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Morphological Variation in Leaf Traits of *Populus euphratica* Oliv. Natural Populations

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ABSTRACT

Populus euphratica oliv. is one of the most important species in the arid and semi-arid regions of Iran. Variations in its leaf morphology have been studied in the eleven growing sites with different ecological conditions. Thirteen biometric characteristics of leaf were measured in all natural stands. In this study, leaf length, maximum leaf width, leaf area, distance between middle of maximum leaf width and leaf blade, distance between maximum leaf width and midrib, and the ratios of petiole length to leaf length provided good discriminating criteria for classifying various populations. Correlation between morphological traits and geo-climatic factors demonstrated an existing cline controlling the distribution of euphrat poplar populations. Three distinct groups of populations were recognized in different geographical and ecological conditions. Significant differences in morphological features have been observed in populations from eastern and western parts of Iran.

Key Words: *Populus euphratica*; Poplar; Leaf morphological traits; Natural population; Iran

INTRODUCTION

The ability of intra- and inter-specific hybridization within the *Populus* genus has caused the creation of a high number of sub-species and transient forms. Numerous studies have demonstrated that leaf variation is of adaptive significance for growth and competitive survival in a wide range of plants (Raschke, 1960; Parkhurst & Loucks, 1972; Givnish, 1979; Hinckley *et al*., 1989; Gurevitch, 1992). Callaham (1964) worked on provenance with the aim to define the genetic and environmental components of phenotypic variability between trees from different geographic origins. Several studies suggest that the extent of the geographic range correlated closely with the within population genetic variation in tropical tree species (Hamrick *et al*., 1979 & 1981; Hamrick & Godt, 1989). *Populus euphratica* oliv. is the oldest tree species in genus populus and has a wide distribution range naturally from west and central Asia to north Africa.

According to the leaf fossil discovered in China, it is estimated that *P. euphratica* has existed for about 3 - 6 million years (Shiji *et al*., 1996). The best known feature of this poplar in its areas of occurrence is its strong tolerance to high temperatures and salinity (FAO, 1979). It also plays a very important role in maintaining ecosystem function in arid and semi-arid regions, because of its tolerance to severe drought and high salinity and alkalinity in soil (Chen *et al.,* 2004). This species distributed naturally in the vast regions of Iran. It is extended from sub-tropical zone such as, Khuzistan to cold zone such as, Azarbaidjan and Zandjan provinces (Sabeti, 1976). The largest growing areas are in Khuzestan province, where they occur chiefly along large river banks and flood plains of Karoon, Kharkheh and Dez, and are economically important as a source of wood production, fodder and wildlife (Calagari, 1998). Geographical and climatical differences prevailing in *P. euphratica* growing sites affect on leaf morphology (e.g. size & shape of leaves) among the populations.

The main objective of this study is to determine the efficiency of *in situ* conservation, as well as the possibility of selecting individuals with superior performance in the generative progeny. By cloning superior individuals, positive characters of parental species can be preserved. Furthermore, this study aims to provide basic knowledge regarding the intra-specific variation that may be useful for tree improvement.

MATERIALS AND METHODS

A total of 55 even-aged and mature trees of *P. euphratica* were sampled from 11 places in Iran (Table I & Fig. 1). The method and pattern of sampling were similar for all places. Thirty fully expanded leaves were randomly selected from five trees (six leaves for each tree) and their morphological characteristics were measured. The leaf area (LA) was measured with a ΔT-Area meter. The thickness of leaf (TL) was measured with a micrometer. The leaf length (LL), maximum of leaf width (MLW), petiole length (PL), distance between the middle of maximum leaf width and leaf blade (DLL), distance between leaf maximum width and midrib (DLM) and maximum depth of leaf serrate (MDL) were measured with a vernier. The angle between the midrib and the second lower lateral vein (AMV) was measured with a protractor. The leaf dry matter (LDM) was determined by drying leaves in oven at 80ºC for 48 h. All statistical analyses were done using SAS computer software,

