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Review Article

A Major Loss of Phenyl Ethyl Alcohol by the Distillation Procedure of *Rosa damascene* Mill.

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ABSTRACT

Rosa damascena Mill. is an aromatic plant, having different components that usually use in natural products. The main industrial products from oil-bearing rose are rose oil, rose water, rose concrete, and rose absolute which are produced by hydro-distillation and solvent extraction processes. However, essential oil productionis the best part that the industry is eager to it. Hydro-distillation with cohobation is a widely used method for producing volatile oils from oil-bearing rose. Newly, different methods of extraction of volatile compounds of rose oil, rose water and residue water from the flower of *R. damascena*, such as headspace solid-phase microextraction (HS-SPME) is developed. However, every method of extraction affects the quality of *Rosa* product. In this way the hydro-distilled rose oil analyzed by gas chromatography (GC-FID/GC-MS) revealed that the percentages of alkanes or steaoreptens like hexadecane (1.3%), nonadecane (7.2%) and heneicosane (1.8%) were higher than those of HS-SPME. However, there is a preference of rose oil extraction by hydro- distilation that the rose water (hydrosol) and residue water have phenylethyl alcohol (PEA), which is easily soluble in water, rose water and residue water. Actually rose water is a by-products of the hydro-distillation method, and it contained very high amounts of phenylethyl alcohol and can be employed as the best natural fragrance of the oil-bearing rose due to its very high phenylethyl alcohol content. Phenyl ethyl alcohol is a naturally occurring aromatic compound found in various flowers including roses, lavender, ylang-ylang, geranium and champagne. For determining the content of phenyl ethyl alcohol in the flower *R. damascene*, according to the geographical similarity, the flowers were collected from different provinces of Iran by scientists of RIFR. In a research in 2021, after the extraction of essential oil and rose water, it was conducted on the remaining water in the still. The leftover water inside the distillation pot was extracted with eight different solvents (chloroform, cyclohexane, dichloromethane, diethyl ether, ethyl acetate, hexane, petroleum ether and toluene) by a separatory funnel and extracted crude were analyzed by (GC) and (GC/MS). The amount of phenylethyl alcohol obtained by different solvents containing chloroform, cyclohexane, dichloromethane, diethyl ether, ethyl acetate, hexane, petroleum ether and toluene was 96.1%, 46.3%, 96.1%, 46.3%, 89.6%, 100%, 0.42% and 74%, respectively. Therefore, the research revealed the phenylethyl alcohol compound can be separated from the wastewater in the still and used.

INTRODUCTION

The genus *Rosa* belongs to the Rosaceae family and includes 200 species and more than 18,000 cultivars [1]. One of the most important rose species is *R. damascena* Mill. and some of its varieties are very important for essential oil production and the rest are widely cultivated as garden roses [2]. The *R. damascena* Mill. is cultivated for its medicinal properties, and this aspect is steadily increasing in the world. In recent years, the anti-HIV, antibacterial and antioxidant activities of *R. damascena* Mill. essential oil have been presented

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[3-6]. Rose essential oil is one of the most expensive essential oils in the world market due to low amount of essential oils in the *Rosa* genus and the absence of natural and synthetic alternatives [7]. The total global production of rose oil is approximately 5 metric tons. Bulgaria and Turkey are the main producers, followed by Morocco, Egypt, China, Russia, Iran and India [8].

Among the rose family, *R. damascena* Mill species is the most important rose species that is used to produce essential oil from *R. damascena* Mill flowers. Apart from the essential oil can be separated from the petals with a solvent extraction method, it is also extracted during the distillation of rose water, and concrete and absolute which are all valuable and important basic materials for perfumery and cosmetic industries [9].

Cohobation is a widely used method for producing rose essential oil because the flowers tend to aggregate and form clumps that cannot be distilled by steam distillation or direct steam distillation. In the industrial water distillation process, large steel tanks with a capacity of 3000 liters are filled with rose (500 kg) and water (1.5 tons) and then heated with steam for 90 minutes. Evaporated water and rose essence are taken out and enter the concentrator and then collected in the Florentine flask [10].

The essential oil separated in the Florentine flask is known as a straight essential oil, which makes up about 20% of the total essential oil. The water that condenses with the essential oil is drained off and redistilled to obtain the water-soluble fractions of the rose essential oil. After cohobation, the obtained essential oil is called "indirect essential oil", which constitutes the major part of about 80% of the total essential oil. The direct and indirect rose essences are combined to make the final rose essence of the trade (rose iron or attar). The hydrosol part of distillation is known as rose water [11,12].

By the method of solvent extraction, the main products of concrete and absolute are obtained from petals. Concrete is not widely used in perfumery and cosmetics in its native form, but is generally converted into an alcohol-soluble aromatic liquid known as absolute. To produce concrete, flowers are stirred in an extractor with a non-polar solvent such as n-hexane, which removes aromatic compounds as well as other soluble substances such as waxes and pigments. The extractive material is separated by the distillation method in circulating vacuum. The remaining waxy mass is known as concrete. Concrete is stirred with ethyl alcohol at -15 to -20 degrees Celsius to dissolve aromatic compounds and leave wax and other paraffinic substances. The alcohol fraction is evaporated at low pressure, leaving the finished absolute behind [13, 14, 15, 16]. The amount of rose essential oil is generally very low (0.03-0.04%). Out of every 3.5 tons of fresh roses picked by hand in the early hours of the flowering season, only 1 kg of rose oil is obtained after distillation with water (Bidar, 2015). Rose oil is the most widely used essential oil in perfumery and cosmetics, even despite its high price in world markets (more than 9000 euros per kilogram in 2015 value). 1 kg of concrete is extracted from 300-400 kg of fresh flowers (0.3-0.4%) [16] and 1 kg of concrete produces about 0.6 kg of absolute [15], which explains why the price of concrete and absolute is lower than essential oil [15,16].

