OPEN ACCESS Science International

ISSN 2305-1884
DOI: 10.17311/sciintl.2017.84.95

Research Article
Comparative Efficacy of Various Essential Oil Extraction Techniques on Oil Yield and Quality of *Jasminum sambac* L.

[1,2]Ahsan Akram, [1]Adnan Younis, [1]Gulzar Akhtar, [3]Kashif Ameer, [4]Amjad Farooq, [5]Muhammad Asif Hanif, [6]Muhammad Saeed and [2]Ki-Byung Lim

[1]Institute of Horticultural Science, University of Agriculture, 38040 Faisalabad, Pakistan
[2]Department of Horticultural Science, Kyungpook National University, 702-701 Daegu , Korea
[3]Department of Food Science and Technology, Kyungpook National University, 702-701 Daegu, Korea
[4]Department of Environmental Science, COMSATS Institute of Information Technology, 61100 Vehari, Pakistan
[5]Department of Chemistry and Biochemistry, University of Agriculture, 38040 Faisalabad, Pakistan
[6]Directorate of Floriculture (Training and Research) Lahore, Pakistan

Abstract

Objective: The current investigation was conducted to check the comparative efficacy of different essential oil extraction techniques. Essential oil was extracted by supercritical fluid extraction (SCFE) and hydro-distillation. The objective of this study was to optimize standard conditions for maximum oil yield from *jasminum sambac* through SCFE technology and compare essential oil extraction techniques for better oil quality. **Materials and Methods:** An optimized protocol was established for the extraction of essential oil from *Jasminum sambac* through supercritical fluid extraction (SCFE) technique. The essential oil extraction of *Jasminum sambac* also done with another technique i.e., Hydro-distillation. Then comparison was done between both techniques regarding quality and quantity of essential oil. Experiment was laid out according to completely randomized design (CRD). The data were subjected to statistical analysis using one-way analysis of variance technique at 5% probability. **Results:** In supercritical fluid extraction (SCFE), CO_2 was used as solvent for getting concrete oil. The concrete was processed to convert it to absolute oil. *Jasminum sambac* had concrete oil yield of 0.334%, whereas it had absolute oil yield of 0.021%. Physio-chemical studies of absolute oil were also conducted which indicated color as clear yellow to light brown, refractive index as 1.49, specific gravity as 0.964 and congealing point of the concrete as 20°C. Different chemical components contained in the essential oil were identified by GC-MS analysis. The maximum number of chemical components identified from single essential oil sample was 35. **Conclusion:** This technique performed better as compared to hydro-distillation. The present studies revealed that *Jasminum sambac* can be successfully cultivated on commercial scale in the country for the extraction of essential oil with promising results.

Key words: Essential oil, hydro-distillation, *Jasminum sambac*, supercritical fluid extraction, *Jasminum grandiflorum*

Citation: Ahsan Akram, Adnan Younis, Gulzar Akhtar, Kashif Ameer, Amjad Farooq, Muhammad Asif Hanif, Muhammad Saeed and Ki-Byung Lim, 2017. Comparative efficacy of various essential oil extraction techniques on oil yield and quality of *Jasminum sambac* L. Sci. Int., 5: 84-95.

Corresponding Author: Ahsan Akram, Institute of Horticultural Science, University of Agriculture, 38040 Faisalabad, Pakistan Tel: 92 312 6623666

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Jasminum sambac belongs to family *Oleaceae*, which comprised of about 200 species while actual number of its true species is only 89 out of which 40 populate in subcontinent[1]. Among the *Jasminum* species *Jasminum sambac* and *Jasminum grandiflorum* are cultivated commercially for oil extraction.

Essential oils are being obtained from many kinds of plants. The genus *Jasminum*, comprise an important group of plants being used in the whole world for the extraction of its precious essential oil. According to Mitsui[2] there are about 400,000 odorants which include both pleasant and unpleasant odors. Among these odorants author describes Jasmine a foremost perfume with a beautiful fragrance containing about 200 components extending from pleasant jasmine fragrance, lactone to extremely unpleasant indole. Jasmine plants had been given due importance in the past. For example the variety *J. grandiflorum* is under cultivation for the last two centuries in the Grasse region of France. Vaze[3] watched an excellent progress in the Indian essential oil industry during the last 50 years. As a result of their study they considered combined efforts of industry, research, development agencies and farmers necessary to make fast growth in the development of essential oil industry in the world. The countries of Bulgaria, Turkey, Southern France and Morocco are the major producing regions of jasmine[2].

