

Phenotypic Diversity of *Acer ginnala* (Aceraceae) Populations in Shanxi

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Abstract: *Acer ginnala* is the important economic plant, of which gallic acid from leaf has significant anti-liver virus activity. To understand the phenotypic diversity, the systemic investigation on *A. ginnala* populations was carried out. 22 phenotypic traits for 154 individuals from 8 *A. ginnala* populations were analyzed to assess phenotypic variation and their relationship with environment. Variance analysis, nested analysis, correlation analysis and cluster analysis were used in this study. The present results showed that phenotypic traits were variation significantly different among these populations. The mean phenotypic differentiation coefficient among populations was 74.42%, which was far more than that of within populations with 25.58%. Phenotypic variation coefficient ranged from 1.33% to 27.49%, and the average value of variation coefficient of leaf, fruit, and seed were 18.25%, 13.13% and 7.09%, respectively. Principal component analysis showed the total of the first 4 principal components was 87.30% , the contribution rates was as follow: leaf rate > fruit rate > seed rate. Latitude was negative correlated to key fruit traits of *A. ginnala*, while slope and aspect were negative correlated to the leaf length and leaf width. 8 *A. ginnala* populations could be divided into three groups based on the cluster analysis. Understanding the phenotypic diversity of *A. ginnala* and their relationship with its geographical environment, it is most important to the further genetic diversity improvement and protecting.

Key words: *Acer ginnala*, phenotypic diversity, geographical environment, correlation.

1. Introduction

Acer ginnala belongs to the Aceraceae, which is a deciduous spreading shrub [1], with hermaphrodite flower [2]. *A. ginnala* has high rate of fruit under the natural state, the seed maturity level is generally low [3]. The *A. ginnala* has high economic value in light industry, of which the Gallic acid has the activity against liver disease [4]. So protection *A. ginnala* resource is necessary. The phenotypic diversity of plants is formed by the interaction of genes and environment, so Phenotypic traits such as Length/width ratio of leaf (LLW); Length/width ratio of petiole (PLW); Length/width ratio of Key fruit (KFLW); Length/width ratio of Key seed (KSLW), Leaf length to leaf petiole (LP) were calculation by EXCEL software.

phenotypic diversity provide basis for further utilizing and protecting plant [5].

The present study about *A. ginnala* was mainly on applying germplasm resources into breeding, medicine and so on. There is little information about population diversity. The objectives of this study were: (i) to characterize phenotypic diversity of *A. ginnala* in Shanxi using 22 morphological traits, (ii) to determine the relationship between phenotypic traits and environmental factors, (iii) to provide protection method of *A. ginnala*.

2. Materials and methods

2.1 Sample collection

According to the "flora of Shanxi", the sample collection and investigation about *A. ginnala* were mainly in the area where it distributed. In Shanxi province, the study was carried out 8 different areas on October 2016. For each area, 20-30 *A. ginnala* were selected to perform morphometric analysis. Considering the effect of mother trees for this study, the distance between the candidates was about 30m. All the candidate plants were healthy trees. Geographical location and ecological factors of candidate samples were listed in Table 1.

2.2 Measurement of physiological parameters

According to the method of Falkenhagen and Vikram [6]-[7], the 17 important phenotypic traits Leaf length (LL), Leaf width (LW); Leaf area (LA); Petiole length (PL); Petiole width (PW); Petiole end width (PEW); Key fruit length(KFL); Key fruit width (KFW); Fruit thickness (FT); Fruit length (FL); Fruit width (FW); Bears the mark (BM); Key fruit stalk length (KFSL); Fruit stalk length (FSL); Seed length (SL); Seed width (SW); Seed thickness (ST) were measured by ruler, vernier caliper, leaf area instrument, respectively. The other 5

2.3 Data analyze

22 phenotypic traits of *A. ginnala* were used to perform ANOVA [8]-[9]. The phenotypic differentiation coefficient analysis [10], variation coefficient analysis [11] principal component analysis [12], relationship between phenotypic traits and geographical factors were performed correlation analysis using SPSS software version 17.0 [13]. Eight *A. ginnala* populations were performed cluster analysis (UPGMA) [14] using NTSYSps-2102a software. Other statistical analyses were performed using Excel software version 5.