However, the production of compost, methane biogas, phenylethyl alcohol, residual concrete, sanitary sludge, cosmetics as well as a natural antioxidant from the remaining distillation can compensate part of expences [17]. Epidermal cells of flower petals are the main source of aromatic compounds, which are a set of volatile organic molecules, for examples, they are monoterpenes, sesquiterpenes, aromatic alcohols, oxides, ethers, esters and aldehydes [18]. The relative amount of aromatic compounds is the most important parameter that determine the quality of rose products [19].

Monoterpene alcohols such as linalool, citronellol, nerol and geraniol, hydrocarbons such as nonadecene, heneicosane, heptadecane, octadecane and tricosane, sesquiterpene hydrocarbons such as α-guanine, humulene, γ-murolene, and others such as methyl eugenol, esters and aldehydes such as geranyl acetate and geranial, phenols such as eugenol are among the most important compounds in Turkish rose essential oil [13,15,20].

However, the processes of water distillation and solvent extraction significantly affect the yield and quality of essential oil and the extraction of active ingredients of rose. From this point of view, comparing the compositions of rose oil, rose water (hydrosol), and water remaining in the still with *R. damascena* petal compositions, which are combined with other headspace-solid phase microextraction

(HS-SPME) and solid phase microextraction (SPME) methods. Floral compositions are essential during the water distillation process and the testing of water-distilled rose products.

On the other hand, n-hexane, a solvent obtained from petrochemical sources, is the most widely used solvent for the production of extractive material (concrete) from rose petals. Can better and healthier alternatives to n-hexane be discovered among other organic hydrocarbons for solvent extraction? Based on this question, another aim of the study is to determine the efficiency of the extractive material (concrete and absolute) of some non-polar solvents such as cyclohexane, petroleum ether, diethyl ether and chloroform as a substitute for n-hexane and finally using analyzes GC-FID and GC-MS identified the aromatic compounds of the extracts.

According to the method presented in the European Pharmacopoeia, we put 1 kg of fresh rose flowers along with 3 liters of I in a 6 liter balloon and connect the balloon to a water cooling system.Essential oil and rose water (hydrosol) were separated into two upper and lower layers after passing through the cooling in the separator. The amount of rose essential oil measured as percentage (v/w) was 0.045% on average in triplicate analyses. After the water distillation method, the rose water (hydrosol) collected under the rose oil and the remaining water in the distillation flask are also separated [21].

Kukcuoglu and Baser in 2003 extracted rose flowers with high-purity non-polar solvents by a separatory funnel. Multi-stage extraction (3 times) by solvents was done in 30, 20 and 15 minutes consecutively in a funnel in order to increase the yield of the extract. After evaporation of the solvents from the extracted material by a rotary evaporator under vacuum at a temperature below 50 °C, rose concrete, a pale reddish, waxy and semi-solid material was obtained. The concretes were dissolved in 96% ethanol and vigorously stirred at 35-40 °C for 8 hours. The alcoholic solution was cooled at -20 °C for 8 hours in a deep freezer. Crystallized stearopetenes were filtered through filter paper under vacuum condition [15].Aydinli and Tutas 2003 redissolved the isolated stearoptenes remaining on filter paper in 50 mL of ethanol (purity 96%) to obtain liquid filtrates. To obtain an absolute result, the filter is placed overnight in a deep freezer and evaporated in vacuo using a rotary evaporator at 55 °C to remove ethanol and water. In absolute production, concretes were treated with approximately 10 times their respective weight of ethanol [14].

Apart from yield as % (v/w), the concrete and absolute efficiencies were also calculated. While concrete efficiency represents the number of fresh flowers for extracting 1 kg of rose concrete, absolute efficiency represents the amount of the concrete for extracting 1 kg of rose absolute. Solvent extraction analyses were performed in triplicate and the data were analyzed with the analysis of variance (ANOVA) using SAS (SAS, 1999). Means were separated by Duncan's Multiple Range Test ($P \leq 0.05$).

Baydar, *et al*., 2016, Put 2.5 g of fresh rose flower in a 10 ml vial, which is immediately closed with a silicone septum and sealed with a cap. After incubation for 30 minutes at 60 °C, the SPME fiber was pushed through the headspace of a sample vial to absorb volatiles and then directly into the injection port of a gas chromatograph coupled to a mass spectrometer (GC/MS) [22].

Another method of extracting rose essential oil is solid phase microextraction (SPME, Supelco, Germany) with a fiber precoated with a 75 μm-thick layer of Carboxen/Polydimethylsiloxane (CAR/PDMS). 2.5 g of fresh hand-picked flowers were put into a 10 mL vial, which was then immediately sealed with a silicone septum and a crimp cap. After incubation for 30 min at 60^{o}C, SPME fiber was pushed through the headspace of a sample vial to adsorb the volatiles, and then inserted directly into the injection port of the GC-MS.