Enhancing value of crops of small scale farms elevates the income and potential of small scale growers[4]. This can be accomplished by promoting the cultivation of refined horticultural strains/crops with greater production potential and developing and promoting the use of appropriate extraction technology to increase product value. Extraction of essential oils from high value crops is an effective method to transform a delicate product that is difficult to transport with a low density of value into a robust product of high economic density which is easy to transport[5]. For example, 6000 kg of jasmine grown in a rural area trucked from the farm can be sold in local Pakistani market for ~$3000, the equivalent value of 1 kg of jasmine oil which would be most convenient to transport and can be sold on the international market for ~$9000.

Jasmine essential oil is called as otto or attar which is taken from flower petals, has a wide range of application in perfumery, soap and cosmetic industry. It is also medically used for tonning dry, greasy, irritated and sensitive skin, beneficial for respiratory system, helpful in relieving muscle pain, sprains and limbs. Likewise, therapeutically jasmine oil is used as an antidepressant, antiseptic, antispasmodic, sedative and uterine[6]. Jasmine oil has many applications in our daily life. For example, methanol extract of *Jasminum amplexicaule* and its different fractions can be used to cure diarrhea and pains[7]. Friedman *et al*.[8] assessed jasmine oil as active oil against *Campylobacter jejune*. Jasmine flowers can be added to the water content of tea to prepare jasmine tea[9]. Nakayama *et al*.[10] testified that jasmine essential oil could exert a wide variety of psychological and biological effects on human. The above references reveal that essential oil of jasmine contains many useful constituents which can be used in many ways.

In Pakistan no trustworthy data are available regarding production and consumption of essential oils in food and beverages, medicine, cosmetics and other industrial uses, although many companies and individuals claim to produce these oils in small or large quantity. Such claims also need scientific verification to evaluate whether the oils are true to type or not. For this kind of data base to establish, a separate study will be required. Essential oils are being imported in the country on a reasonably large scale. According to Pakistan Statistical Yearbook[11] the total cost of imports of essential oils, perfumes and flovour materials was Rs. 10158.35 million in the year 2003-04. This figure is a mixture of essential oils and other perfumes, etc. Data about the exports of such materials are silent. It can be seen that a huge amount of money is being spent every year on the import of essential oils and flavouring substances. Production of these substances in the country on industrial basis will not serve the local demands but also a lot of foreign exchange may also be saved.

There is various extraction techniques such as hydro-distillation, steam distillation, direct steam distillation and solvent extraction[12] are employed for the purpose of essential oil extraction. The traditional methods for the extraction of plant materials include steam distillation and organic solvent extraction using percolation, maceration or Soxhlet's techniques[13]. These procedures however, have distinct drawbacks such as time consuming and labor intensive operations, handling of large volumes of hazardous solvents and extended concentration steps which can result in the loss or degradation of target analytes. Moreover, there is increasing interest for alternative extraction technologies consuming less organic solvents, because of the rising solvent acquisition and disposal costs and regulatory restrictions. Supercritical fluid extraction (SCFE) is a rapid, selective and convenient method for sample preparation prior to the analysis of compounds in the volatile product of plant matrices[14,15].

Supercritical fluid extraction (SCFE) has ousted all other extraction methods because it is a quick selective and environment friendly. The SFE is usually performed with pure or modified carbon dioxide, which is inert and no hazardous effects[15]. Voluminous research work can be found internationally on oil extraction from different *Jasminum* species. Unfortunately, very little research work regarding this aspect has been conducted in Pakistan. No sound effort has been made so far in the country to exploit the potential of cultivation of *Jasminum* specie for essential oil production. If taken scientifically, jasmine cultivation can become a source of value addition and ultimately a means of foreign exchange earnings in addition to reap many associated benefits. The essential oil was extracted from *Jasminum sambac* through two different extraction techniques like hydro-distillation and supercritical fluid extraction (SCFE), which was followed by their various physio-chemical analyses in order to make assessment of the quality. Particular attention was given to analyze the samples of essential oil of *Jasminum sambac* through latest GC-MS technique for the identification of constituents. The objectives of present study were to Optimize standard conditions for maximum oil yield from *jasminum sambac* through SCFE technology and compare essential oil extraction techniques for better oil quality.

