ISSN 1996-0875 ©2012 Academic Journals

10011 1000 0010 02012 / loadoffilio dodiffial

Full Length Research Paper

Analysis of essential oil from nine distinct genotypes of Iranian Damask rose (Rosa damascena Mill)

Akbar Karami^{1*}, Parviz Zandi², Morteza Khosh-Khui¹, Hassan Salehi¹ and M. Jamal Saharkhiz¹

¹Department of Horticultural Science, College of Agriculture, Shiraz University, Shiraz, Iran. ²Research Center for Agriculture and Natural Resources of Fars Province, Shiraz, Iran.

Accepted 4 September, 2012

The chemical compositions of essential oil from nine distinct genotypes of Iranian *Rosa damascena* (Fars1, Fars2, Tehran, Mazandaran, Gilan, East Azar, Ardabi, Kermanshah and Qom) were determined following gas chromatography (GC) and gas chromatography-mass spectrometry (GC/MS) analysis. The main essential oil components identified in the different genotypes were citronellol, geraniol, neral, linalool and nonadecane. The relative percentage of citronellol was highest in Fars1 (42.2%) and Gilan (40.3%) genotypes and lowest in the Qom (2.2%) and East Azar (0.6%) genotypes, while in Mazandaran, Ardabil and Kermanshah genotypes citronellol was not detected. The highest content of geraniol was detected in the essential oil of the Kermanshah genotype (37.5%); however, in the Fars1, Fars2, Tehran, Gilan, East Azar, Kermanshah and Qom genotypes this component was not detected. Linalool was detected in the essential oil of all genotypes, and the highest amount of this component was in Fars2 (39.2%). Hydrocarbons had the highest contribution to the essential oil of the East Azar genotype. The major hydrocarbons identified in all genotypes were nonadecane (10.7 to 51.2%), heneicosane (3.7 to 18%), eicosane (0.8 to 6.2%) and tricosane (0.5 to 2.4%).

Key words: Rosa damascena, essential oil, genotypes.

INTRODUCTION

The damask rose (*Rosa damascena* Mill.) has been grown in Iran for centuries for the production of rose water and rose oil, which are currently widely applied in the perfumery, cosmetics and the food industry. The blooming of *R. damascena* happens once a year at the beginning of the summer and the blooming period is extremely intensive. The rose bushes produce daily a large number of blooming flower buds and full-blown flowers, which are picked by hand and subjected to steam distillation within the same day (Dobreva and Kovacheva, 2010). The majority of the rose buds that start blooming early in the morning hours reach full-blown before noon and have to be collected on the same day. Faded flowers fully blown within the previous day are not

harvested (Rusanov et al., 2011).

Iran is one of the most important rose oil and rose water producers in the world. Iran has been mentioned as a country of possible origin of Damask roses and in this country, cultivation and consumption of R. damascena has a long history (Kiani et al., 2007; Jalali-Heravi et al., 2008). Interestingly, several molecular marker based studies (Baydar et al., 2004; Rusanov et al., 2005) revealed that the *R. damascena* plants used for essential oil production in Bulgaria and Turkey, which are currently one of the main producers of rose oil in the world, represent a single genotype. This genotype also dominates in the major production zone of Iran although much diversity was found throughout the production areas of Iran (Babaei et al., 2007; Kiani et al., 2007). Cluster analysis based on microsatellite markers of 40 Iranian accessions of R. damascena resulted in nine distinct genotypes which were used in this study (Babaei et al., 2007).

^{*}Corresponding author. E-mail: akarami2004@gmail.com. Tel: +989177366760.

Comparative gas chromatography (GC) and gas chromatography-mass spectrometry (GC/MS) analyses of rose oil composition have been used for characterization of rose oils distilled from flowers of *R. damascena* cultivated in different geographic regions and/or representing different genotypes. In addition GC–MS analysis was applied for the characterization of other rose flower products, including rose concrete, rose absolute and rose water (Kurkcuoglu and Baser, 2003; Ayci et al., 2005; Rusanov et al., 2011). This study was aimed at evaluating and comparing the essential oil composition in nine distinct genotypes of Iranian damask roses, which could be useful for their application in future breeding programs.