In study, HS-SPME coupled with GC-MS system using carboxane / polydimethylsiloxane (CAR/PDMS) fiber was used to characterize volatile compounds emitted from pink roses. The HS-SPME method was obviously a faster sample preparation method compared to classical hydro-distillation (90 min × 300 min) and was performed with a much lower amount of mud than hydro-distillation (2.5 g/kg). In addition, no organic solvents are required. Through the use of HS-SPME to extract volatile compounds from rose flowers, it was possible to identify a total of 46 flower compounds, representing about 100% of the total composition by direct injection in GC-MS. Phenylethyl alcohol, citronellol, and geraniol were the main compounds of fresh rose petals (43.2%, 16.6%, and 10.3%, respectively).

Dobreva (monitored the daily dynamics of compounds in *R. damascena* flowers using the HS-SPME technique and identified 37 compounds; geraniol (0.6-32.3%), citronellol (1.2-30.9%), nerol (0.4-8.6%) and phenylethyl alcohol (1.3-13.4%) whose presence at the respective time was highly dependent on the moment of the day and abiotic factors (air temperature, relative humidity, intensity of sunlight and wind)

The yield of rose oil after hydro-distillation was on average 0.045% (v/w). According to the GC and GC/MS analysis, in rose oil, a total of 15 volatile compounds (calculated aspercentage of peak area of GC-FID analysis) were characterized by a high percentage of non-monoterpene alcohols. The ring was detected, especially shown. compounds of geraniol (35.4%), citronellol (31.6%), and nerol (15.3%), and long-chain hydrocarbons (alkanes) in particular such as nonadecane (7.2%), hexadecane (1.3%) and heneicosane (1.8%) show given. Although phenylethyl alcohol was the main component of the fresh floral scent, only 1.3% of it was found in hydro-distilled rose oil.High-quality rose essential oil is characterized by a (citronellol + nerol) to geraniol ratio that should be between 1.2 and 1.3 [11].

Babu, *et al*., 2002, reported ratio was 0.9 percentage. This ratio changes mainly with the fermentation of fresh flowers. If the flowers are picked late and distilled late in the day, they undergo fermentation. During fermentation, while the proportion of citronellol increases, the proportion of geraniol and nerol decreases [19, 20, 23]. Since unfermented fresh flowers were used as material in this study, the ratio of (citronellol + nerol) to geraniol was less than 1.2-1.3. Generally, the percentage of total alcohol (55.3–83.4%) in rose essential oil increased with increasing distillation pressure and temperature [23].

The percentage of hexadecane (1.3%), nonadecane (7.2%), and heneicosane (1.8%) in rose petal essence obtained by hydro-distillation method was higher than HS-SPME method in fresh rose flowers. The percentage of alkanes or steareptenes in rose oil increases and the percentage of monoterpene alcohols decreases with increasing distillation time with water and slides of the second fraction [20]. While fresh roses contained only 0.2 percent rose furan and cis-rose oxide, hydro-distilled products contained neither. Many other compounds in rose oil, such as β-damascenone and β-ionone, are present only in small amounts but are also very important for overall quality [24].

The compounds identified in the rose water and the remaining water in the still were different from the rose oil compounds. The amount of essential oil obtained from rose water (hydrosol) and water remaining in the pot was about 0.1-0.2%. The main compounds in rose water (hydrosol) and the remaining water in the pot were phenylethyl alcohol with 35.6% and 98.2%, respectively. Due to the solubility in water, the compounds of rose water and the remaining water in the distillation pot, the by-products of distillation with water, have higher amounts of phenylethyl alcohol compound than rose oil.Therefore, rose water probably exhibits a more natural rose fragrance than essential oil due to its very high phenylethyl alcohol content. Other compounds of rose essential oil are: geraniol (27.9%), nerol (12.7%), citronellol (8.3%) and eugenol (6.2%). Volatile combinations of rose water and residual water containing long chain hydrocarbons (stearopetenes) were absent except for ecosane, which was the highest alkane remaining in rose water and residual water at both 1.8%.

The amount of volatile compounds in rose water (hydrosols) depends on the solubility and specific weight of aromatic compounds. In normal production, rose water has very small amounts of essential oil (below 0.1%) and its main composition is phenylethyl alcohol [25]. It is important to mention that the main source of phenylethyl alcohol is 2-phenylethyl β-D-glucopyranoside, which accumulates in the rose oil at the time of harvesting and can be easily hydrolyzed during hydro-distillation with the juice of the petals inside the still [26]. Although rose oil has a strong antioxidant and antimicrobial effect [5,6, 27-29]. Effects similar to rose water are relatively low [28],

since rose water provides an ideal growth environment for bacteria, yeasts and fungi, the use of physical preservation methods such as pasteurization, and UV treatment or the addition of approved chemicals. After the production of rose water, it is necessary to ensure the health of consumers and the quality of rose water [17]. Methyl eugenol is a valuable aromatic chemical used in perfumes and cosmetics. However, due to negative and allergic side effects on human health, more than a certain concentration in essential oils is not desirable [30,31].

Rose oil is one of the essential oils containing methyl eugenol, the percentage of which can increase up to 5%, especially in rose oils distilled from fermented flowers and distilled with a lot of water or for a long time [20].

HS-SPME analysis of fresh flower and measurement and identification of compounds from hydro-distilled rose oil showed similar percentages of methyl eugenol of 0.9% and 0.8% by GC-FID, respectively. While the aromatic compounds of rose water had 1.23% of the residue. HS-SPME-GC-MS results were observed in fresh roses and GC-FID in distilled products.

