Leaf Morpho–physiology and Leaf-Fe Content of Selected Quince Genotypes from Different Parts of Iran

Mitra MIRABDULBAGHI (✉)
Hamid ABDOLLAHI

Summary

The objectives of this study were to compare genotype variability of leaf morpho-physiology and leaf-Fe content, as well as to select quince genotypes possessing desirable characteristics for possible use in breeding projects. Leaves were sampled from 28 quince genotypes that were selected from different parts of Iran. Selected genotypes were grown under the same environmental conditions in nursery of Seed and Plant Improvement Institute. The results suggest that estimated variations of studied leaf chlorophyll fluorescence parameters were slight, but statistically significant. The highest variability was estimated for the leaf area, and somewhat lower for the specific leaf area. The leaves of genotype *KM1* had the smallest amount of leaf area and leaf laminar length. Leaf chlorophyll (SPAD-Values) and leaf laminar petiole were the highest for the genotype *NB2*. The genotype *SHA1* had the highest minimum chlorophyll fluorescence (F0). The highest value of fluorescence variable (FV) and chlorophyll fluorescence (FM) belonged to *Moghavem2*. The lowest minimum chlorophyll fluorescence (F0) and the highest value of photochemical capacity of photosystem 2 (FV/FM) belonged to the *Khosro.* The highest amount of leaf laminar width, leaf dry weight and leaf area belonged to *sahelborgmoghavem*. The leaves of genotype *KVD1* had the highest amount of specific leaf area. Simple correlation analysis showed significant negative and positive correlations for some important characteristics. Factor analysis revealed that chlorophyll fluorescence (FM), fluorescence variable (FV), minimum chlorophyll fluorescence (F0) and leaf area were related to the main factor components. Cluster analysis for selective factors divided quince genotypes to five main groups.

Key words

leaf morphology, chlorophyll fluorescence parameters, leaf chlorophyll, leaf-Fe content, quince genotypes

[1] Department of Horticulture, Seed and Plant Improvement Research Institute, Karaj, Iran
✉ e-mail: mitra_mirabdulbaghi@yahoo.com

Received: January 11, 2014 | Accepted: February 23, 2014

Introduction

Quince (*Cydonia oblonga* Mill.) belongs to the *Maloideae* subfamily of the *Rosaceae* family, which includes commercially important fruits such as apples and pears. This subfamily comprises approximately 1,000 species in 30 genera and is characterized by a distinctive fruit, pome, and a base chromosome number of 17 (Rodger and Campbell, 2002). Quince was thought to have originated in Persia, Turkistan and the Caucasus. Its tree is used as rootstock for pear cultivars, while its fruit is used for making preserves, jam, marmalade, sauce and juice (Halasz et al., 2009). The quince tree shows high genetic variability, the following authors studied the genetic variability in this species: Scaramuzzi (1957), Reina et al. (1985), Janda and Gavrilovic (1987), Rotaru and Lobachev (1990) Shao and Lu (1995), Ercisli et al. (1999), Yarlgac (2001) and Srivastava et al. (2005). Though quince is no longer a popular fruit tree species, it is still the most important rootstock for pear cultivation. Being highly sensitive, they are continuously subjected to environmental conditions as well as to phenological cycles and growth rhythms (Bussoti et al., 2000), and these occurrences involve visual symptoms and physiological/ultra-structural changes. Numerous studies have shown that various water, light, temperature and CO2 regimes can influence leaf morphology, structure and physiology in various tree species (Onofrio and Bellocchi, 1998; Rodríguez-Guisado et al., 2009). To our knowledge, a very small number of Iranian authors studied the within-species variability of the leaf structure of quince genotypes (Abdollahi and Ghahremani., 2011; Abdollahi et al., 2013). According to Abdollahi et al. (2013) quince genotypes from the North of Iran with most similarity to the wild ancestors demonstrated low fruit quality, late to very late fruit maturity and high fruit set. These genotypes also clustered as the most dwarfing and showed the lowest level of leaf chlorosis in calcareous soils.