Table I. Environmental characteristics of the sampling sites

Site Code	Provence	Region	River	Latitude Lat	Longitude Long	Altitude (m) Alt	Mean annual Rainfall (mm) MAR	Mean annual Temperature (°C) MAT
S1	Khorasan	Sarakhs	Tajan	36° 15' N	61° 10' E	260	203.3	17.6
S2	Golestan	Dashly-Bouron	Atrak	37° 16' N	54° 56' E	50	201.9	17.1
S3	Tehran	Khojir	Djaj-roud	35° 39' N	51° 45' E	1320	231.9	17.6
S4	Gilan	Manjil	Shah-roud	36° 48' N	49° 12' E	350	196.4	17.3
S5	Zanjan	Mahneshan	Ghezel-Ozen	36° 46' N	47° 43' E	1820	207.0	14.6
S6	Azarbaijan	Gherekhlar	Shor-Chay	38° 26' N	45° 35' E	1070	342.2	12.0
S7	Azarbaijan	Jolfa	Aras	38° 50' N	45° 47' E	710	179.8	14.8
S8	Lorestan	Malavi	Khoram-Abad	32° 15' N	47° 55' E	850	523.1	16.3
S9	Khuzistan	Dezful	Dez	32° 15' N	48° 20' E	140	444.3	24.0
S10	Khuzistan	Gotvand	Karoon	32° 08' N	48° 52' E	80	295.9	24.8
S11	Khuzistan	Hamidiyeh	Karkheh	31° 30' N	48° 25' E	55	194.5	24.2

version 6.03 (SAS, 1989). The obtained data were analyzed based on estimates of simple statistical parameters, correlation and principal component analysis (PCA). Regression analyses were conducted to investigate relationships between morphological traits and environmental factors.

RESULTS

Leaf areas (LA) vary from 12.5 cm² (in site S6) to 36.8 cm² (in site S10) (see Table III). The least variation coefficients (CV) of LA are seen in sites S3 (14.5%) and S6 (15%). The thicknesses of leaves (TL) range from 0.28 mm (in site S9) to 0.38 mm (in site S6). The leaf lengths (LL) range from 4.28 cm (in site S6) to 8.06 cm (in site S8). The variation coefficient of LL varies in value from 16.8% (in site S3) to 29.8% (in site S10). Maximum leaf widths (MLW) range from 4.33 cm (in site S6) to 6.36 cm (in site S7). The petiole lengths vary from 2.33 cm (in site S5) to 3.75 (in site S3). The high LL/MLW ratios are observed in sites S3 (1.25) and S8 (1.38), while the low ratios are seen in sites S6 (0.93) and S1 (1). The PL/LL ratios vary from 0.36 (in site S5) to 0.67 (in site S1). The angle between the midrib and the second lower lateral veins (AMV) range from 27.55 (in site S2) to 36.90 (in site S9). The largest and the smallest number of main serrate on the leaves (NMS) are seen in site S4 (11.67) and S8 (4.75), respectively. The maximum depths of leaf serrate ranges from 1.8 mm (in sites S9 & S11) to 8.9 mm (in site S5).

There is a good correlation among some of the leaf morphological traits (e.g. LL-MLW, LL- LL/MLW, LL-PL/LL & LL-DLL). Correlation coefficients among leaf morphological traits indicate that the LL correlates very well ($P < 0.01$) with MLW, LL/MLW, PL/LL and DLL (see Table IV). LA positively correlates ($P < 0.01$) with LL, MLW and DLM, and negatively with TL. Results of regression analyses and relationship of important traits are shown in (Fig. 2a, 2b & 3). Leaf morphology traits were regressed against the geographic and climatic variables. Significant cline population variation detected geographic (latitude) and climatic (mean annual temperature) gradients for some of traits ($P < 0.05$). The parameters MDL and NMS correlate with geographic latitude (LAT) (see Table III) and the plots of MDL with LAT and NMS with LAT display positive slopes (Fig. 2a & 2b). The parameters MDL negatively correlates with mean annual temperature (MAT) (see Table IV) and the MDL with MAT plot exhibits a negative slope (Fig. 3). The parameters LDM and MDL also negatively correlate with MAT (see Table IV). Almost all the parameters of the morphological traits show no