The water was completely free of methyl eugenol compound. Concrete efficiency and absolute efficiency of different solvents for extraction were studied. The yield of concrete from *R. damascena* flower using n-hexane solvent of about 0.25% (1 kg from 400 kg of fresh flower) was reported in Turkey [15], Bulgaria [32] and Pakistan [33].

The absolute yield of rose petal concrete using ethyl alcohol extraction has been reported from 55% to 68% [15,17,32]. The efficiency of concrete is between 0.30 and 0.66% and the absolute efficiency is reported between 52.1 and 70.9%. While the highest yield of concrete (0.66 percent) and productivity (1 kg of 150.1 kg of fresh mud) for extraction from diethyl ether solvent, the highest absolute yield (70.9 percent) and productivity (1 kg of 249.7 kg of fresh mud) to was achieved Ethyl alcohol extraction from n-hexane and diethyl ether concretes were obtained respectively.

The difference between concrete and absolute productivity and productivity with petroleum ether, cyclohexane and chloroform solvents was not statistically significant ($P \leq 0.05$). These solvents produced higher concrete yields, but lower absolute yields compared to n-hexane extraction. Chloroform solvent ranked second after diethyl ether in absolute productivity with 450.5 kg of fresh roses required to produce 1 kg absolute.The volatile compounds of concrete are rich in monoterpenes and aromatic alcohols such as phenylethyl alcohol (16.6-23.3%), citronellol (3.8-5.4%), nerol (2.2-3.3%) and geraniol (5.0-7.4%) and chain hydrocarbons and long-chain hydrocarbons such as nonacosane (19.2-38.5%), hexadecane (6.1-17.3%), eicosane (0.0-13.1%) and nonadecane (0.0-11.8%). The number of volatile compounds in concrete samples changed from 11 (in diethyl ether concrete) and chloroform concrete to 15 (in n-hexane concrete).

Phenylethyl alcohol and nonacosane were the main compounds in all the concretes tested. The highest percentage of phenylethyl alcohol (23.3%) and the lowest percentage of citronellol, nerol and geraniol (3.8%, 2.2% and 0.5% respectively) were observed in the concrete extracted with diethyl ether solvent. On the other hand, concrete extraction with hexane solvent obtained the highest percentage of geraniol, citronellol and nerol (7.4, 5.4 and 3.3% respectively) and the lowest percentage of nonacosane (19.2%).

The solvents tested in this study were selective for some volatile compounds. For example, eicosane was only extracted by hexanes in similar percentages, while nonadecena was only extracted by ethers. Some hydrocarbons, e.g. Nonacosane and hexadecane were extracted with all the solvents used in the study. These results may be mainly related to the solubility, polarity and other chemical properties of the solvents.

The main compounds extracted with solvent from rose petals, including absolute from various solvents extracted from concrete, are: phenylethyl alcohol (35.2-38.4%), hexadecane (28.3-36.3%), geraniol (6.9-10.8%), citronellol (5.6-8.4%) and nerol (3.4-5.0%). The number of absolute compounds identified was between 10 and 11, which was less than the number of concrete compounds shown. Ayci *et al*., 2005 reported that absolute extraction of rose flower from concrete with n-hexane solvent contained 14 compounds, mainly phenylethyl alcohol, citronellol, geraniol, nerol, methyl eugenol, geranyl acetate, benzyl alcohol, nonadecane, nonadecene. and farnesol [16]. The substances obtained from absolute are similar to the volatile compounds of rose oil. However, the extraction with diethyl ether solvent from the absolute lower percentage of geraniol, citronellol and nerol compounds (6.9, 5.6 and 3.4%, respectively) than the others and a higher percentage of phenylethyl alcohol (8.8) 37%) gave absolute ether after petroleum solvent. Methyl eugenol was detected absolutely in the range of 0.4 to 0.5% with the exception of n-hexane concrete which had only 0.3%.

Another interesting finding, nonacosane, the major compound of rose concretes, was not detected in absolute rose. This compound is probably insoluble in ethyl alcohol and remains as a solid by-product. This view is confirmed by a study conducted by Ayci *et al*. 2005, who emphasized that the remaining solid hydrocarbon fractions are composed of long-chain, high-molecular-weight saturated hydrocarbons, including nonacosane [16].

In Iran, the cultivation and consumption of roses has a long history. Crude distillation of roses oil is believed to have originated in Iran in the late 7th century AD and spread to the provinces of the Ottoman Empire in the late 14th century. Iran was a major producer of rose oil until the 16th century and exported it to the whole world [2,9].

At present, in Iran, only one type of rose (*R. damascene* Mill.) is cultivated, and it is mostly produced by hydro-distillation, rose water and essential oil, and its waste is used to make jam, sugar flower, etc. All kinds of rose water are used as astringent, tonic, mild laxative and antibacterial. They have various uses in treatment such as: sore throat, enlarged tonsils, heart problems, eye diseases and gallstones, and they are also used as a cooling effect and as other medicines [34-37].

The amount of rose oil is very low due to the production method. The composition of essential oil is different in flower stages, flower parts and harvest period [9]. One kilogram of rose oil can be obtained from approximately 3000 kg of petals [12]. Flower of *R. damascene* Mill., only once a year and flowering lasts approximately 4-6 weeks [38]. The rose bushes produce a large number of blooming flower buds daily, which are hand-picked and distilled the same day. Due to the short flowering period and the large number of flowers, a significant amount of roses wait a long time until distillation. Depending on the waiting of the petals, not only the yield of the essential oil will decrease, but also its quality will decrease. Therefore, only fresh rose petals are preferred for essential oil production [39].