MATERIALS AND METHODS

The present study was conducted at Rose project laboratory, Institute of Horticultural Sciences, University of Agriculture, Faisalabad during 2013-14. Jasmine flowers were picked, their fresh weights were taken, then their petals were separated and used for hydro-distillation. Prior to placement in steam distillation apparatus the petals were spread in a tray under shade at room temperature in order to remove the excessive moisture and then used for extraction.

Collection of plant material: The healthy plants of jasmine were selected for the collection of flower petals from Rose Project, Institute of Horticultural Sciences, University of Agriculture, Faisalabad. This is a plain region, with relatively mild climate and can be considered more suitable for the cultivation of plants like jasmine. This area is near to Pattoki (flower growing and marketing hub of Punjab, Pakistan). Thus the climatic conditions of the area provide a nice opportunity for ideal growth of the plants. Gunkel *et al.*[16] reported that geographical location of the area also affects the quality of finished flower oils of jasmine. It was therefore, perceived during the conduct of the experiment that essential oil thus obtained would be of fine quality. Flowers were harvested before sunrise in order to ensure the minimum loss of volatile aromatic substances with maximum oil recovery.

Handling of plant material: Every time the flower petals were collected they were immediately packed in cardboard perforated boxes lined with polyethylene sheet from inside. These boxes were kept in refrigerator till the time of starting the extraction process. The material was then taken to the oil extraction laboratory for extraction of essential oil.

Preparation of plant material for extraction: Sepals, leaves and any other undesirable plant part were separated from the petals to avoid contamination in the final product. Flowers were weighed carefully after cleaning. These cleaned petals were put in extraction vessel of SCFE plant and for hydro-distillation put the petals in distillation flasks immediately for further working.

Extraction of essential oil: To capture the aroma present in the botanical material, essential oil is obtained which may be in the form of concrete and absolute. Essential oil extraction was carried out by two methods, viz., supercritical fluid extraction (SCFE) and hydro-distillation. Calibrated glassware and chemicals of analytical grade were used in the experiments. All the apparatus used was thoroughly washed and dried every time the extraction and distillation process was carried out.

Supercritical fluid extraction (SCFE): Any Compound above its critical temperature and pressure is called supercritical. At supercritical stage, neither liquid nor gaseous form exists. An intermediate of both liquid and gas known as fluid can be found with properties of liquid and gas molecules. The extraction with SCFE was performed in a pilot unit in Rose Project, Institute of Horticultural Sciences, University of Agriculture, Faisalabad. The SCFE pilot plant has one CO_2 tank, heat exchanger, gas booster, one extractor and one separator vessel.

The SCFE include following steps:

- CO_2 conditioning
- Extraction process
- Extract recovery

A gas booster received liquid CO_2 (99% pure) from a cylinder and pressurized a jacketed surge tank which in turn provided gas to a jacketed extraction vessel. The jacketed surge tank was placed between the gas booster and the extraction vessel in order to avoid potential pressure overshoots allowing a better pressure control. The temperature of the surge tank and extraction vessel was controlled by a thermostatic water bath. The *Jasminum sambac* flowers were filled in the extraction vessel after weighing of 20 kg weight. The extraction pressure was maintained by the gas booster, monitored by a pressure tansductor and controlled by a pneumatic control valve. The samples were collected at different time intervals in a separation vessel at specified temperature and pressure ranges as mentioned in treatments section. That allows the separation of the oil by changing the CO_2 phase. The temperature and pressure maintained through a connected computer having SCFE run software. Extraction is carried out typically at temperatures between 35-60°C[14]. Absolute oil of *Jasminum sambac* was collected in a flask.

Yield attributes: Essential oil yield was obtained as concrete and absolute oil recovery.