Table II. List of measured morphological characters of leaf of *P. euphratica*

LL (cm)	Leaf length
MLW (cm)	Maximum of leaf width
LL:MLW	
LA (cm²)	Leaf area
TL (mm)	Thickness of leaf
PL (cm)	Petiole length
PL:LL	
NMS (no.)	Number of main serrate
DLL (cm)	Distance between leaf widest width middle and leaf blade
MDL (mm)	Maximum depth of leaf serrate
DLM (cm)	Distance between leaf widest width and midrib
AMV (°)	Angle between the midrib and the 2 nd lower lateral vein
LDM (%)	Leaf dry matter

Fig. 1. Geographical locations of 11 sampling sites of *P. euphratica* on the index map of Iran

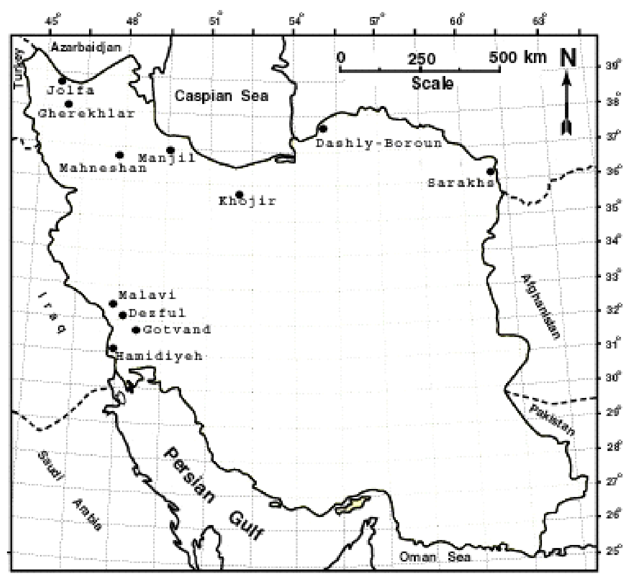


Table III. Statistical parameters, means, standard deviation (SD) and coefficient of variation (CV) of the leaf traits of *P. euphratica* for 11 sampling sites (S1 to S11)