The chemical composition of rose oil, rose water, concrete and absolute has been investigated in Iran and abroad [14,16,25,26, 40- 42].

Lohani, *et al*., 2013, on the volatile compounds of rose (*R. damascene* Mill.) from Uttarakhand Himalayas, reported phenylethyl alcohol as the major compound, ranging from 77.35-89.37%, followed by α-citronell (0.95 -5.71%) is mentioned. along with other minor compounds such as geranial (0.07-2.49%), n-eicosane (0.08-0.20), n-heneicosane (0.08-1.07%), n-tricosane (0.01-0.09%), and n-tetracosane (0.04- 0.31%) [43]. Rose water compositions varied qualitatively and quantitatively among locations. Therefore, it was concluded that the climatic conditions of the hills of Uttarakhand are suitable for the production of high-quality rose water, especially from the Joshimath region, which was identified as an elite source among the investigated samples. The amount of phenylethyl alcohol compound was very low in rose oil due to its high polarity and as a result its solubility in water. Therefore, the maximum proportion of phenylethyl alcohol is dissolved in the aqueous phase/distilled water during the essential oil extraction process, i.e. distillation. For this reason, the difference is in the amount of fragrance and composition of the natural rose oil and rose water [40]. Due to the presence of a higher percentage of phenylethyl alcohol, the smell of rose water increases. It can be used as an ingredient in flavorings and perfumery, so we came to the conclusion that it can be used to produce rose water of excellent quality by examining different weather conditions.

In research conducted from 2003 to 2005 on species collected from all over Iran and cultivated in the Research Institute of Forests and Rangelands, the compounds of essential oil of the Damask rose flower were identified. A total of 40 Damask rose landraces were collected from 28 provinces of Iran. According to geographical similarity, the landraces were divided into 13 origin sites (Fig. 1) .These landraces were grown during 2000-2005 in the experimental field of the Research Institute of Forests and Rangelands (RIFR). Tehran, Iran (latitude 35° 44´ N. longitude 51° 10´ E. altitude 1320 m) using a randomized complete block design with three replications. The experimental plots were 2.5 m long 2 m wide and comprised three plants per plot.

Rezaei *et al*. 2003, reported in a project entitled "Comparison of laboratory and industrial samples of *R. damascene* Mill. in terms of quantity and quality of major compounds from Kashan region". Os1 (Isfahan) samples were collected in the laboratory of the Medicinal Plants Research Department of the Forests and Rangelands Research Institute and extracted by water distillation.

Fig. 1 The origin sites of Damask rose landraces (Os1 – Os13) on the map of Iran.

Os1 (Isfahan), Os2 (East & Weast Azarbayjan- Ardabil), Os3 (Kermanshah – Eilam), Os4 (Tehran- Markazi), Os5 (Chaharmahal -Kohkulooie – Lorestan), Os6 (Razavi Khorasan -South Khorasan), Os7 (Khozestan – Hormozgan- Sistan), Os8 (Zanjan – Qazvin), Os9 (Semnan -Qom), Os10 (Pars -Kerman), Os11 (Kurdistan -Hamadan), Os12 (Guilan -Mazandaran- Gulestan), Os13 (Yazd).

The extracted compounds were analyzed by gas chromatography (GC) and gas chromatography-mass spectrometry (GC/MS) devices. The amount of phenyl ethyl alcohol were (18.9%) [44]. Rezaei *et al*. in 2003, reported a project entitled "Study of *R. damascene* Mill essential oil. Central and northwestern regions of the country" were presented. Samples of Kashan, Ghamsar with code Os1 (Isfahan) and samples of Azerbaijan with code Os2 (Esco) were extracted by water distillation method after collection in the laboratory of Medicinal Plants Research Department in Forests and Rangelands Research Institute. The extracted compounds were analyzed by gas chromatography (GC) and gas chromatography-mass spectrometry (GC/MS) devices. The amount of phenyl ethyl alcohol were (7.1%). For the samples of Azerbaijan (Osko) the main compounds were phenyl ethyl alcohol (9.4%), [44]. An appropriate way therefore genotype identified with a high percentage of major essential components, from Osko, despite cultivation in a different envirment from its geographical origin. Jaimand *et al*. in 2005, reported in a project entitled "Comparison of the quantity and quality of *R. damascene* Mill essential oil. From different designs of water distillation apparatus" has been presented. Samples of Kashan, Ghamsar with code Os1 (Isfahan) were collected by distillation with water after collection in the laboratory of Medicinal Plants Research Department in the Forests and Rangelands Research Institute of the country. The extracted compounds were analyzed by gas chromatography (GC) and gas chromatography-mass spectrometry (GC / MS) devices. One sample was made with a water distillation method but with different device designs. Different apparatuses like Clevenger (0.6%), MaQuell (2.3%), the Analytical official Chemistry Association (AOAC), and two designs proposed by the author called Designs 1 and 2, were not detected any phenyl alcohol [44]. Jaimand *et al*. In 2004, reported a project entitled "Study of *R. damascene* Mill essential oil. Different regions of Isfahan province" has been presented. Samples of Kashan, Ghamsar with code Os1 (Isfahan) were collected by distillation with water after collection in the laboratory of Medicinal Plants Research Department in the Forests and Rangelands Research Institute of the country. The extracted compounds were analyzed by gas chromatography (GC) and gas chromatography - mass spectrometry (GC / MS) devices. Four samples of camouflage from Kamoo village containing phenyl ethyl alcohol from 5.8% , 8.8%, 18.9% and to 30.2%. one sample from Qahroud contains phenyl ethyl alcohol (15.2%), from Ghamsar two samples contain phenyl ethyl alcohol (7.1%) and also (17.5%) and from Mashhad Ardhal two samples and phenyl alcohol (4.5%) And (10.3%) were obtained [46].

