

Essential Oil Composition of Rose Flowers from Karangpring Village Jember District Extracted by Distillation and Enfleurage

Komposisi Minyak Atsiri Bunga Mawar dari Desa Karangpring Kabupaten Jember yang diekstrak melalui Distilasi dan Enfleurasi

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ABSTRACT

Karangpring is one tourist destination villages in Jember district which is popular with beautiful large rose field areas. Therefore, this area grows to be a leading producer of rose flowers in Jember district. However, the bulky presence of these flowers made its price becomes lower in regular days. Local community only uses and sells these fresh flowers as the flower for funeral. The rose flower has a great potency to be explored as a source of rose essential oil production. To date, there is no previous research on studying rose flowers from Karangpring village for its potency on the essential oil production. In this research, rose flowers were subjected to be extracted of its essential oil using two extraction methods, distillation, and enfleurage. Hydrodistillation resulted two phases of distillates, above part formed a cloudy white phase as a normal essential oil extracted from plants, and the lower phase was an aqueous phase containing rose hydrosols. Both phases of these condensates were analyzed using GCMS. Data explained that above phase, with a yield oil of 0.07%, only contains long-chain hydrocarbons such as *n*-nonadecane, *n*-heptadecane, 9-nonadecene, and eicosane, while the lower phase only contains 2-phenylethyl alcohol. On the other hand, enfleurage of fresh rose flowers resulted in 0.06% oil yield. GCMS analysis of this oil shows that 2-phenylethyl alcohol, eugenol, and phenylacetic acid are three major compounds which take more than 85% of total rose absolute. The results show that enfleurage is a better method for extracting rose oil in better quality than using the distillation method, in term of the variety of volatile components. Meanwhile, hydrodistillation is still benefiting from producing rose water that is qualified as an industrial additive agent for food and cosmetic productions or even a new potent of agromedicine products.

Keywords: rose, rose oil, rose water, rose absolute, distillation, enfleurage.

INTRODUCTION

Rose flower has an economic and social value in the world, especially the Indonesian community. Some communities regularly use the flower as cut roses for many special events, such as birthday and wedding parties, also for funeral and cemetery flowers. Karangpring village is a leading producer and supplier for rose flowers in Jember district (Derap Desa, 2010; Arifin, 2017b). Local communities in Karangpring village frequently use rose flowers only as formal flowers for cemetery and funeral aim. Some also produce healthy labeled juice from rose flowers, or even as rose candies, teas, and crackers (Arifin, 2017a). Others also use rose water in cooking as a natural coloring agent (Saati *et al.* 2011), flavoring agents on desserts and many meat dishes (Karizaki *et al.* 2016; Alalwan *et al.* 2017). However, another valuable product

from rose flowers which attracts the attention of the world is the production of rose oil, known as rose *otto*, in flavor and fragrance industry (Kovats, 1987). Since rose oil is one of the most expensive essential oil in the world market today, we proposed the way on the diversification product of rose flower as essential oil production in this research (Agaoglu, 2000; Kovacheva *et al.* 2010).

The main flavor component on the scent of rose flowers are β -damascone and β -damascenone (Pal, 2013). Nevertheless, it is also known that characteristic odor of rose flowers came from 2-phenylethyl alcohol which being a principal component in rose water (Watanabe *et al.* 2001; Sakai *et al.* 2007; Sakai *et al.* 2008; Kumar *et al.* 2014; Gunes, 2005; Hirata *et al.* 2016). Previous researches explained that bioactive molecule of 2-phenylethyl alcohol inside rose flowers is in

the form of 2-phenylethyl β -D-glucopyranoside (Eikani *et al.* 2005; Watanabe *et al.* 2002). However, most publications only mention the finding on aglycon part of it, *i.e.* 2-phenylethylalcohol. It was assumed that instead of the presence of β -glucosidase inside the plant (Watanabe *et al.* 2001; Sakai *et al.* 2008), applying various pretreatment and extraction methods could hydrolyze the glycoside as well to produce 2-phenylethyl alcohol.

Some efforts have been made to extract rose oil with higher quality, meaning that it has a high content of rose alcohol, but low or even free of ester, higher alkanes, and fat (Eikani *et al.* 2005). Various methods to extract rose oil are distillation (Najem *et al.* 2011; Hosni *et al.* 2010; Agarwal *et al.* 2005), solvent extraction (Rusanov *et al.* 2011; Watanabe *et al.* 2001; Kurkeoglu and Baser, 2003), enfleurage (Yulianingsih *et al.* 2007), and supercritical CO₂ (Reverchon *et al.* 1997). Kumar *et al.* (2016) have applied distillation of Damask rose flowers with saline water containing 75 g NaCl/kg of rose flowers to increase about 42% higher of rose oil content compared to distillation in water only. But there is a special consideration on applying this method in a significant scale process due to the adverse effect of salt on damaging steel apparatus in a longer period.