Concrete oil yield: At first step the concrete oil yield was obtained. For this purpose the dissolved organic residue in n-hexane was collected in a flask which was taken to rotary evaporator to recover n-hexane for further use and to collect extract of essential oil. Recovery of n-hexane from the solution started at 40°C. After the recovery of n-hexane the remaining extract was taken in falcon tubes and subject to centrifuge at 3500 rpm. The process was repeated 4-5 times till a thick viscous substance was achieved. This was collected in pre-weighed flask and the weight of concrete was determined again by weighing the flask. This concrete oil was kept in refrigerator at 2-4°C till the commencement of next process of making absolute. However, in case of hydro-distillation almost pure oil was obtained. Therefore, that oil was not subjected to any purification process. The percentage of concrete oil yield was calculated by the following equation.

$$\text{Percentage of concrete oil (on petal basis)} = \frac{\text{Weight of fresh petals}}{\text{Weight of concrete oil}} \times 100$$

Absolute oil yield: Concrete oil is not considered pure because it contains waxes, traces of solvent and other undesirable materials. A procedure was adapted to convert the concrete to absolute by removing waxes and other undesirable materials. Moisture contained in the organic residue remaining in the flask (concrete oil) was removed by adding anhydrous sodium sulphate followed by its filtration through Whatmann filter paper No. 43. Then a small volume of absolute alcohol was added to this concrete. That absolute alcohol was removed by performing distillation process using rotary evaporator. Least traces of alcohol were removed by bubbling nitrogen gas through this oil.

The percentage of absolute oil yield was calculated by the following equation.

$$\text{Percentage of absolute oil} = \frac{\text{Wt. of fresh petals}}{\text{Wt. of absolute oil}} \times 100$$

Concrete oil percentage: The extract of previous existed plant tissues which hold some soluble substances and waxy stuff is called concrete oil. First the 100 mL flask was weighed and then concrete oil was added to it. Again the flask with concrete oil was weighed and weight of concrete oil was recorded.

Concrete oil content (%) was calculated as:

$$\text{Percentage of concrete oil} = \frac{\text{Weight of concrete oil}}{\text{Weight of petals}} \times 100$$

Absolute oil percentage: Absolute oil was recovered from concrete oil according to the operation discussed earlier.

The absolute oil content (%) was calculated as:

$$\text{Percentage of absolute oil (on petal basis)} = \frac{\text{Weight of absolute oil}}{\text{Weight of petals}} \times 100$$

$$\text{Percentage of absolute oil (on concrete oil basis)} = \frac{\text{Weight of absolute oil}}{\text{Weight of concrete oil}} \times 100$$

Storage of essential oil: The essential oil, both left-over concrete and absolute were stored in tightly packed Falcon tubes and small HPLC vials which were kept in refrigerator at low temperature of 2-4°C in order to assure the minimum loss of volatile substances from this oil.

Physical attributes: For the evaluation of quality and to determine the constituents of essential oil, certain physical and chemical properties were analyzed using the available facilities. Maximum care was taken during the conduct of these analyses to achieve satisfactory results.

Color: Color is a sensation that is aroused when light falls on the retina of the eye[17]. Color of absolute oil was determined by using spectrophotometer. The instrument recorded transmittance measurement between 400 and 700 nm.

Specific gravity: Specific gravity is the ratio of the density of a given substance to the density of reference at standard temperature and pressure. The standard of reference for solids and liquids is the density of water[17]. Specific gravity acts as important criteria for the determination of quality and purity of an essential oil. For the determination of this physical property of the essential oil the specific gravity bottle was used. The pre-weighed 10 mL specific gravity bottles were filled one by one with absolute oil of all the three varieties, leaving no air bubbles and then weighed. The density of oil was computed by using the following equation.

$$\text{Density} = \frac{\text{Weight}}{\text{Volume}}$$

$$\text{Specipic gravity} = \frac{\text{Density of liquid at 20°C}}{\text{Density of water at same temperature}}$$

Congealing point: In case of mixtures, such as essential oils, congealing point is determined instead of melting point. In its determination, the oil is super cooled so that upon congealation liberation of heat occurs with immediate crystallization[16]. For the determination of congealing point, about 10 mL of absolute oil was placed in a dry test tube of 20 mm diameter. This was cooled in water the temperature of which was about 5°C lower than the supposed congealing point of the essential oil. The inner walls of the tube were rubbed with a thermometer quickly up and down in the oil. The temperature was noted constantly. There was a rise in temperature which soon approached a constant value. This value was taken as congealing point of the oil. The process was repeated many times in order to ensure accuracy.