Phenethyl alcohol, or 2-phenylethanol, is the organic compound that consists of a phenethyl group ($C_6H_5CH_2CH_2$) group attached to OH. It is a colourless liquid that is slightly soluble in water (2 ml/100 ml H_2O), but miscible with most organic solvents. It occurs widely in nature, being found in a variety of essential oils. It has a pleasant floral odor. Properties: Chemical formula ($C_8H_{10}O$), Molar mass (122.16 g/mol), Odor (Soft, like roses), Density (1.017 g/cm^3), Melting point [(−27 °C (−17 °F; 246 K)], Boiling point [(219 to 221 °C (426 to 430 °F; 492 to 494 K)]. Soluble in (Alcohol, Water, dipropylene glycol, fixed oils, kerosene, mineral oil, slightly propylene glycol), Insoluble in (paraffin oil), Stability in (alcoholic lotion, antiperspirant, bleach, deo stick, detergent perborate, fabric softener, hair spray, hard surface cleaner, liquid detergent, non-discoloring in most media, shampoo).

Phenethyl alcohol is found in extract of rose, carnation, hyacinth, Aleppo pine, orange blossom, ylang-ylang, geranium, neroli, and champaca. It is also an autoantibiotic produced by the fungus *Candida albicans.*

It is therefore a common ingredient in flavors and perfumery, particularly when the odor of rose is desired. It is used as an additive in cigarettes. It is also used as a preservative in soaps due to its stability in basic conditions. It is of interest due to its antimicrobial properties. phenyl ethyl alcohol was the principal component of fragrant rose flowers among the 13 rose varieties. When this phenyl ethyl alcohol is so distilled, it is added back to the original distillate, in the correct proportion, to form a complete and whole rose oil, and is then called Rose Otto.

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It is therefore a common ingredient in flavors and perfumery, particularly when the odor of rose is desired. It is used as an additive in cigarettes. It is also used as a preservative in soaps due to its stability in basic conditions. It is of interest due to its antimicrobial properties.

Soluble in (Alcohol, Water, dipropylene glycol, fixed oils, kerosene, mineral oil, slightly propylene glycol.), Insoluble in (paraffin oil), Stability (alcoholic lotion, antiperspirant, bleach, deo stick, detergent perborate, fabric softener, hair spray, hard surface cleaner, liquid detergent, non-discoloring in most media, shampoo).

In a research conducted in 2021, after Extraction essential oil and rose water, it was conducted on the remaining water in the still. The water inside the distillation pot was extracted with eight different solvents (chloroform, cyclohexane, dichloromethane, diethyl ether, ethyl acetate, hexane, petroleum ether and toluene) by a separatory funnel. Then, the extracted materials were measured and identified by (GC) and (GC/MS). The amount of phenylethyl alcohol obtained by different solvents in the form of chloroform 96.1%, cyclohexane 46.3%, dichloromethane 96.1%, diethyl ether 46.3%, ethyl acetate 89.6%, hexane 100%, petroleum Ether was 0.42% and toluene was 74%. Phenylethyl alcohol compound can be separated from the water in the still and used.

CONCLUSION

There are numerous methods for isolating floral compounds from flowers containig aromatic components. For industrial production of aromatic oils and other extracts from the roses, the common methods are hydro-distillation with cohobation and extraction with organic solvents [47]. The main target in the distillation and extraction processes is to produce the aromatic oils and extracts which have compounds close to genuine scent compounds secreted from the rose flowers. For this reason, it is important to identify the scent compounds not only in the products but also in the flowers. The use of headspace solid phase microextraction (HS-SPME) is shown to be a convenient and effective analytical tool for the sampling of floral compounds of oil-bearing rose by Dobreva [23] and Jirovetz [48].

In our study, while a total of 46 floral compounds of the fresh rose flower were identified by SPME-GC-MS analysis, the aromatic products from hydro-distillation and solvent extractions contained between 10 and 15 compounds detected by GC-FID/GC-MS analysis. These results show that both hydro-distillation and solvent extraction processes significantly change the natural scent composition or chemical profile of the oil-bearing rose. For example, while phenylethyl alcohol was one of the main floral scent compounds in the fresh rose flower (>40%), hydro-distilled rose oil contained a very small amount of it (<1.5%), which explain why the smell of rose oil does not resemble the genuine odour of rose flower.

Some of the compounds extracted from the rose flowers undergo denaturing or chemical breakdown mainly due to the high temperatures during hydro-distillation. Moreover some valuable floral compounds such as phenylethyl alcohol remain in the rose water and residual water by-products during the hydro-distillation process. While there is a commercial value of rose water, residue water does not. However, residue water can be economically utilized as a source of phenylethyl alcohol which is used in cosmetic industry as ingredient in perfume and other formulations because of its popular rose-like smell.

