280**	0.253**	0.417**	-0.016	0.019	0.047	-0.239**	0.418**	1.000
Partial R^2	-0.001	0.015	0.001	-0.003	-0.005	-0.001	0.003	0.048	-0.003	-0.032	-0.00	0.008	-0.010	-0.001	0.416	-0.0001	0.001	0.001	0.010	0.408	

* & **represent significant at 5% & 1% respectively

Residual effect= 0.389

1. DAY_FLW: Days to flowering
2. PLT_HGT: Plant height (cm)
3. BRN_PLT: No. of main branches/plant
4. SBRN_PLT: No. of sec. branches/plant
5. LF_LT: Leaf length (cm)
6. LF_WD: Leaf width (cm)
7. PLT_PLOT: No. of plant/plot
8. BERR_PLT: No. of berries/plant
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10. PLT_WTF: Fresh plant weight/plant (g)
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12. ROT_LT: Main root length (cm)
13. ROT_DIA: Main root diameter (cm)
14. ROT_PLT: Root branches/plant
15. ROT_WGTF: Fresh root weight (g)
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17. FIB%: Fiber content in root (%)
18. CARBO%: Carbohydrate content in root (%)
19. PRO%: Protein content in root (%)
20. HI%: Harvest index (%)
21. DMC%: Dry matter content (%)