3 Result and analysis

3.1 Phenotypic diversity

The F value of *A. ginnala* phenotypic traits among and within populations was listed in Table 2. By F test, 22 phenotypic traits among populations were significantly different. Leaf shape index within populations such as LL, LW, LA were significant or extremely significant difference (respectively 11.843, 6.948, 9.045). The fruit morphological indexes KFL, FL and BM were extremely remarkable difference (respectively 7.319, 4.279, 7.139). There was no significant difference in seeds morphological index. This result showed the morphological characteristics of seed were relative stability.

Coefficient of variation is used to show the discretion characteristics. General speaking, the variation coefficient was higher; the degree of discrete traits was bigger. The average coefficient of variation of leaves, fruits and seeds had some differences (table 3), the leaf (18.25%) > fruit (13.130%) > seed (7.09%), this explained that leaf had high variation level, and seed traits had stability variation level. The results of this study were similar with the conclusion of phenotypic diversity of *Acer mono* maxim population [16].

The first 4 principal components of the cumulative contribution rate were 87.3%, they reflected most basically information of the primitive variables (Table 4). The first principal component contribution rate was 39.83%, LL, LA, and LW played the decisive role (respectively -0.902, 0.891, -0.873). The second principal component contribution rate was 22.52%, which played the decisive role in PW (0.899), PEW (0.815). The third principal component contribution rate was 16.50%, KFL and KFW played important roles (respectively 0.875, 0.783). The most important variables contributing to the fourth principal components were SL (0.841), SW (0.575), and total account for 8.45%. The contribution source of Shanxi *A. ginnala* phenotypic diversity: leaf contribution rate > fruit contribution rate > seed contribution rate, this result was consistent with the variation coefficient analysis, and also similar with the result of phenotypic diversity analysis of *Acer mono* [16].

3.2 Phenotypic differentiation and genetic structure

The results of variance analysis were listed in Table 5. The present results confirmed that phenotypic differentiation coefficient was diversity among and within populations. For total *A. ginnala*, the phenotypic differentiation was 74.42%. The variance component among populations was 50.70%, and the variance component within populations was 16.45%. It

revealed phenotypic variation among populations was higher than it within populations. Phenotypic variation of the Shanxi *A. ginnala* was the main from among population.

Eight *A. ginnala* populations in Shanxi were mainly divided into 3 groups with euclidean distance of 0.09 (Fig. 1). Four populations (JMLC, PQG BDG and HJG) were divided into group I. Three populations (HHG, QLY and XTS) with similar phenotypic characteristics were divided into group II. YDS population was divided into group III. The clustering results and population actual geographical distribution level were not completely consistent; this explained the discontinuity phenotypic variation of *A. ginnala*.

3.3 Correlation analysis

The average value of 22 phenotypic traits of 8 *A. ginnala* populations and environmental factors were preformed partial correlation and significant difference test. The results were given in table 6. Latitude was negative correlated to KFLW, KFL and KFW, the correlation coefficient was -0.730, -0.813, 0.784, respectively. Slope and aspect were negative correlated to the LL, LW. However, slope and aspect were positive correlated to FL and SW, the correlation coefficient was 0.799, 0.853, 0.869, 0.776, respectively. A high correlation was also observed between the slope and SW ($r=0.869$), the slope and FL were positively correlated ($r=0.853$). But correlation matrix also revealed a very strong incorrelation between phenotypic traits of *A. ginnala* and the longitude, altitude. The results indicated that fruit trait variation of *A. ginnala* was relevant to the increasing latitude. In the Shanxi area from the south to the north, fruit shape from the long oval gradually to narrowly short shape. With slope and aspect gradually increasing, leaf variation from rectangular to square, and the key fruit shape was also gradually to long elliptic variation.