Most of all, the use of hydrodistillation in kinds of literature is quite widespread for extracting rose oil from fresh rose flowers. It has been known that hydrodistillation product market value is higher than other product from different extraction methods, by the fact that the total number of essential oil components in hydrodistillation is over (Pal 2013). Research by Karmous *et al.* (2004) mentioned that hydrodistillation has a drawback that the effect of boiling floral material and water at the same time could lead a light burned odor in the final product of essential oil. Since hydrodistillation is also quite easy and inexpensive, then this method became economical for essential oil production and suitable to be applied to the local community as a kind of home industry.

Enfleurage also becomes a popular extraction method to produce absolutely because it applies a low temperature for extraction. Hence, enfleurage prevents the hydrolysis of essential oil component during extraction that can be occurred at high temperature process, and creates an unpleasant odour because of the presence of acetic acid,

dimethyl ketone, acetaldehyde, sulfides, ammonia and methanol (Soe'uib *et al.* 2016; Muchtar *et al.* 2013; Yulianingsih *et al.* 2007; Oktavianawati *et al.* 2017).

Various rose extracts depending on the way they were extracted, have their names. Rose oil is normally obtained from the upper phase of distillate from steam, steam-water, and hydrodistillation methods. Rose concrete is obtained by solvent extraction of rose flowers and has a smooth like semi-solid performance, while rose absolute is derived from rose flower extraction using ethanol as solvent. Rose water is collected from the lower phase of distillate which also contains a valuable oxygenated compound which is responsible for the great odor of rose flowers (Ulusoy *et al.* 2009).

To date, there was no previous investigation on exploring essential oil from rose flowers in Jember district. Then, this current paper will discuss the potency of rose essential oil production and compare the oil quality, in terms of GCM analysis data, because of applying different extraction methods.

METHODS

Materials

Fresh rose flowers were obtained from a rose farmer community, Sumber Mawar, in Karangpring village, Jember district, from May to July 2018. Rose plants that were used in this research has been identified by Lembaga Ilmu Pengetahuan Indonesia (Indonesian Institute of Sciences) Balai Konservasi Tumbuhan Kebun Raya Purwodadi as *Rosa hybrida* Hort.

Extraction Procedures

Hydrodistillation

The whole flower of roses was used in new conditions and extracted by hydrodistillation method using a Clevenger apparatus for 4 hours. The condensate formed two phases of distillate. The upper phase was collected as rose oil, while the lower phase was rose water. Rose oil was subjected to further analysis using GCMS. Rose water was extracted using diethyl ether to obtain rose concrete and then was run for GCMS analysis as well.

Enfleurage

Chasis (50x50x7) was smeared with 500 grams of shortening (white butter). 200 grams of fresh rose flowers were placed on the top of fat from shortening for five days. The flowers were changed in every 24 hours and substituted with the new fresh one in a similar weight. After five days, the fat containing essential oil was extracted using alcohol in a ratio of 2:1. Then the mixture was kept in 0-5°C during 24 hours. Filtration of this mixture results in pomade and extract. Extract was evaporated for its

alcohol content which then produced a rose absolute. Rose absolute was also analyzed for its volatile content using GCMS.

Identification of Volatile Components

Volatile compounds of the essential oil extracts were analyzed by GCMS-QP2010S Shimadzu with Rtx 5 MS column (length 30 m; ID 0.25 m; film 0.25 um), Helium gas as a carrier, EI 70 Ev. Column oven temp.70°C, injector temp. 300°C, pressure 12 kPa, total flow 27.3 mL/min, column flow 0.49 mL/min, ion source temp. 250°C, interface temp.305°C, solvent cut time 3 min, detector gain +0.00 kV. MS table, start time 3.20 min, end time 70 min, scan speed 1250, event time 0.50 sec. The constituents of the oils were identified based on their chromatogram and by comparing their mass spectral data with those from Wiley mass spectral database.

RESULTS AND DISCUSSION

Hydrodistillation is a kind of distillation method that boils the sample together with water at the same time. Water molecules will diffuse into plant tissues and carry out essential oil from oil glands to evaporate and condense. The oil will naturally be separated from the aqueous phase in the distillate. In this current research, the expected rose oil from hydrodistillation method was presented in the above phase of distillate and was separated from rose water to yield 0.07% oil. The performance of this rose oil was in semisolid form with a white transparent waxy look when it was cooled at 0-5°C or even at room temperature. However, the odor of this rose oil does not represent the fragrance of rose flowers.