Traits	Stat Param	S1	S2	S3	S4	S5	S6	S7	S8	S9	S10	S11
	Mean	20.88	28.81	23.30	20.34	23.06	12.50	27.76	35.96	23.20	36.80	22.05
LA	SD	6.09	6.39	3.37	4.17	8.89	1.87	6.09	6.33	9.05	16.40	4.62
	CV (%)	29.20	22.20	14.50	20.50	38.60	15.00	29.20	22.80	39.00	44.60	20.90
	Mean	0.31	0.31	0.36	0.29	0.35	0.38	0.30	0.31	0.28	0.29	0.32
TL	SD	0.03	0.03	0.02	0.02	0.01	0.03	0.02	0.02	0.04	0.04	0.02
	CV (%)	9.34	9.90	14.50	11.40	4.20	15.00	8.20	6.40	12.50	12.40	5.60
	Mean	3.50	3.26	3.75	2.93	2.33	2.73	2.93	2.76	2.58	3.41	3.05
PL	SD	0.64	0.42	0.67	0.69	0.65	0.37	0.49	0.44	0.52	0.41	0.57
	CV (%)	18.30	12.90	17.90	23.70	27.90	13.60	16.80	16.10	20.10	11.90	18.70
	Mean	5.42	6.90	6.71	5.97	7.13	4.28	6.58	8.06	5.72	7.21	6.05
LL	SD	1.35	1.33	1.13	1.54	1.87	1.16	1.27	1.96	1.45	2.15	1.58
	CV (%)	24.90	19.30	16.80	25.70	26.20	27.20	19.20	24.30	25.40	29.80	26.10
	Mean	5.59	6.31	5.48	5.39	6.01	4.33	6.36	5.90	5.59	6.17	5.21
MLW	SD	0.87	0.87	0.57	1.05	1.61	0.62	0.95	0.70	0.96	1.50	0.58
	CV (%)	15.50	13.80	10.30	19.50	26.80	14.40	15.00	11.90	17.20	24.2	11.10
	Mean	1.00	1.11	1.25	1.20	1.24	0.93	1.06	1.36	1.02	1.17	1.18
LL:MLW	SD	0.35	0.26	0.30	0.58	0.46	0.37	0.29	0.30	0.19	0.30	0.35
	CV (%)	35.30	24.00	24.10	48.50	37.00	40.50	27.00	22.20	18.70	25.80	29.80
	Mean	0.67	0.49	0.58	0.56	0.36	0.66	0.45	0.37	0.48	0.52	0.54
PL:LL	SD	0.15	0.12	0.18	0.18	0.14	0.15	0.09	0.14	0.16	0.18	0.15
	CV (%)	22.40	24.70	31.20	20.50	37.60	22.70	19.80	37.40	32.80	34.30	27.60
	Mean	2.29	2.80	2.41	2.10	2.82	1.30	2.17	2.54	2.15	2.24	2.06
DLL	SD	0.47	0.63	0.37	0.35	0.56	0.17	0.32	0.45	0.29	0.48	0.23
	CV (%)	20.50	22.60	15.20	16.70	19.70	13.40	14.90	17.80	13.70	21.40	10.90
	Mean	2.78	3.05	2.78	2.69	2.44	2.14	3.01	2.98	2.75	3.00	2.65
DLM	SD	0.50	0.45	0.29	0.54	0.82	0.37	0.38	0.32	0.55	0.70	0.27
	CV (%)	17.80	14.90	10.50	20.00	33.50	17.30	12.90	10.90	20.20	22.80	10.20
	Mean	8.96	9.09	7.21	11.67	8.89	7.17	8.20	4.75	7.43	6.00	6.00
NMS	SD	2.06	2.45	1.51	1.87	2.30	1.47	1.42	0.96	2.37	0.82	1.82
	CV (%)	23.00	22.20	20.20	16.00	25.80	20.50	17.40	20.10	31.90	13.60	30.40
	Mean	35.30	27.55	34.06	35.30	32.90	34.37	35.41	32.10	36.90	36.20	36.37
AMV	SD	5.90	3.66	3.41	1.93	3.96	1.80	5.53	3.53	3.90	3.63	3.16
	CV (%)	16.70	13.30	10.00	5.40	12.00	5.20	15.60	11.00	10.60	10.00	8.70
	Mean	32.30	30.50	31.97	30.80	33.03	33.03	34.37	35.37	30.33	31.70	29.67
LDM	SD	0.40	0.61	0.70	4.17	0.40	0.86	1.22	0.74	0.55	0.36	0.45
	CV (%)	1.20	2.00	2.20	2.30	1.20	2.60	3.50	2.10	1.70	1.14	1.50
	Mean	4.30	3.37	2.47	5.16	8.90	4.40	4.85	2.07	1.80	2.32	1.80
MDL	SD	0.80	0.73	0.35	0.81	2.50	0.32	0.64	0.15	0.23	0.61	0.35
	CV (%)	18.50	21.70	14.30	15.80	28.20	7.40	13.10	7.40	13.00	26.10	19.50

susceptible correlation with mean annual rainfall (MAR) (see Table IV).

Three distinct populations of *P. euphratica* have been recognized on tri-dimensional plot in (Fig. 4). Each group of samples collected from Mahneshan (S5) and Gherekhar (S6) form a separate population, while all other groups of samples taken from the rest of the country develop a single population.