MATERIALS AND METHODS

Plant material and essential oil distillation

The flowers of nine distinct genotypes of Iranian *R. damascena* Mill. (Fars1, Fars2, Tehran, Mazandaran, Gilan, East Azar, Ardabi, Kermanshah and Qom) as described by Babaei et al. (2007), were harvested before sunrise in full bloom stage from plants grown in the Estahban Research Center. Fresh flowers (400 g) were hydrodistilled (2.0 L of water) for 3 h using an all glass Clevenger-type apparatus to extract oils according to the method outlined by the European Pharmacopoeia. The extracted oils were dried over anhydrous sodium sulphate and stored in sealed vials at low temperature (4°C) before GC and GC/MS analysis.

GC analysis

GC analysis was performed using a Shimadzu GC-9A gas chromatograph equipped with a DB-5 fused silica column (30 m \times 0.25 mm, film thickness 0.25 μM). The oven temperature was held at 40°C for 5 min and then programmed to 220°C at a rate of 4°C/min. In brief, 1 μI of sample was injected with splitless injection methods. Injector and detector (FID) temperature were 240°C; helium 5.0 was used as carrier gas with a linear velocity of 32 cm/s. Percentage were calculated by electronic integration of flame ionization detector (FID) peak areas without the use of response factor correlation.

GC/MS parameters

The GC/MS analysis was performed using a Saturn- 3400 gas chromatograph, equipped with a DB-5 fused silica column 30 m \times 0.25 mm, film thickness 0.25 mm). The oven temperature was held at 60°C and programmed to 230°C, 4°C min⁻¹. Carrier gas helium 5.0 with a linear velocity of 31.5 cm s⁻¹, split ratio 1/60, ionization energy 70 eV; scan time 1 s and mass range of 40 to 300 amu.

Identification of the essential oil components

The components were identified by comparing their retention indices and mass spectra with those of authentic standards. The confirmation of identity was done by comparison of their mass spectra with those reported in the literature and reference compounds. The percentage evaluation of the oil components was made by

by area normalization.

Cluster analysis

Cluster analysis was used to classify nine distinct genotypes based on the oil compositions according to Ward's minimum variance method by means of the statistical package SPSS software (IBM).

RESULTS AND DISCUSSION

The chemical composition of the essential oils isolated from nine distinct genotypes of *R. damascena* is presented in Table 1. The applied GC-MS metabolite profiling resulted in the identification of a total of 24 compounds based on comparison with MS library, consisting of compounds from rose essential oils. The number of compounds identified in each genotype is presented in Table 1. As shown in the table, the highest number of identified compounds was in Tehran, Qom and Kermanshah.

Moreover, the main essential oil components identified in the different R. damascena genotypes were citronellol, geraniol, neral, linalool and nonadecane. The relative percentage of citronellol was highest in Fars1 (42.2%) and Gilan (40.3%) genotypes and lowest in the Qom (2.2%) and East Azar (0.6%) genotypes, while in Mazandaran, Ardabil and Kermanshah genotypes citronellol was not detected. The highest content of geraniol was detected in the essential oil of the Kermanshah genotype (37.5%); however, in the Fars1, Fars2, Tehran, Gilan, East Azar, Kermanshah and Qom genotypes this component was not detected. Linalool was detected in the essential oil of all genotypes as the highest amount of this component was in Fars2 (39.2%). Hydrocarbons had the highest contribution to the essential oils of the East Azar genotype. This fraction was dominated by nonadecane (10.7 to 51.2%), heneicosane (3.7 to 18%), and eicosane (0.8 to 6.2%), which were detected in all genotypes. Verma et al. (2011) investigated the chemical composition of rose volatiles obtained from the bud, half bloom and full bloom stages essential oil from two different locations. Phenyl ethyl alcohol, citronellol and benzyl alcohol were higher at half bloom stage, while geraniol, neral, (Z)-9-nonadecane and β-damascenone were higher at full bloom stage. Furthermore, the amount of tricosane, heneicosane, pentacosane and geranial recorded were higher during the bud stage. Stoyanova and Genova (2007) also investigated the essential oil composition of roses at different stages and found that the amount of geraniol and citronellol increased with the advancement of flower developmental stage (Picone et al., 2004; Stoyanova and Genova, 2007). In this study the amounts of geraniol and citronellol in some of the analyzed nine Iranian damask rose genotypes were found to be similar to the amounts

Table 1. Essential oil composition of nine *Rosa damascena* genotypes from Iran.