Our hypothesis was that structural characteristics of leaves differ among selected quince genotypes from different parts of Iran within the same stand and that these differences are under genetic control. Hence, objective was to compare the variability of selected characteristics, as well as to select genotypes possessing desirable characteristics for possible use in breeding projects, for example, selecting the genotypes with the lowest level of leaf chlorosis in calcareous soils

Material and methods

The plant material used in this investigation belonged to the breeding programs of Iranian National Quince collection from different parts of Iran (Isfahan, Khorasan Orumia, Ardebil, Astara and Tehran) during 2006-2009 (Fig. 1). All selected quince genotypes were budded on quince seedling rootstocks in 2012, and then grown under the same environmental conditions in nursery of Seed and Plant Improvement Institute. In the present work, leaves were sampled from twenty eight quince genotypes that were selected from Central, Central-North, North, North West and North East regions of Iran (Fig. 1). For specific leaf area (SLA), leaf area, leaf laminar width, petiole and laminar lengths and also leaf-Fe content, five leaves were sampled per quince genotypes in July. The mean leaf area of individual genotypes (cm^2) was determined by portable leaf area meter LI — 3000 (Li-Cor, USA). These leaves were dried in paper bags at 105°C to constant weight, and their mass was measured to estimate a value for specific leaf area (cm^{2}g dry matter^{-1}). Leaves from each genotype were oven-dried and ground for chemical analysis. Iron was determined by atomic absorption spectophotometry (AOAC, 1980). The plant chlorophyll was indirectly measured during the experimental period using a portable SPAD-502 device (Minolta Camera CO, Ltd., Japan) in two young expanded leaves with two readings per leaf. Chlorophyll fluorescence parameters (F0: minimum fluorescence; Fm: maximum fluorescence; Fv = Fm - F0: variable fluorescence) and value of photochemical capacity of photosystem 2 (FV/FM) were measured with a portable fluorimeter (Plant Efficiency Analyser, PEA, Hansatech Instruments Ltd., England). Prior to the measurements, the leaves were kept in the dark for 30 min using cuvettes. A 5-s light pulse at 400 μmolm−2 s−1 was used. Leaf chlorophyll (Chl) concentration was estimated using a portable SPAD-502 meter (Minolta, Osaka, Japan). The experiment was conducted in a Randomized Complete Block Design with three replications. The statistical evaluation was done by using analysis of variance (ANOVA). The statistical analysis included: an analysis of variance (ANOVA) evaluated using Duncan's multiple range test at P=0.05 to compare and detect any significant differences among the studied quince genotypes; a test for evaluating the relationships between studied parameters by using Pearson's correlation coefficients at $P \leq 0.05$, and a factor analysis where relative variance of any factor indicates the percent importance of related factor in total variance of studied traits. In the present study cluster analysis (based on Ward's method) was carried out based on the main factors that demonstrated the highest of total variance.

Figure 1. Geographic sources of the 28 Iranian quince genotypes used in this study

Results and discussion

Table 1 showed that there were significant differences ($p \leq 0.01$), between studied quince genotypes in respect to the all studied traits. The results suggest that estimated variations of studied leaf chlorophyll fluorescence parameters were slight, but statistically significant. The highest variability was estimated for the leaf area, and somewhat lower for the specific leaf area (11.97) and the leaf area (13.95%). These quantitative differences, illustrating intraspecies variability of parameters were studied. According to Castro-Diez et al. (1997), the within-species variability of leaf morphology and structure may improve plant performance, laminar width (8.1 cm), leaf dry weight (0.48 g) and leaf area 37.8 cm^2) belonged to *sahelborgmoghavem*. The leaves of genotype *KVD1* had the highest amount of specific leaf area (83.27$cm^2 \cdot g^{-1}$). An earlier investigation on the Iranian quince genotype from different parts of Iran showed that Quince genotypes from the North with most similarity to the wild ancestors demonstrated low fruit quality, late to very late fruit maturity and high fruit set. These genotypes also clustered as the most dwarfing and showed the lowest level of leaf chlorosis in calcareous soils (Abdollahi et al., 2013).