4 Discussion

4.1 Phenotypic variation

Phenotypic variation was found in 22 phenotypic traits of 8 *A. ginnala* populations. In this study, variation index of vegetative organs was higher than variation index of reproductive organs, the similar results were found in plant *Acer grosser* and *Amygdalus ledebouriana* [17]-[18]. Also principal component analysis was in line with this study. Generally speaking, vegetative characters were easily affected by environmental change, variation of reproductive organs changed little due to they had complex developmental and differentiation. Different environment was the main reason for the variation of vegetative organs [12].

TABLE I
GEOLOGICAL ECOLOGY FACTORS OF *A. GINNALA* POPULATIONS

Popula tion	Location	Latitude(E)	Longitude(N)	Altitude(m)	Slop e	Aspect
BDG	Ba daogou area	41.1333	115.4667	1580	33°	0.22°
HJG	Haojiagou area	38.5333	111.4332	1450	32°	0.34°
HHG	Hou huaigou area	36.8234	111.7564	1200	39°	0.56°
JMLC	Jiemiao forestry	36.8453	111.7552	1450	44°	0.95°
PQG	Pang quangou	37.7921	111.5118	1800	30°	0.27°
QLY	Qiliyu area	36.6167	111.9833	1560	42°	0.21°
XTS	Xing tangsi area	36.4212	110.8334	1530	53	0.85
YDS	Yunding	37.8789	111.6204	1000	46	0.82

TABLE II
THE ANOVE ANALYSIS OF PHENOTYPIC TRAITS AMONG /WITHIN POPULATIONS OF *A. GINNALA*

Phenotypic trait	Among populations <i>F</i> value	Within populations <i>F</i> value
LL	12.973**	11.843**
LW	35.996**	6.948**
LA	47.819**	9.045**
LLW	44.807**	0.205
PL	21.479**	2.148
PW	37.88**	2.013
PLW	16.520**	1.078
PFW	3.753**	3.548*
KFL	142.749**	7.319**
KFW	107.616**	2.332
KFLW	44.169**	0.608
FT	8.126**	1.176
FL	83.096**	4.279**
FW	15.042**	0.55
BM	846.528**	7.139**
KFSL	84.123**	3.705*
FSL	52.631**	1.414
SL	33.767**	1.035
SW	11.555**	0.538
ST	21.384**	1.156
KSLW	11.802**	1.279
LP	13.060**	0.239

*mean significant difference at 0.05 level; ** mean significant difference at 0.01 level

TABLE III
VARIATION COEFFICIENT BASED ON PHENOTYPIC TRAITS OF A.GINNALA POPULATIONS

Phenotypic trait	Variation coefficient	Phenotypic trait	Variation coefficient	Phenotypic trait	Variation coefficient	Populations	Variation coefficient
LL	12.135	KFL	27.493	FSL	26.168	BDG	20.982
LW	15.582	KFW	5.649	SL	10.67	HJG	15.675
LA	22.473	KFLW	5.709	SW	1.326	HHG	20.191
LLW	12.9	FT	10.924	ST	1.639	JMLC	23.833
PL	23.21	FL	5.5	KSLW	14.706	PQG	10.298
PW	14.068	FW	8.775	LP	20.885	QLY	26.53
PLW	24.375	BM	2.25			XTS	20.255
PEW	18.613	KFSL	25.699			YDS	29.314