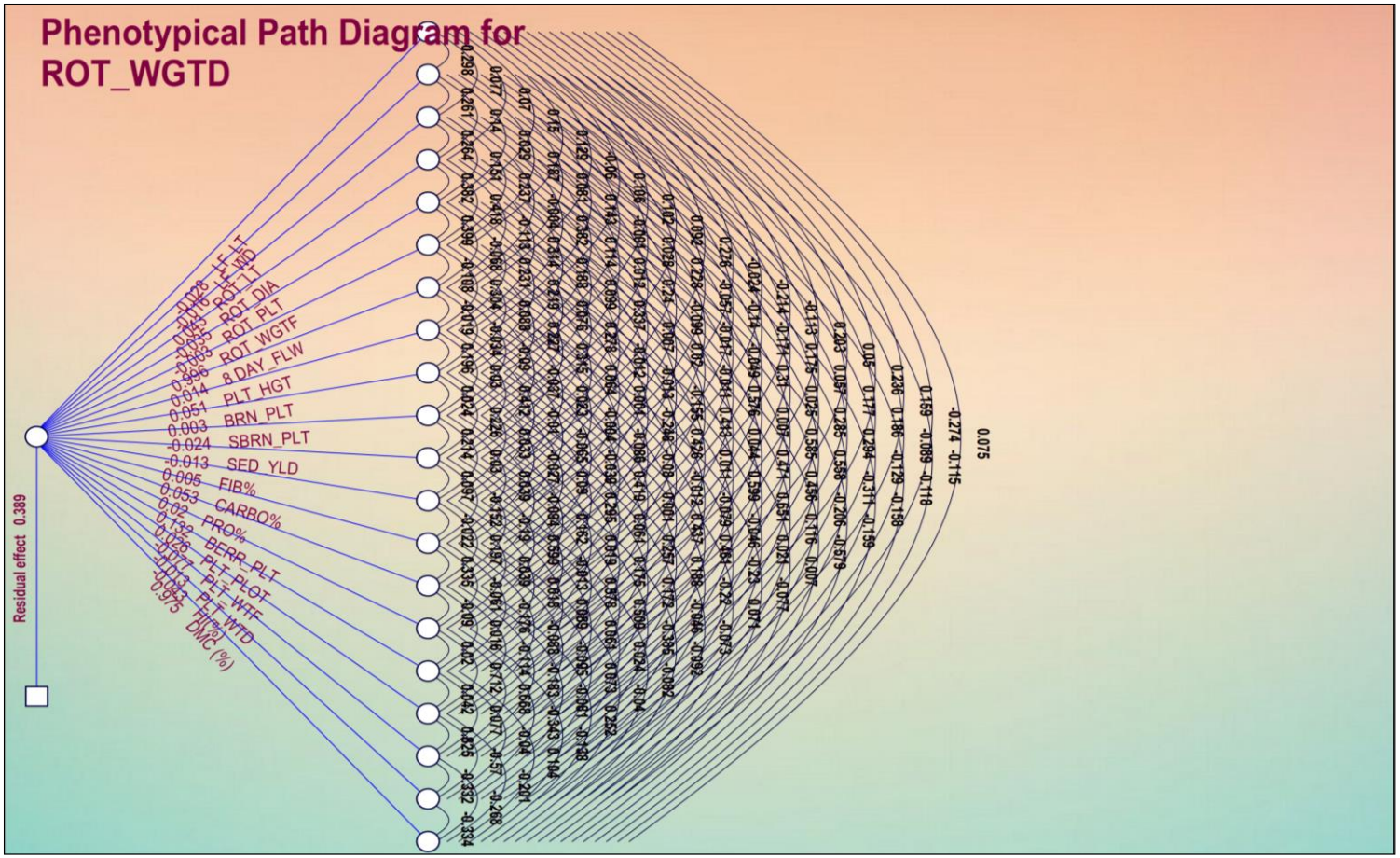

Fig 1: Phenotypic Path coefficient analysis for different traits in ashwagandha

Table 6: Path coefficient analysis showing direct and indirect effects on dry root weight at genotypic level in ashwagandha