Table 1. Leaf structural characteristics of selected quince genotypes from different parts of Iran

Quince genotype	SPAD-Value	F0	FM	FV/Fm	Leaf area (cm^2)	Leaf-Fe (mg/L)	FV (Fm-F0)	Leaf laminar width (cm)	Leaf laminar length (cm)	Leaf dry weight (g)	Specific leaf area ($cm^2 \cdot g^{-1}$)	Leaf laminar petiole (g)
KVD2	18.5	53	509	0.895	16.73	32.86	456	5.15	6.15	0.31	54.70	1.13
ASM3	40.1	116	522	0.77	16.22	57.65	406	5.40	6.05	0.32	50.51	2.00
KVD3	20.6	66	351	0.811	26.62	37.81	285	6.55	7.90	0.39	66.66	1.27
SVS2	25.1	50	423	0.881	17.19	36.85	373	5.50	6.00	0.33	52.63	1.51
KVD4	14.4	117	566	0.793	20.69	29.18	449	5.90	7.20	0.35	58.96	0.88
Khosro	28.2	34	400	0.915	30.52	22.21	366	7.15	8.40	0.43	71.47	1.69
PH2	14.6	128	311	0.588	16.93	32.40	183	5.35	6.45	0.32	53.08	0.90
Sahelborgmoghavem	16.8	151	497	0.696	37.80	35.68	346	8.1	8.75	0.48	78.16	1.01
ET1	20.0	130	554	0.765	24.44	39.18	424	6.4	7.63	0.38	64.34	1.20
NB2	85.4	111	491	0.773	24.23	27.77	380	5.95	7.7	0.35	69.04	2.01
KM1	17.7	190	571	0.667	15.88	23.90	381	6.05	5.85	0.36	44.12	1.08
ASP1	27	192	514	0.626	25.35	28.22	322	6.35	7	0.38	67.04	1.62
Esphehanoghaf	19.4	108	520	0.792	36.26	18.17	412	7.9	8.9	0.47	77.30	1.18
SVS1	12.5	107	477	0.775	24.15	41.08	370	6.25	7.15	0.37	64.82	0.76
ASM1	27	112	495	0.773	16.96	49.01	383	5.6	6.15	0.33	50.93	1.62
Unknown	32.9	120	486	0.753	31	32.99	366	7.1	8.75	0.42	73.50	1.98
KVD1	20.6	116	602	0.807	37.58	36.93	486	7.55	8.2	0.45	83.28	1.25
ASM2	20.8	111	572	0.805	20.77	35.07	461	5.5	6.1	0.33	64.39	1.26
PK2	29	122	500	0.756	17.08	34.86	378	5.2	6.3	0.31	54.43	1.74
Behtorsh	19.5	107	510	0.79	24.13	40.65	403	7.1	7.45	0.42	57.23	1.19
ASP2	17.3	120	463	0.74	24.93	28.99	343	6.9	6.40	0.41	61.11	1.03
SHA1	21.5	224	582	0.741	21.9	31.27	358	5.95	7.10	0.35	61.80	1.31
NB3	22.2	123	533	0.769	31.77	28.22	410	7.20	8.00	0.43	74.31	1.33
NB4	14.6	123	604	0.796	17.18	26.80	481	7.00	7.90	0.41	41.42	0.90
AS2	27.8	154	517	0.702	23.03	25.78	363	6.25	7.00	0.37	61.94	1.67
Moghavem1	27.4	159	573	0.722	29.35	34.40	414	6.87	7.80	0.41	71.97	1.66
Moghavem2	23.3	116	626	0.814	22.70	32.99	510	6.25	7.00	0.37	61.32	1.40
Gardandar	25.4	122	575	0.787	16.35	41.17	453	5.58	6.40	0.33	49.33	1.53
LSD5%	0.64	1.03	0.78	0.01	5.45	0.19	1.26	0.68	0.72	0.04	12.21	0.04
CV (%)	1.59	0.47	0.10	0.395	13.945	1.10	0.19	6.26	6.00	6.3	11.97	1.74