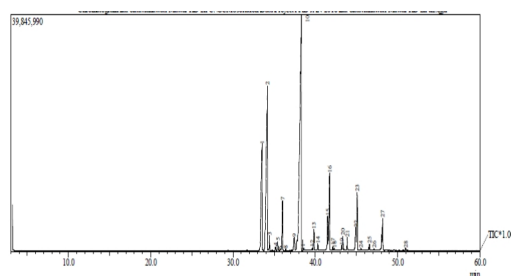


Figure 1. A chromatogram of rose oil from hydrodistillation method

Chromatogram of this rose oil can be seen in Figure 1, while the name list of those volatile compounds can be seen Table 1. GCMS analysis showed that this rose oil only contains saturated and unsaturated aliphatic long chain hydrocarbon compounds, which is

also known as stereoptanes, including *n*-nonadecane, *n*-heptadecane, 9-nonadecene, and eicosane. In this term, the form of rose oil as a wax-like material is possibly originating from the protective coatings of flowers which then makes the fat hardens and solidifies at room temperature (Sterrett 1962). The presence of this stearopten guarantees the durability of the scent (Dobrevva *et al.* 2013).

Table 1. Chemical components in rose oil from Karangpring rose flowers

No	Retention time	Compound Names	%
1	33.509	9-nonadecene	11.64
2	34.167	<i>n</i> -heptadecane	26.52
3	34.419	7-hexadecyne	1.00
4	35.426	1-octadecene	0.59
5	35.708	<i>Z</i> -7-hexadecene	0.09
6	37.427	10-heneicosene	1.59
7	38.292	<i>n</i> -nonadecane	38.52
8	38.476	trans-1,4-dihydroxycyclohexane	0.23
9	41.502	<i>E</i> -5-octadecene	4.43
10	41.735	eicosane	10.81
11	43.877	stearaldehyde	1.40
12	45.546	tridecanaldehyde	0.08
13	48.178	<i>n</i> -hexatriacontane	2.84
14	50.982	octadecyl chloride	0.16

This result research was relevant with the result of Moein *et al.* (2010) mentioning that *Rosa damascena* Mill. from South of Iran contains nonadecane, heneicosane, docosane and 9-nonadecene in a total amount of 92.06% of the rose oil, while the rest comprises citronellol and germacrene-D. Our result showed a relative big different on the composition of rose essential oil from other research that also used hydrodistillation to extract rose oil. Babu *et al.* (2002) confirmed that damask rose oil from the western Himalayas contains rose alcohols (such as citronellol, nerol, and geraniol) up to 68.13% and long-chain hydrocarbons including nonadecane, heptadecane, 9-eicosene, and docosane for about 21.23% of the total oil. Dobrevva *et al.* (2013) also enlighten the major volatile contents of three genotypes of oil-bearing roses in Kazanluk are mostly consist of citronellol, geraniol, nerol and nonadecane. Phenylethyl alcohol, as a marker compound of rose flower, is also obtained from this research though, in trace quantity, 0.26-1.31% of total rose oil since it is a polar compound and more soluble in water.

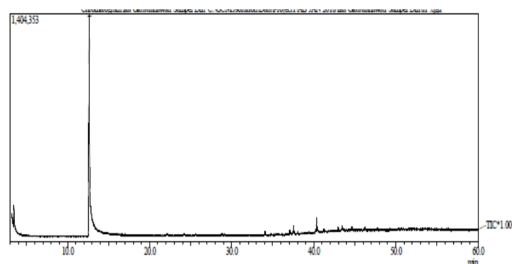


Figure 2. A chromatogram of rose water extracted with diethyl ether

It has been discussed before that upper phase of distillate, rose oil, contains only aliphatic hydrocarbons, while the lower phase only contains one polar and soluble compound in water, 2-phenylethyl alcohol (see chromatogram in Figure 2). This marker compound of rose oil was obtained when the lower distillate, as we know as a hydrosol, was extracted using diethyl ether. Evaporation of diethyl ether resulted in a clear colorless liquid form of rose concrete. The odor of this extract represents the right fragrance of rose flowers with a pleasant odour. In our research, we do not find any other volatile compounds that could be extracted from this rose water, like other research which are conducted by Kurkeuoglu & Baser (2003), Eikani *et al* (2005), Najem *et al* (2011), Agarwal *et al* (2003) and Babu *et al* (2002), using various solvents of n-hexane, n-butylacetate, pentane, and dichloromethane, respectively, to isolate citronellol, geraniol, nerol, linalool and eugenol, instead of obtaining 2-phenylethyl alcohol.