For the perfumery industry, the production of concrete and absolute is done by low temperature solvent extraction method. Since essential oil compounds are usually non-polar molecules, they can be largely extracted by dissolving in a non-polar solvent. The most common non-polar solvent used is hexane. As a result of this study, it seems to be the most feasible alternative solvent for hexane, diethyl ether due to its higher concrete performance and absolute productivity, as well as its higher phenylethyl alcohol content. However, diethyl ether is highly flammable and may be explosive under standard operating procedures.

Considering the adverse effects on human health and the environment, healthier and more productive alternatives for hydrocarbon solvents should be explored for extraction. Today, supercritical CO2 has become an important commercial and industrial solvent due to its high purity, low toxicity, and non-flammable property compared to traditional solvents [49].

In a summary, it is necessary to improve the most efficient distillation and extraction methods that capture the majority of flower compounds without significant changes in their chemical and molecular structure. Findings from this and similar studies may provide an important scientific contribution to these requirements.

Regarding the composition of phenylethyl alcohol, in fact, this composition remains in the still after extracting essential oil and rose water. As you can see in table No. 6, it is possible to separate phenyl ethyl alcohol by different solvents: hexane (100%), chloroform (96.1%), dichloromethane (96.1%) and ethyl acetate (89.6%) from the water in the pot after distillation and used.

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REFERENCES

1. Gudin S. Rose genetics and breeding, Plant Breed. Rev. 200;17:159-189.
2. Rusanov K., Kovacheva N., Vosman B., Zhang L., Rajapakse S., Atanassov A., Atanassov I. Microsateilite analysis of *Rosa damascene* Mill. Accessions reveals genetic similarity between genotypes used for rose oil production and oil Damask rose varities. Theor. Appl. Genet. 2005;111: 804-809.
3. Mahmood N., Piacente S., Pizza C., Burke A., Khan A., Hay A.J. The Anti-HIV Activity and Mechanisms of Action of Pure Compounds Isolated from *Rosa damascena*. Biochem Biophys Res Communication. 1996;229:73- 79.
4. Achuthan CR, Babu BH, and Padikkala J. Antioxidant and Hepatoprotective Effects of *Rosa damascena*. Pharmaceu. Biol. 2003;41:357- 361.
5. Basim E., and Basim H., Antibacterial Activity of *Rosa damascena* Essential Oil. Fitoterapia 2003;74:394-396.
6. Özkan G, Sağdic O, Baydar NG, and Baydar H, Antioxidant and Antibacterial Activities of *Rosa damascena* Flower Extracts. Food Sci Technol Intl. 2004;10:277-281.
7. Baydar H., Baydar N.G. The Effects of Harvest Date, Fermentation Duration and Tween 20 Treatment on Essential Oil Content and Composition of Industrial Oil Rose (*Rosa damascena* Mill.). Ind Crops Prod. 2005;21:251-255.
8. Verma R.S., Padalia R.C., Chauhan A., Singh A., Yadav A.K. Volatile constituents of essential oil and rose water of damask rose (*Rosa damascena* Mill.) cultivars from North Indian hills. Natural Product Res. 2011;25(17), 1577-1584.
9. Guenther E. Oil of rose. In: The essential oils. Vol 5. Kreiger, kregen publishing company Malabar, Florida, USA. 1952; 3-48.
10. Erbaş S., Baydar H. Variation in Scent Compounds of Oil-Bearing Rose (*Rosa damascena* Mill.) Produced by Headspace Solid Phase Microextraction, Hydrodistillation and Solvent Extraction. Rec Nat Prod. 2016;10:5,555-565
11. Baser KH.C. Turkish rose oil. Perfumer & Flavorist. 1992;17, 45-52.
12. Bayrak A., Akgul A. Volatile oil composition of Turkish rose (*Rosa damascene* Mill.). Sci. Food Agr J. 1994;64:441-448.
13. Anac O. Gas chromatographic analysis on Turkish rose oil, absolute and concrete, P. & F.1984;1-14.
14. Aydinli M. Tutas M., Production of rose absolute from rose concrete. Flavour Frag. J., 2003;18: 26-31.
15. Kukcuoglu M., Baser KH.C. Studies on Turkish rose concrete, absolute and hydrosol. Chem Nat Compd. 2003; 39:457-464.
16. Ayci F., Aydinli M., Bozdemir O.A., Tutas M. Gas Chromatographic investigation of rose concrete, absolute and solid residue. Flavour Frag. J. 2005; 20:481-486.
17. Baydar N.G., Baydar H. Phenolic compounds, antiradical activity and antioxidant capacity of oil-bearing rose (*Rosa damascena* Mill.) extracts, Ind. Crop. Prod. 2013;41:75-380.
18. Dudareva N., Pichersky E. Biochemical and molecular genetic aspects of floral scents. Plant Physiol. 2000;122: 627-633.
19. Kovacheva N., Rusanov K., Atanassov L. Industrial cultivation of oil bearing rose and rose oil production in

Bulgaria during 21st century, directions and challenges, Biotechnol. & Biotechnol. Eq. 2010;24: 1793-1789.