TABLE IV
ANALYSIS ON FEATURE VALUE OF PRINCIPAL COMPONENT, CONTRIBUTION RATE AND ACCUMULATION CONTRIBUTION

Phenotypic	Principal component			
	1	2	3	4
LL	-0.902	0.567	0.003	-0.027
LW	-0.873	0.334	0.423	0.194
LA	0.891	0.784	0.185	0.102
LLW	0.551	0.003	-0.663	-0.329
PL	-0.731	0.230	0.179	0.015
PW	-0.136	0.899	0.056	0.276
PLW	-0.813	-0.465	0.107	-0.147
PEW	-0.456	0.815	-0.043	-0.125
KFL	0.652	0.551	0.875	-0.139
KFW	0.831	0.535	0.783	-0.048
KFLW	-0.860	-0.397	0.187	-0.066
FT	0.129	-0.107	0.118	-0.273
FL	0.735	0.067	0.097	-0.022
FW	0.285	-0.324	0.459	-0.378
BM	0.805	0.344	0.437	-0.139
KFSL	-0.582	0.293	-0.016	0.002
FSL	0.194	0.561	-0.253	0.502
SL	0.229	-0.057	0.418	0.841
SW	0.820	-0.255	0.450	0.575
ST	-0.065	-0.790	0.536	0.173
KSLW	-0.397	0.262	0.463	0.042
LP	0.718	-0.038	-0.263	-0.067
Eigen value	8.762	4.955	3.630	1.860
Contribution rate	39.825	22.522	16.499	8.454
Cumulative Contribution	39.825	62.347	78.846	87.300

TABLE V
VARIANCE COMPONENTS AND DIFFERENTS AND DIFFERENTIATION COEFFICIENTS (V_{ST}) OF PHENOTYPIC TRAITS AMONG AND WITHIN POPULATIONS OF *A. GINNALA*

Phenotypic	Variance components			Percentage of variance portion		V_{ST}
	Within populations	Among populations	Random errors	Within populations	Among populations	
LL	54.307	21.247	8.552	64.570	25.262	71.878
LW	16.083	13.303	9.127	41.759	34.542	54.729
LA	662.260	536.864	282.922	44.686	36.225	55.229
LLW	1.011	0.2000	0.461	60.467	11.962	83.485
PL	12.585	5.393	11.969	42.023	18.009	70.001
PW	0.388	0.088	0.209	56.596	12.889	81.451
PLW	17.598	4.921	21.761	39.742	11.113	78.148
PEW	0.291	1.181	1.586	9.528	38.605	19.794
KFL	0.713	0.157	0.102	73.375	16.124	81.984
KFW	0.473	0.044	0.090	77.960	7.241	91.501
KFLW	0.451	0.027	0.209	65.726	3.875	94.433
FT	0.011	0.007	0.028	24.190	15.001	61.723
FL	0.152	0.033	0.037	68.188	15.049	81.921
FW	0.028	0.004	0.038	39.764	6.233	86.449
BM	3.355	1.210	0.081	72.213	26.044	73.494
KFSL	0.231	0.044	0.056	69.853	13.183	84.123
FSL	1.472	0.170	0.571	66.520	7.661	89.672
SL	0.081	0.011	0.049	57.593	7.564	88.391
SW	0.024	0.005	0.043	33.701	6.719	83.377
ST	0.029	0.007	0.028	45.722	10.598	81.183
KSLW	0.402	0.187	0.696	31.297	14.532	68.290
LP	1.451	1.140	2.270	29.850	23.452	56.002
Mean				50.697	16.449	74.421

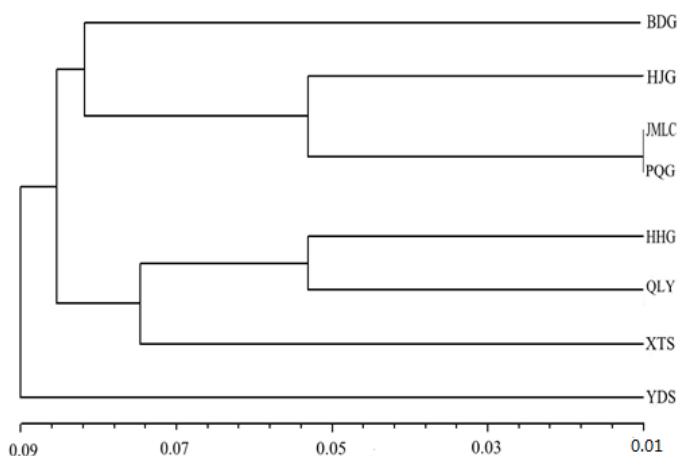


Fig.1 UPGMA-derived dendrogram based on Euclidean distances showing of the 22 phenotype traits of *A. ginnala*