Refractive index: The refractive index of the oil was determined at room temperature of 25°C by using Abbe's Refractometer[18].

Optical rotation: Optical rotation is the turning of the plane of linearly polarized light about the direction of motion as the light travels through certain materials.

A 10 mL polarizer and analyzer was taken in filling the tube to avoid that there was no air bubble, which could disturb the field of light. Analyzer was slowly turned until both the halves of the field were viewed through the telescope. The direction of rotation was determined if the analyzer was turned counter clockwise from the zero position to obtain the final reading, the rotation if anticlockwise is levo (+) and if clockwise is dextro (-).

Chemical attributes

Acid number: Acid number is the number of mg of potassium hydroxide (KOH) required to neutralize the free fatty acids in 1 g of oil. Acid number was determined through indicator method[18].

Absolute oil was weighed 1.5 mg accurately in a 100 cm^3 sponification flask. About 15 cm^3 of neutral, 95% alcohol and 3 drops of 1% phenolphthalein solution were added. Titration of the free acid with a standard 0.1 N aqueous solution of NaOH was done by adding alkali drop wise at a uniform rate. The contents of the flask were continuously shaken as an end point was observed. Process was repeated 3 times to get mean value.

The acid number was calculated by the following equation:

$$\text{Acid number} = \frac{56.1\ (\text{Number of } cm^3 \text{ of } 0.1\text{N NaOH})}{\text{Weight of the sample in grams}}$$

Ester number: The ester number is the number of mg of potassium hydroxide required to sponify the ester contained in 1 g of oil. Another sample of absolute oil was taken. About 10 cm^3 of 0.5 N solution of potassium hydroxide was added. A glass air cooled condenser was attached to the flask and contents were refluxed for 1 h on a water bath. The apparatus was removed and allowed to cool at room temperature for 15 min. Excess alkali was titrated against standardize 0.5 N aqueous HCl.

The ester content was calculated by the following equation:

$$\text{Ester number} = \frac{28.05\ (a)}{S}$$

Where:

a = Number of cm^3 of 0.5 N HCl used in saponification

s = Weight of sample in grams

Hydro-distillation: In hydro-distillation process, the compounds of a mixture of liquid (or liquids) are separated by creating difference in their vapor pressure. In the present study the flower petals were separated from sepals, etc. as usual and their weighed quantity of 1000 g was put in the flask containing sufficient quantity of water. The flower petals remained in direct contact with the boiling water for about 3 h. On boiling the water vapors containing oil raised and passed through the condenser. The vapors and oil drops cooled down and water came back in the flask containing flowers while the essential oil, being light in weight, started

settling on the top of water in the jacket. Resultantly a thin layer of oil was separated from the petals, usually combined with water in the bottom. This thin layer of essential oil was separated with the help of micro pipette and stored in Eppendorf or HPLC vials. The oil extracted was not purified further.

Comparison of SCFE and hydro-distillation techniques: The SCFE optimized treatment's best result was used to compare with hydro-distillation extraction technique's result. This comparison was made on the basis of following attributes, efficiency of extraction technique and time. The detail of each attributes discussed in SCFE and hydro-distillation extraction as well.

Statistical analysis: Experiment was laid out according to completely randomized design (CRD). The data were subjected to statistical analysis using one-way analysis of variance technique at 5% probability[19].

RESULTS

In current investigation, standardization of SCFE technology to get maximum essential oil yield of *J. sambac* from this modern extraction technology. Simultaneously, a conventional extraction technique hydro-distillation was also used to extract essential oil from *J. sambac* flowers.

Ultimately, it compare the efficacy of both the extraction methods under local condition and to make recommendations in the light of results.

Physical properties: The color of the absolute oil of jasmine flowers was clear yellow in case essential oil obtained through SCFE and it was very light brown in case hydro-distillation. The refractive index at 20°C for oil extracted through SCFE was 1.49, while from essential oil extracted hydro-distillation it was 1.37 (Table 1). The highest temperature at which an essential oil is solidified is called its congealing point. The congealing point of essential oil obtained through SCFE was 20°C while it was 57°C in case hydro-distillation. The optical rotation of *Jasminum sambac* essential oil from SCFE was +3.30, while for hydro-distillation essential oil extraction, it was +2.34. The specific gravity at 30°C of essential oil from SCFE was recorded as 0.964, while for oil extracted through hydro-distillation it was recorded as 0.895 (Table 1).