This current research confirmed that hydrodistillation method is not a suitable method to extract the true rose oil since it does not contain any marker compound of rose flower, *i.e.* 2-phenylethyl alcohol, or even any other highly polar and water-soluble compound, while the rose water contains. Thus, the distillation method could be chosen as the right method to produce valuable rose water or hydrosol because of a big content of 2-phenylethyl alcohol. This rose hydrosol was proven to be a safe material or additive in aromatherapy, cosmetics, and flavoring agents since it did not contain any lipophilic compounds that could interact and react with body tissues in topical application method (Adams *et al.* 2005).

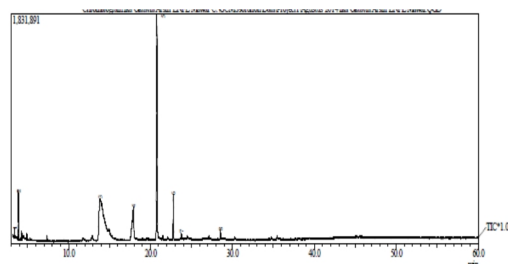


Figure 3. A chromatogram of rose absolute from five days enfleurage of fresh rose flowers.

Enfleurage method is chosen as an alternative method to extract essential oil from fresh rose flowers instead of hydrodistillation. Enfleurage applies cold fat as an adsorbent of volatile compounds deliberated by flowers. This research used a white butter or shortening as an adsorbent.

Table 2. Chemical components of rose absolute from enfleurage of fresh rose flowers

No.	Retention time	Compound Names	%
1	3.448	2-propenoic acid	0.96
2	3.889	glycolamide	3.56
3	13.857	Phenylethyl alcohol	40.48
4	17.948	phenylacetic acid	12.91
5	20.789	eugenol	35.16
6	22.793	β -caryophyllene	5.21
7	23.778	β -selinene	0.63
8	28.541	9-heptadecanone	1.09

Enfleurage of fresh rose flowers for five days resulted in a 0.06% oil yield with 'a light yellow liquid majorly containing 2-phenylethyl alcohol, eugenol and phenylacetic acid (Figure 3 and Table 2). Research of Yulianingsih *et al.* (2007) shows that seven days enfleurage on similar adsorbent, *i.e.* white butter, of Middle Java red roses brings about 0.239% of rose absolute containing 2-phenylethyl alcohol, citronellol, geraniol, nerol and also eugenol. Although that we did not obtain any citronellol, geraniol and nerol in our rose oil, we found that it also contains phenylacetic acid. Although Watanabe *et al.* (2002) have explained that one way of biogenesis of 2-phenylethyl alcohol (route c) was from phenylacetic acid intermediate the presence of phenylacetic acid in this research result was mostly predicted to become from the oxidation of 2-phenylethyl alcohol during enfleurage process. The

presence of natural oxidators such as rose oxides and acids in enfleurage and defleurage process could influence the stability of 2-phenylethyl alcohol. Moreover, possibility of the presence of microbes inside chassis and the fat, such as *Acetobacter acetic*, will oxidize 2-phenylethyl alcohol into corresponding acid and aldehyde (Gandolfi *et al.* 2004).

Nevertheless, the rose absolute performance from enfleurage is quite impressive with a light yellow color oil and pleasant odor effect. There was no fat left from adsorbent to be the presence in the rose absolute, similar to another enfleurage we have done previously for *Cananga odorata* (Oktavianawati *et al.* 2017). This enfleurage also has been applied practically on producing rose absolute to local community in Karangpring village, where those rose flowers came from. Moreover, enfleurage pomade or white butter residue has been extended to be the main material of soap making as well (Oktavianawati *et al.* 2018).

The use of hydrodistillation will result in a big problem on the extraction of the valuable oxygenated compound, 2-phenylethyl alcohol, since it can pass into solution in rose water for over than two-thirds of the oil, while the rose oil only contains less than one-third (Eikani *et al.* 2005). While the use of enfleurage on extracting high quality of rose absolute can be performed successfully. However, enfleurage and defleurage processes require a big effort on the preparation, extraction and purification of fragrance components from the adsorbent.

Overall, hydro distillation and enfleurage are compatible methods to obtain valuable fragrance compounds depend on our need, in the form of rose water or absolute. However, in order to be applied as home industry of rose oil from hydrodistillation is entirely not economical since it only produces low oil recovery yield from fresh rose flowers.

CONCLUSION

Bulky amount of rose flowers in Karangpring village can be a valuable natural resource on producing rose essential oil, and aromatic rose water depend on the extraction method to use. Rose oil is better extracted using enfleurage of fresh rose flowers than using hydrodistillation process by showing diverse compound types in it, from rose alcohols into stearoptenes. This rose absolute also has a more pleasant odor like the true fragrance of rose flowers than rose oil.

Beside, rose water showed a mature content of rose flower marker compound, 2-phenylethyl alcohol, making this aromatic water to be a precious candidate of flavor and aromatherapy agent obtaining from the distillation process.

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