DISCUSSION

In this study, the morphological traits of leaves provided discriminatory grounds for separating various populations of *P. euphratica*. Discrepancies of morphological characteristics among populations may furnish basis for selecting and breeding of *P. euphratica*. The relationships among morphological traits and geo-climatic parameters show that there is a controlling cline in distribution of *P. euphratica* populations. Several workers such as Barnes (1975), Sokal *et al*. (1986), Kundu and Tigerstedt (1997), Bruschi *et al*. (2003) investigated on other species and reported results similar to this study. Clinal variations in morphological characteristics among populations of *P. euphratica* may be caused by geographical parameters (e.g. altitude, latitude, longitude). The good correlations among morphological parameters such TL, LDM and MDL with mean annual temperature (MAT) changes may indicate the adapting capability of *P. euphratica* to a wide range of temperature. Similar relationships also were reported for European black poplar and black spruce by Kajba *et al*. (2004) and Wei *et al*. (2004), respectively. The lack of correlation between mean annual rainfall (MAR) and the morphological parameters may suggest that *P. euphratica* can acquire its water requirements from the river, since it grows normally along the river banks and active flood plains. The European black poplar, however, grows in temperate climatic zones, where the average annual precipitation changes strongly affect on some of its morphological traits (Kajba *et al*., 2004). The changes of petiole length (PL) of the *P. euphratica* among

Table IV. Correlation coefficients among the leaf morphological traits and the environmental factors of *P. euphratica*

Traits	LA	TL	PL	LL	MLW	LL/MLW	PL/LL	DLL	DLM	NMS	AMV	LDM	MDL
TL	-0.60 *												
PL	0.31	-0.04											
LL	0.74 **	-0.29	0.04										
MLW	0.85 **	-0.56	0.10	0.78 **									
LL/MLW	0.37	-0.05	0.02	0.84 **	0.36								
PL/LM	-0.46	0.24	0.56	-0.78 **	-0.64 *	-0.61 *							
DLL	0.56	-0.24	0.07	0.80 **	0.80 **	0.60 *	-0.64 *						
DLM	0.84 **	-0.70 *	0.43	0.65 *	0.83 **	0.30	-0.33	0.52					
NMS	-0.29	-0.11	-0.04	-0.30	0.05	-0.24	0.21	0.10	-0.13				
AMV	-0.13	-0.23	-0.03	-0.39	-0.33	-0.24	0.30	-0.56	-0.17	-0.12			
LDM	0.01	0.25	-0.22	0.32	0.17	0.21	-0.34	0.05	0.09	-0.29	-0.14		
MDL	-0.25	0.33	-0.45	-0.05	0.09	-0.03	-0.20	0.20	-0.40	0.60 *	-0.17	0.27	
MAT	0.49	-0.62 *	0.24	0.14	0.16	0.10	-0.01	0.07	0.35	-0.32	0.45	-0.63 *	-0.62 *
MAR	-0.33	-0.11	-0.32	0.13	-0.13	0.28	-0.27	0.12	-0.47	-0.01	-0.16	0.57	-0.45
Alt	-0.01	-0.11	-0.35	0.16	-0.14	0.14	-0.28	-0.13	-0.01	0.60 *	0.04	0.34	-0.45
Long	0.05	-0.14	0.62 *	-0.07	0.16	0.16	0.43	0.36	0.23	0.30	-0.26	-0.25	-0.07
Lat	-0.33	0.40	-0.06	-0.33	-0.03	-0.41	0.23	-0.09	-0.20	0.65 *	-0.37	0.26	0.64 *

**, * Significant at 1% and 5% levels of probability, respectively0

the various populations may be due to clinal variation pattern in western, northwestern and eastern parts of the country. Moreover, the maximum depth of leaf serrate (MDL) among northern populations is greater than that of the southern ones. There are great differences in the morphological parameters between populations in the eastern zone (e.g. S1) and the ones in the western zone (e.g. S6). Populations growing in similar geo-climatic conditions (e.g. S9, S10 & S11) exhibit similar morphological

Fig. 2. Relationship between geographic latitude (LAT) and MDL (a), NMS (b) of *P. euphratica* from 11 sampling sites