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
1	-0.252	-0.063	0.087	0.042	0.155	0.038	0.022	0.107	0.118	0.072	0.046	0.019	0.082	0.068	0.054	-0.032	-0.001	0.058	-0.093	-0.004	-0.259**
2	-0.073	-0.292	0.094	-0.031	-0.017	-0.173	-0.014	-0.165	-0.123	-0.219	-0.241	-0.192	-0.196	-0.096	-0.214	0.016	0.037	0.037	0.13	0.159	0.426**
3	0.301	0.282	-0.872	-0.159	0.005	-0.049	-0.188	-0.390	-0.16	-0.223	-0.150	0.118	-0.266	-0.112	0.038	-0.157	-0.067	-0.251	0.433	-0.221	0.339**
4	-0.051	0.032	0.055	0.304	0.088	0.098	0.046	0.144	0.164	0.105	0.097	0.071	0.075	0.034	0.109	0.012	0.029	-0.053	-0.07	-0.038	0.357**
5	0.592	-0.054	0.006	-0.278	-0.963	-0.529	-0.023	-0.471	-0.54	-0.3891	-0.299	0.023	-0.092	-0.405	-0.323	0.022	0.300	0.214	0.417	-0.063	0.494**
6	-0.026	0.103	0.010	0.056	0.096	0.174	-0.009	0.082	0.093	0.083	0.083	0.097	0.083	0.061	0.112	-0.019	-0.033	-0.059	-0.040	-0.083	0.340**
7	0.010	-0.006	-0.026	-0.018	-0.003	0.006	-0.120	-0.042	-0.035	-0.029	-0.017	-0.005	0.001	-0.006	0.003	-0.003	0.052	-0.001	0.040	-0.047	0.461**
8	-0.505	0.667	0.529	0.559	0.579	0.557	0.415	1.182	0.967	1.214	1.171	0.335	1.0329	0.844	1.034	0.116	-0.188	-0.245	-0.916	-0.518	0.676**
9	0.544	-0.490	-0.213	-0.623	-0.65	-0.618	-0.337	-0.949	-1.159	-0.94	-0.919	-0.427	-0.843	-0.740	-0.815	-0.191	0.316	0.369	0.701	0.4017	0.546**
10	-0.266	0.697	0.237	0.319	0.375	0.443	0.225	0.953	0.752	0.928	0.902	0.317	0.7943	0.626	0.764	0.108	-0.127	-0.133	-0.641	-0.4	0.602**
11	0.745	-3.338	-0.695	-1.294	-1.26	-1.930	-0.584	-4.006	-3.206	-3.931	-4.043	-1.586	-3.605	-2.917	-3.509	-0.33	0.632	0.967	2.530	2.28	0.550**
12	-0.035	0.309	-0.064	0.109	-0.01	0.262	0.020	0.134	0.173	0.161	0.185	0.470	0.166	0.045	0.188	-0.088	-0.016	-0.044	-0.066	-0.145	0.269**
13	-0.134	0.275	0.125	0.101	0.039	0.195	-0.004	0.357	0.298	0.350	0.365	0.145	0.409	0.238	0.346	0.006	0.004	-0.009	-0.219	-0.229	0.478**
14	-0.202	0.247	0.097	0.083	0.317	0.266	0.040	0.537	0.479	0.508	0.542	0.071	0.437	0.751	0.533	-0.053	-0.103	-0.219	-0.294	-0.273	0.497**
15	-0.434	1.480	-0.087	0.720	0.676	1.294	-0.049	1.764	1.418	1.661	1.750	0.807	1.707	1.43	2.016	0.147	-0.089	-0.641	-0.455	-1.483	0.546**
16	0.088	-0.037	0.124	0.028	-0.016	-0.075	0.020	0.068	0.114	0.081	0.057	-0.129	0.0098	-0.049	0.050	0.691	-0.034	0.143	0.036	-0.082	-0.038
17	-0.001	0.0311	-0.019	-0.023	0.077	0.048	0.109	0.039	0.068	0.034	0.039	0.008	-0.002	0.034	0.011	0.012	-0.248	-0.093	-0.030	0.0003	0.010
18	0.134	0.074	-0.167	0.1	0.129	0.195	-0.004	0.120	0.184	0.083	0.138	0.054	0.012	0.168	0.184	-0.120	-0.216	-0.579	0.104	-0.223	0.055
19	-0.707	0.856	0.955	0.442	0.833	0.445	0.646	1.491	1.163	1.329	1.204	0.269	1.031	0.754	0.434	-0.099	-0.236	0.345	-1.924	0.476	-0.516**
20	0.011	-0.347	0.162	-0.080	0.042	-0.307	0.251	-0.280	-0.221	-0.275	-0.359	-0.197	-0.358	-0.232	-0.469	-0.075	-0.001	0.246	-0.158	0.638	0.142**
21	-0.259**	0.426**	0.339**	0.357**	0.494**	0.340**	0.461**	0.676**	0.546**	0.602**	0.550**	0.269**	0.478**	0.497**	0.546**	-0.0375	0.010	0.055	-0.516**	0.142**	1.00
Partial R^2	0.0652	-0.1243	-0.2956	0.1086	-0.4757	0.059	-0.0552	0.7989	-0.6327	0.5584	-2.2215	0.1265	0.1956	0.3735	1.1001	-0.0259	-0.0025	-0.0318	0.9929	0.0905	