allowing species to maintain their fitness in resource availability. The results of the leaf parameters taken are shown in Table 3. The leaves of genotype *KM1* had the smallest amount of leaf area (15.88 cm^2) and leaf laminar length (5.85 cm). Leaf chlorophyll (SPAD-Values) (85.4) and leaf laminar petiole (2.01 cm) were the highest for the genotype *NB2*. The genotype *SHA1* had the highest minimum chlorophyll fluorescence (F0) (224). The highest value of fluorescence variable (FV) (510) and maximum chlorophyll fluorescence (FM) (626) belonged to *Moghavem2*. The lowest minimum chlorophyll fluorescence (F0) (34) and the highest value of photochemical capacity of photosystem 2 (FV/FM) (0.915) belonged to the *Khosro*. The highest amount of leaf

Simple correlation analysis showed significant negative and positive correlations for some important characteristics. Positive correlation was observed between maximum chlorophyll fluorescence (FM) and minimum chlorophyll fluorescence (F0) as well as fluorescence variable (FV) (Table 2). In contrast, photochemical capacity of photosystem 2 (FV/FM) had negative correlation with minimum chlorophyll fluorescence (F0). There was positive significant correlation between leaf laminar width and leaf area, as well as between leaf area, leaf laminar width and leaf laminar length. Changes in the fluorescence variables cause alterations in the Fv/Fm ratio, indicating a disturbance in the photochemical activity of photosynthesis. The Fv/Fm ratio

Table 2. Similarity coefficient among selected quince genotypes from different parts of Iran

	SPAD-Value	F0	FM	FV/Fm	Leaf-area	Leaf-Fe	FV	Leaf laminar width	Leaf laminar length	Leaf dry weight	Specific leaf area	Leaf laminar petiole
SPAD-Value	1											
F0	-.053	1										
FM	-.044	.452*	1									
FV/Fm	.050	-.77**	.069	1								
Leaf-area	-.016	.003	.011	.023	1							
Leaf-Fe	.042	-.118	-.013	.077	-.309	1						
FV	-.016	-.115	.834**	.554**	.010	.059	1					
Leaf laminar width	-.170	.058	.100	-.005	.881**	-.371	.075	1				
Leaf laminar length	.051	-.048	.005	.088	.859**	-.356	.036	.860**	1			
Leaf dry weight	-.170	.058	.100	-.005	.881**	-.371	.075	1.000**	.860**	1		
Specific leaf area	.125	-.021	-.028	.043	.938**	-.253	-.018	.673**	.747**	.673**	1	
Leaf laminar petiole	.741**	-.033	-.008	.087	-.007	.203	.012	-.176	-.004	-.176	.117	1

** - Correlation is significant at the 0.01 level; * - Correlation is significant at the 0.05 level.

Table 3. Eigen values, relative variance and cumulative variance percentage for selected quince genotypes from different parts of Iran

Component	Initial Eigenvalues		
	Total	% of variance	Cumulative %
Maximum chlorophyll fluorescence (FM)	4.532	37.770	37.770
Fluorescence variable (FV)	2.120	17.669	55.440
Minimum chlorophyll fluorescence (F0)	1.957	16.310	71.750
Leaf area	1.694	14.1107	85.867

has been inferred as an indicator of environmental stress, such as high temperature, drought and excess light, as it is easy and fast to measure (Maxwell and Johnson, 2000). Furthermore, as shown in Table 2, there were significant positive relationships between leaf laminar width, leaf laminar length, leaf dry weight, specific leaf area and leaf area. Leaf chlorophyll (SPAD-Values) had positive significant correlation with leaf laminar petiole. In contrast to Prado and Vara (2011), we did not observed any significant correlation between leaf–Fe content, leaf dry weight and Leaf chlorophyll (SPAD-Values).