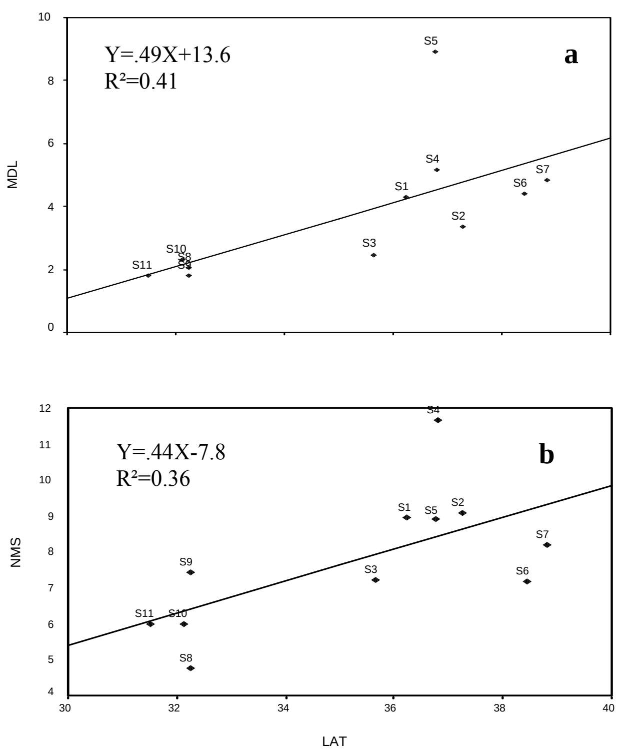
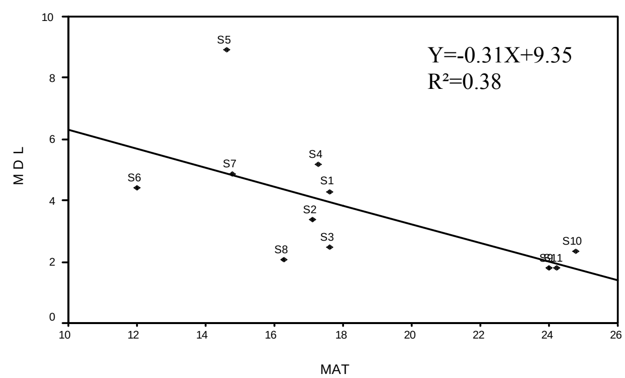


Fig. 3. Relationship between mean annual temperature (MAT) and MDL of *P. euphratica* from 11 sampling sites



characteristics. The population in site S6 representing growing zone of high latitude and altitude generally displays lower LL, LA and MLW. The population in site S5, which represents growing zone of relatively higher altitude features characteristically higher number of main serrate (NMS) and maximum depth of leaf serrate (MDL). The third population shows intermediate morphological characteristics.

In conclusion, all the populations of *P. euphratica* are well adapted to the environmental conditions of Iran and may be interesting source of genetic diversity. The data also furnishes reasonable basis for improvement of afforestation operations.

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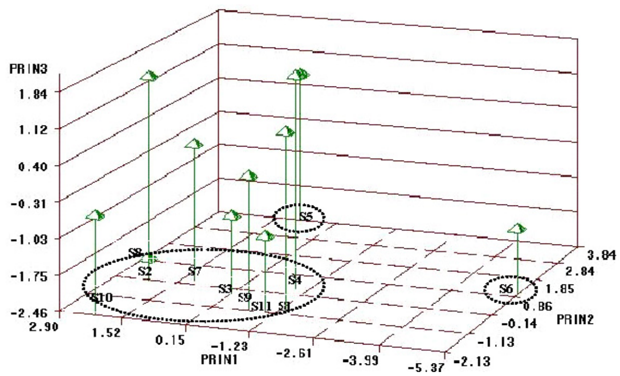
Fig. 4. Relationship among first, second, and third principal components showing the distribution of sampling sites on the basis of PCA. The first component (X axis) represents morphological traits such as LA, LL and MDL; the second component (Y axis) represents latitude and MDL; the third component (Z axis) represents mean annual temperature (MAT)



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