TABLE VI
CORRELATION COEFFICIENT BETWEEN PHENOTYPIC CHARACTERS AND GEOGRAPHICAL FACTORS OF
A.GINNALA

Phenotypic	Longitude	Latitude	Altitude	Slope	Aspect
LL	0.630	0.149	0.674	-0.787*	-0.710*
LW	0.377	0.143	0.411	-0.631	-0.748*
LA	0.297	0.119	0.538	-0.699	-0.474
LLW	-0.051	-0.182	-0.107	0.354	0.645
PL	0.286	0.585	0.460	-0.682	-0.488
PW	0.095	-0.100	0.309	-0.441	-0.110
PLW	0.223	0.689	0.216	-0.380	-0.430
PEW	0.413	0.001	0.563	-0.572	-0.190
KFL	-0.348	-0.730*	-0.041	0.471	0.388
KFW	-0.293	-0.813*	-0.188	0.489	0.544
KFLW	0.185	0.784*	0.336	-0.441	-0.607
FT	-0.344	0.104	-0.657	0.037	0.291
FL	-0.381	-0.765*	-0.532	0.799*	0.853**
FW	-0.516	-0.222	-0.706	0.169	-0.070
BM	-0.491	-0.792*	-0.270	0.652	0.550
KFSL	-0.595	-0.617	-0.480	0.526	0.719*
FSL	-0.479	0.053	-0.263	-0.414	0.020
SL	-0.369	-0.476	-0.201	0.592	0.344
SW	-0.626	-0.634	-0.739*	0.869**	0.776*
ST	-0.474	0.297	-0.497	0.355	0.062
KSLW	0.042	-0.094	0.415	-0.051	-0.274
LP	-0.003	-0.635	-0.209	0.405	0.175

Seed of *A. ginnala* is the transfer of genetic material, higher stability, less affected by the environment, so seed has a low level variation. *A. ginnala* habitat had diversity types. It not only growth in the valley wetlands, forest edges, but also growth in bush fallow and adret slope. In order to adapt to diverse habitat, *A. ginnala* had high phenotypic diversity.

4.2 Differentiation coefficient

Differentiation degree of *A. ginnala* among population was high due to the following 2 reasons: Firstly, *A. ginnala* had bisexual flowers which increase the probability of selfing [19]. Secondly, although in the open areas *A. ginnala* Samara can perform the long-distance transmission with the wind, but its distribution area was often covered with evergreen broad-leaved forest, dense forest edge may be a barrier to the dispersal of seeds, the seeds flow of long distance be affected.

4.3 Correlation analysis

In order to know the relationship between some plants traits and geographical factors, correlation analysis for phenotypic variation was preformed. Latitude reflected the temperature changes. With latitude increasing, the temperature gradually reduced. The slope and aspect reflected differences light conditions, longitude mainly reflected water change. According to this study, the phenotypic variation of *A. ginnala* was largely influenced by latitude, slope and aspect, and was less influenced by longitude. All of these results correspond to

the *A. ginnala* growth environment. Due to the *A. ginnala* belongs to positive species, like wet soil, but resistance to dry, and *A. ginnala* often growth on sunny hillside or semi sunny slope [20].

Conservation considerations

A. ginnala is one of the main economic plants in China. Natural reserves of *A. ginnala* should be established, especially for the low phenotypic diversity populations. In addition, seed flow among populations could be improved by human activity, such asexensive seed collection, distribution of seeds into different populations, and transplanting individuals from one habitat to another.

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