In factor analysis, four main and independent factors with Eigen values ≥ 1 interpreted 86% of total variance (Table 3). In the first factor, maximum chlorophyll fluorescence (FM) with positive coefficients interpreted 37.77% of total variance. In the second fluorescence variable (FV) with positive efficient interpreted about 17.67% of total variance. In the third factor, minimum chlorophyll fluorescence (F0) with positive coefficients interpreted 16.31% of total variance. The forth factor, leaf area demonstrated 14.11% of total variance. Factor analysis had great potential to differentiate the highlighted distinctions between studied genotypes (Kaufmane et al., 2002; Ogasanovic et al., 2007).

In the present study cluster analysis was carried out based on four selective main factors (maximum chlorophyll fluorescence (FM), fluorescence variable (FV), minimum chlorophyll fluorescence (F0) and leaf area that demonstrated 86% of total variance. According to the cluster analysis for selective factors (Fig. 2), quince genotypes were divided to five main groups (Khoramdel, 2013).

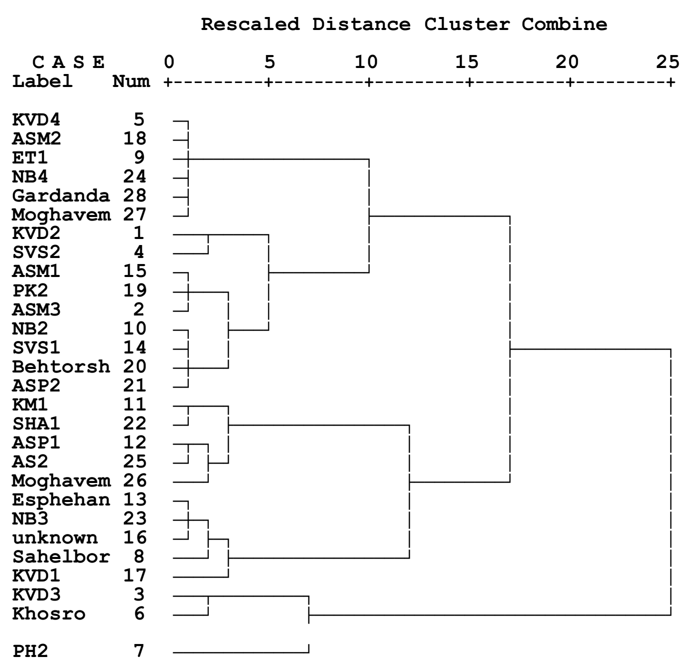

Figure 2. Cluster analysis (based on Ward's method) for four selective factors of quince genotypes from different parts of Iran

Conclusion

Our results approve the genetic basis of leaf morpho- physiology and leaf-Fe content differences observed among studied quince genotypes and also select quince genotypes possessing desirable characteristics for possible use in breeding projects, for example, propagations of quince genotypes that induced a higher tolerance to iron deficiency.

References

AOAC (1980). Official Methods of Analysis Association of Official Analytical Chemists. 13th Edn., Association of Official Analytical Chemists, Washington, DC. USA.

Abdollahi H., Ghahremani Z. (2011). The Role of Chloroplasts in the Interaction between Erwinia amylovora and Host Plants. Acta Hort 896: 215-221

Abdollahi H., Alipour M., Khorramdel Azad M., Ghasemi A., Adli M., Atashkar D., Akbari M., Nasiri J. (2013). Establishment and primary evaluation of quince germplasm collection from various regions of Iran. Acta Hort 976:199-206

Bussoti F., Borghini F., Celesti C., Leonzio C., Bruschi P. (2000). Leaf morphology and macronutrients in broadleaved trees in central Italy. Trees 14: 361-368