* & **represent significant at 5% & 1% respectively

Residual effect= 0.389

1. DAY_FLW: Days to flowering
2. PLT_HGT: Plant height (cm)
3. BRN_PLT: No. of main branches/plant
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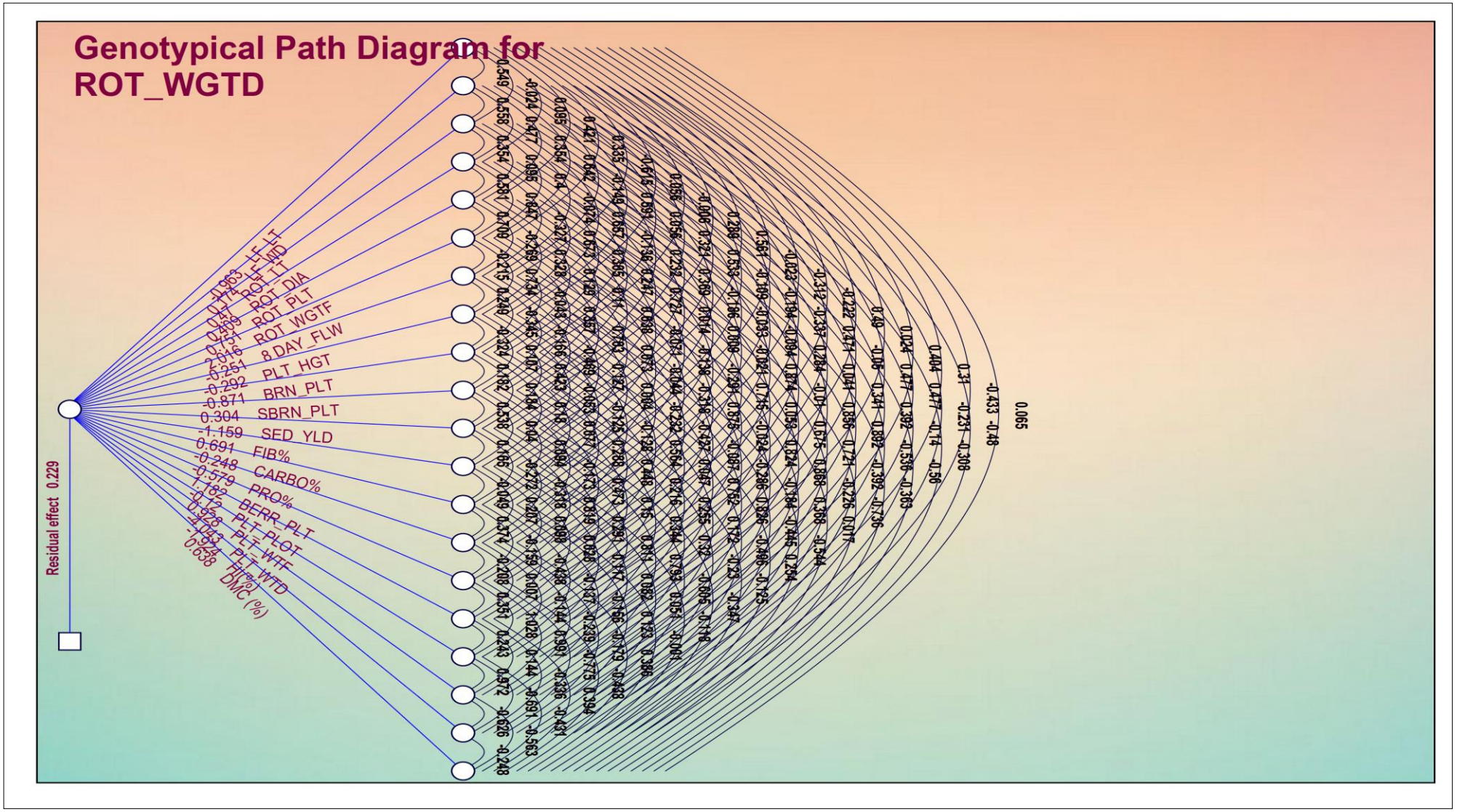

Fig 2: Genotypic Path coefficient analysis for different traits in ashwagandha

Conclusion

The aim of the study was to determine genetic variability and the relationships between morphological and biochemical traits and root yield. For all of the traits studied, the analysis of variance revealed a wide range of variation and major variations. Fiber content in root, carbohydrate content in root, dry plant weight, fresh root weight per plant, and fresh plant weight all had high heritability combined with genetic advance as percent of mean. Positive and significant association of dry root weight with fresh plant weight and no. of berries per plant at phenotypic and genotypic level respectively. According to path study fresh root weight, dry plant weight, fresh plant weight, no. of berries, root diameter, and root branches should all be considered for direct selection in ashwagandha, as these traits would increase dry root weight of the plant. The fresh root weight per plant was found to be direct main contributor to the dry root weight at both the phenotypic and genotypic level. The magnitude of residual effect was found to be high for both phenotypic (0.389) and genotypic (0.229) level. Similarly, Srivastava *et al.*, (2017) reported high residual effect (0.49). As residual effect 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.

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