Component	RI*	Genotypes								
		Fars1	Mazandaran	Tehran	Gilan	East Azar	Qom	Ardabil	Kermanshah	Fars2
Isopentyl butanoate	1058	0.7 ^{\lambda}	0.5	0.3	-	-	0.5	-	0.2	0.2
Dihydro-linalool	1134	6	1.7	17.8	3	1	30.8	12.3	11.7	39.2
Citronellal	1151	-	-	-	4.9	-	-	-	-	-
Linalyl formate	1228	-	-	-	-	-	11.9	-	-	-
Citronellol	1234	42.2	-	26	40.3	0.6	2.2	-	-	19
Neral	1245	7.1	44.8	3.7	-	-	-	31.7	-	7
Geraniol	1251	-	2	-	-	-	-	1.9	37.5	-
Geranial	1264	12.3	-	6.4	-	-	4.9	-	3.6	11.2
Citronellyl formate	1273	-	7.5	-	-	-	-	7.8	14.6	-
Undecanal	1305	2.3	-	1.2	-	-	5.5	0.7	0.7	2.7
n-Undecanol	1367	-	-	-	0.7	1.1	-	-	0.6	-
n-Pentadecane	1500	-	-	-	1.3	-	-	-	-	-
n-Tetradecanol	1673	-	-	-	-	5	-	-	-	-
Occidentalol acetate	1681	3.3	4	3.6	-	3	2.5	3.8	2.7	-
n-Tetradecanol	1693	-	-	-	5	-	-	-	-	1.3
n-Pentadecanol	1760	-	-	0.5	-	-	-	-	-	-
1-Octadecene	1771	0.4	-	-	0.7	-	-	-	0.3	-
Phenyl ethly octanoate	1864	4.4	-	-	-	-	-	-		-
n-Hexadecanol	1878	-	7.2	6.4	-	7.8	4.8	7	4.5	2.7
n-Nonadecane	1900	15.9	20	22.4	26.3	51.2	26.5	20.5	13.9	10.7
n-Eicosane	2000	0.9	1.3	1.4	2.3	6.2	1.7	1.5	0.9	0.8
n-Heneicosane	2100	3.5	5.7	5.3	8.9	18	7.4	6.5	3.8	3.7
n-Docosane	2200	-	-	2.9	-	0.8	0	-	-	-
n-Tricosane	2300	0.5	1	0.7	1.4	2.4	0.9	1.1	0.6	0.6
Total		95.5	95.7	98.6	94.8	97.1	99.6	94.8	95.6	99.1

^{*}RI: Retention indices; -: not detected; λ: numbers represent percentage from the total peak area.

of the same compounds identified in previously analyzed genotypes (Stoyanova and Genova, 2007; Verma et al., 2011). However, in some of the genotypes these components were not detected.

Cluster analyses of the nine distinct genotypes

of *R. damascena* based on 24 oil components revealed five groups including citronellol, geraniol, neral, linalool and nonadecane as the major compounds (Figure 1).