Castro-Diez P., Villar- Salvador P., Perez-Rontome C., Maestro-Martinez M., Montserrat-Marti G. (1997). Leaf morphology and leaf chemical composition in three *Quercus* (*Fagaceae*) species along a rainfall gradient in NE Spain. Trees 11: 127-134

Ercisli S., Guleryuz M., Esitken A. (1999). A study on the fruit properties of native quince cultivars in Oltu. Anadolu 9: 32–40

Janda L., Gavrilovic J. (1987). Technological characteristics of the fruit in some quince varieties and their suitability for processing. Jugoslovensko vocarstvo 21: 41-46

Halász J., Hoffmann V., Szabó Z., Nyéki J., Szabó T., Hegedûs A. (2009). Characterization of quince (*Cydonia oblonga* Mill.) cultivars using SSR markers developed for apple. Int J Hortic Sci 15: 7-10

Kaufmane E., Ikase L., Trajkovski V., Lacis G. (2002). Evaluation and characterization of plum genetic resources in Sweden and Lativa. Acta Hort 577: 207-213

Khoramdel M., Jaber Nasiri A., Abdollahi H., (2013). Genetic Diversity of Selected Iranian Quinces Using SSRs from Apples and Pears. Biochem Genet 51: 426-442.

Maxwell K., and Johnson G. N. (2000). Chlorophyll fluorescence - A pratical guide. Exp Bot 51: 659-668

Ogasanovic D., Plazinic R., Rankovic M., Stamenkovic S., Milinkovic V. (2007). Pomological characteristics of new plum cultivars developed in Cacak. Acta Hort 734: 165-168

Onofrio C. D., Morini S., Bellocchi G. (1998). Effect of light quality on somatic embryogenesis of quince leaves. Plant Cell Tiss Org 53: 91-98

Prado ı R. M., Alcantara-Vara E., (2011). Tolerance to iron chlorosis in non-grafted quince seedlings and in pear grafted onto quince plants. J Soil Sci Plant Nutr 11: 119-128

Reina A., Fanizza G., Giorgio V., (1985). Study of the relationships between fruit characters in a quince population in Puglia and Basilicata. Informatore Agrario 41: 74-75.

Rodger C. E., Campbell C.S. (2002). The origin of the apple subfamily (Maloideae; Rosaceae) is clarified by DNA sequence data from duplicated GBSSI genes. Am J Bot 89: 1478–1484

Rodríguez-Guisado I., Hernández F., Melgarejo P., Legua P., Martínez R., Martínez J. J. (2009). Chemical, morphological and organoleptical characterisation of five Spanish quince tree clones (*Cydonia oblonga* Miller). Sci Hortic 122: 491-496

Rotaru G.I., Lobachev A. (1990). Comparative anatomical characteristics of fruits of new quince cultivars Nakhodka and Volgogradskaya Myagkoplodnaya. Izvestiya Akademii Nauk Moldavskoi SSR Biologicheskikh i Khimicheskikh Nauk 1: 16-21

Shao Z. X., Lu B. (1995). Resources of Chinese quince in Yunnan province. J Fruit Sci 12 (Suppl.): 155-156

Scaramuzzi F., (1957). Contributo allo Studio delle cultivar di cotogno da frutto. Rivista di Ortoflorofrutticoltura Italiana 41: 575-615

Srivastava K. K., Jabeen A., Das B., Sharma A. K. (2005). Genetic variability of quince (Cydonia oblonga) in Kashmir valley. Indian J Agr Sci 75: 766-768

Yamamoto T., Kimura T., Soejima J., Sanada T., Ban Y., Hayashi T. (2004). Identification of quince varieties using SSR markers developed from pear and apple. **Breed** Sci 54: 239-244

Yarlgac T. (2001). Morphological characteristics of wild quince forms grown in Gevasdistrict (van). Ondokuz Mays Universitesi, Ziraat Fakultesi Dergisi 16: 43-49

acs79_16