20. Baydar H., Schulz H., Kruger H., Erbas S., Kineci S. Influences of fermentation time, hydro- distillation time and fractions on essential oil composition of Damask Rose (*Rosa damascene* Mill.), J Essen Oil Bear Plant. 2008; 11: 224-232.
21. European Pharmacopoeia, Maissonneuve Sainte Ruffine. 1975; 3:68.
22. Baydar H., Erbas S., Kazaz S. Variations in floral characteristics and scent composition and the breeding potential in seed-derived oil-bearing roses (*Rosa damascena* Mill.), Turky J Agric. 2016; 40: 560-569, doi:10.3906/tar-1512-57
23. Dobreva A. Dynamics of the headspace chemical components of *Rosa damascena* Mill. Flowers J Essent Oil Bear Pl. 2013;16, 404-411.
24. Babu K.G.D., Singh B., Joshi V.P., Singh V. Essential oil composition of Damask rose (*Rosa damascena* Mill.) distilled under different pressures and temperatures. Flavour Frag J. 2002;17: 136-140.
25. David F., De Clercq C., Sandra P. GC/MS/MS analysis of β-damascenone in rose oil. Varian GC/MS App, Note 52. 2006.
26. Agarwal S.G., Gupta A., Kapahi B.K., Thappa R.K., Suri O.P. Chemical composition of rose water volatiles. J. Essent.Oil Res. 2005;17:265-267.
27. Eikani M.H., Golmohammad F., Rowshanzamir S., Mirza M. Recovery of water-soluble constituents of rose oil using simultaneous distillation–extraction. Flavour Frag J. 2005;20: 555-558.
28. Aridogan B.C., Baydar H., Kaya S., Demirci M., Ozbasar D., Mumcu E. Antimicrobial activity and chemical composition of some essential oils. Arch Pharmacal Res. 2002;6: 860-864.
29. Ulusoy S., Tinaz G., Seçilmis-Canbay H. Tocopherol, carotene, phenolic contents and antibacterial properties of rose essential oil, hydrosol and absolute. Curr Microbiol. 2009;59: 554-558.
30. Anonymous. 2005. The Bulgarian Rose Co. /www.bulgarskarosa.com
31. Harris B. Methyl eugenol – the current bête noire of aromatherapy. Int J Aromather. 2005;12: 193-201.
32. Garnero J., Buil P. Evolution of the composition of the rose essential oils and concrete during the production campaign. Riv. Ital Ess Prof Peante Offic Aromic Sap, Aerosol. 1976;58: 537-540.
33. Khan A.S., Rehman S.U. Extraction and analysis of essential oil of *Rosa* species. I.J.A.B.E. 2005;7:973-974.
34. Anonis D.P. Rose in perfumery and cosmetics. Drug and Cosmetics Industry. 1982;130:63-64.
35. Hunt S.R. The rose in pharmacy. Pharmaceutical J. 1962;189: 589-591.
36. Kaul V.K. Supplement to cultivation and utilization of aromatic plants Jammu: Regional Res Laboratory. 1998.
37. Schweisheimer W. Roses in manufacture of perfumes. Perfumes Cosmetics and Savons. 1961; 4: 62–65.
38. Kazaz S., Erbaş S., Baydar H. The effects of storage temperature and duration on essen- tial oil content and composition oil rose (*Rosa Damascena* Mill.) Turk. J Field Crops. 2009;14:89–96.
39. Baydar H., Göktürk-Baydar N. The effects of harvest date, fermentation duration and Tween 20 treatment on essential oil content and composition of industrial oil rose (*Rosa damascena* Mill.). Ind Crop Prod. 2005;21:251–255.
40. Shawl A.S., Adams R. Rose oil in Kashmiri India. Perfumer & Flavorist. 2009;34: 2-5.
41. Gupta R., Mallavarapu G.R., Ramesh S., Kumar S. Composition of rose essential oils of *Rosa damascena* and *Rosa indica* grown in Lucknow. J Medicinal and Aromatic Plant Sci. 2000;22: 9–12.
42. Lawrence B.M. Essential oils. Carol Stream, IL: Allured Business Media USA.2003.
43. Lohani H., Andola H.C., Chauhan N.K., Gwari G., Bhandari U. Volatile constituents of rose water of Damask rose (*Rosa damascene* Mill.) from Uttarakhand Himalayas. Medicinal Plants. 2013;5: 1-3.
44. Rezaee M.B., Jaimand K., Tabaei-Aghdaei S.R., Brazandeh M.M. Comparative study of laboratory and industrial essential oils samples of *Rosa damascena* Mill. For quantitative and qualitative constituents from Kashan Iranian J Med & Aroma Plants Res. 2003; 19 :63-72.
45. Jaimand K., Rezaee M.B., Assareh M.H., Brazandeh M.M. Comparison of Quantity and Quality of the Essential Oil of *Rosa damascena* Mill. by Different Apparatus of Hydrodistillation. Iranian J Med & Aroma Plants Res. 2005; 21:283-292.
46. Jaimand K., Rezaee M.B., Tabaei-Aghdaei S.R., Brazandeh M.M. Comparative study of essential oils of *Rosa damascena* Mill. from Isfahan province. Pajouhesh & Sazandegi. 2004;65:86-91.
47. Collin H.A. Extraction and Industrial Processes. In: Encyclopedia of Rose Science. (eds. A. Roberts, T. Debener, and S. Gudin), Elsevier Ltd. Academic Press. 2003;726-735.
48. Jirovetz L., Buchbauer G., Stoyanova A., Balinova A., Guangjiun Z., Xihan M. Solid phase microextraction/gas chromatographic and factory analysis of the scent and fixative properties of the essential oil of *Rosa damascena* Mill. from China. Flavour Frag J. 2005;20:7-12.
49. Karale C.K., Dere P.J., Honde B.S., Kothule S., Kote A.P. An overview on supercritical fluid extraction for herbal drugs. Pharmacology online. 2011;2:575-596.