Therefore, the studied genotypes of *R. damascena* revealed five different chemotypes,

each of which was rich in one of the oil constituents. Verma et al. (2011) investigated the rose oil chemical composition of two cultivars of Damask roses obtained from the same growing area during two consecutive years (Purara I and III). The concentration of citronellol, phenyl ethyl

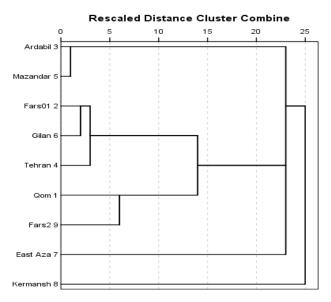


Figure 1. Dendrogram of the dissimilarity using average linkage (between groups) among nine distinct genotypes of *Rosa damascena* based on 24 oil components.

alcohol, α -pinene, β -myrcene and neral was found to be comparatively higher in Purara III, whereas the amount of geraniol, neral, linalool and geranyl acetate recorded was relatively higher in Purara I. This could be due to the variation of the altitude, climatic conditions and pedogenetic characters. In the current study, we investigated the essential oil component of Damask rose in the same year and location. Therefore, the differences in the oil content and composition of the genotypes in this present study could be attributed to their genetic variability.

Conclusion

The cluster analyses of nine distinct genotypes of *R. damascena* based on 24 oil components revealed five groups, including citronellol, geraniol, neral, linalool and nonadecane, as the major components for each group. The studied genotypes of *R. damascena* represent different chemotypes, each of which was rich in one of the oil constituents. Consequently, the differences in the oil content and composition of the genotypes could be attributed to their genetic variability and they could be a good genetic source for breeding purposes.

REFERENCES

- Ayci F, Aydinli M, Bozdemir OA, Tutas M (2005). Gas chromatographic investigation of rose concrete, absolute and solid residue. Flav. Fragr. J. 20:481-486.
- Babaei A, Tabaei-Aghdaei SR, Khosh-Khui M, Omidbaigi R, Naghavi MR, Esselink GD, Smulders MJM (2007). Microsatellite analysis of Damask rose (*Rosa damascena* Mill.) accessions from various regions in Iran reveals multiple genotypes. doi:10.1186/1471-2229-7-1212. BMC Plant Biol. 7:1- 6.
- Baydar NG, Baydar H, Debener T (2004). Analysis of genetic relationships among *Rosa damascena* plants grown in Turkey by using AFLP and microsatellite markers. J. Biotechnol. 111:263-267.
- Dobreva A, Kovacheva N (2010). Daily dynamics of essential oils of Rosa damascena Mill. and Rosa alba L. Agric. Sci. Technol. 2:271-274.
- Jalali-Heravi M, Parastar H, Sereshti H (2008). Development of a method for analysis of Iranian damask rose oil: Combination of gas chromatography—mass spectrometry with Chemometric techniques. Anal. Chim. Acta 623:11-21.
- Kiani M, Zamani Z, Khalighi A, Fatahi R, Byrne DH (2007). Wide genetic diversity of *Rosa damascena* Mill. germplasm in Iran revealed by RAPD analysis. Sci. Hort. 115:386-392.
- Kurkcuoglu M, Baser KHC (2003). Studies on Turkish rose concrete, absolute, and hydrosol. Chem. Nat. Comp. 39:457-464.
- Picone JM, Clery RA, Watanabe N, MacTavish HS, Turnbull CGN (2004). Rhythmic emission of floral volatiles from *Rosa damascena* semperflorens cv. 'Quatre Saisons'. Planta 219:468-478.
- Rusanov K, Kovacheva N, Rusanova M, Atanassov I (2011). Traditional *Rosa damascena* flower harvesting practices evaluated through GC/MS metabolite profiling of flower volatiles. Food Chem. 129:1851-1859.
- Rusanov K, Kovacheva N, Vosman B, Zhang L, Rajapakse S, Atanassov A, Atanassov I (2005). Microsatellite analysis of *Rosa damascena* Mill. accessions reveals genetic similarity between genotypes used for rose oil production and old Damask rose varieties. Theor. Appl. Gen. 111:804-809.
- Stoyanova A, Genova E (2007). Traditional Bulgarian essential oilbearing raw materials rose (*Rosa damascena* Mill.). Indian Perfumer 51:29-34.
- Verma RS, Padalia RC, Chauhan A, Singh A, Yadav AK (2011). Volatile constituents of essential oil and rose water of damask rose (*Rosa damascena* Mill.) cultivars from north Indian hills. Nat. Prod. Res. 25:1577-1584.