Chemical properties: The acid number of *Jasminum sambac* essential oil extracted for both SCFE and hydro-distillation essential oil extraction was 0.950 and 0.913, respectively. It was observed that the ester number of essential oils of

Table 1: Comparison of essential oil physio-chemical properties of *Jasminum sambac* (SCFE and HD extraction techniques)

Physio-chemical properties	SCFE	HD
Color	Clear yellow	Very light brown
Refractive index	1.49±0.0035	1.37±0.002
Congealing point	20°C±1.00	57°C±1.00
Optical rotation	+3.30 at 20°C	+2.34 at 20°C
Specific gravity	0.964 at 30°C±0.0001	0.895 at 30°C±0.0001
Acid number	0.950	0.913
Easter number	213	204

Standard deviation (SD) Value: 0.0015

Jasminum sambac essential oil extracted through SCFE and hydro-distillation essential oil extraction was 213 and 204, respectively (Table 1). Esters are very important in regard to the odor of the oil. In the present study, oil extracted through SCFE had the highest ester number, meaning essential oil had a strong odor.

Comparison of essential oil yield of *Jasminum sambac* (SCFE and HD extraction techniques): Data regarding concrete and absolute oil (%) of *Jasminum sambac* essential oil obtained in SCFE and hydro-distillation extraction techniques. Essential oil yield of *jasminum sambac* under study varied from 0.334-0.024% in supercritical fluid extraction. The absolute oil yield obtained from this concrete was 0.021%. It means that absolute oil recovery from concrete was about 54%, i.e., almost half of concrete.

Hydro-distillation is widely used method for the extraction of essential oils, particularly for delicate flowers like those of jasmine and roses. In the present study this method gave oil yield of concrete 0.291% and the absolute oil yield obtained from this concrete was 0.018% for *J. sambac*. Essential oil yield obtained by hydro-distillation is usually low as compared to supercritical fluid extraction. The above results regarding comparison of concrete and absolute oil (%) are also represented graphically in Fig. 1.

Chemical components of *jasminum sambac* essential oil: In this comparison experiment of two essential oil extraction techniques, supercritical fluid extraction and hydro-distillation through physio-chemical, yield attributes and chemical constituents, SCFE technique performed well in quality, quantity and batch extraction performance GC-MS analysis of essential oil extracted through SCFE and hydro-distillation extraction techniques was carried out. Results of GC-MS chromatogram of *Jasminum sambac* essential oil showing in Fig. 2 and 3, respectively, represented number of chemical components in both extraction techniques. SCFE exhibited maximum chemical components (35) while hydro-distillation, extraction technique was at the bottom with (30) chemical components.

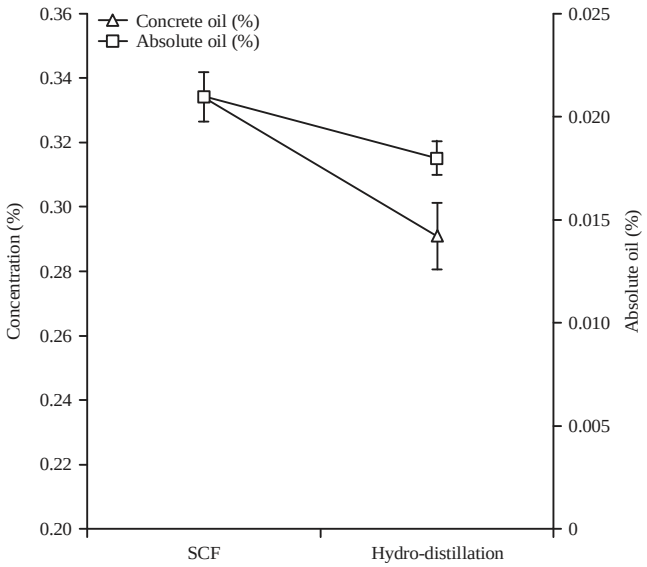

Fig. 1: SCFE and hydro-distillation showing copmparison of concrete and absolute oil yield (%), standard deviation (SD)

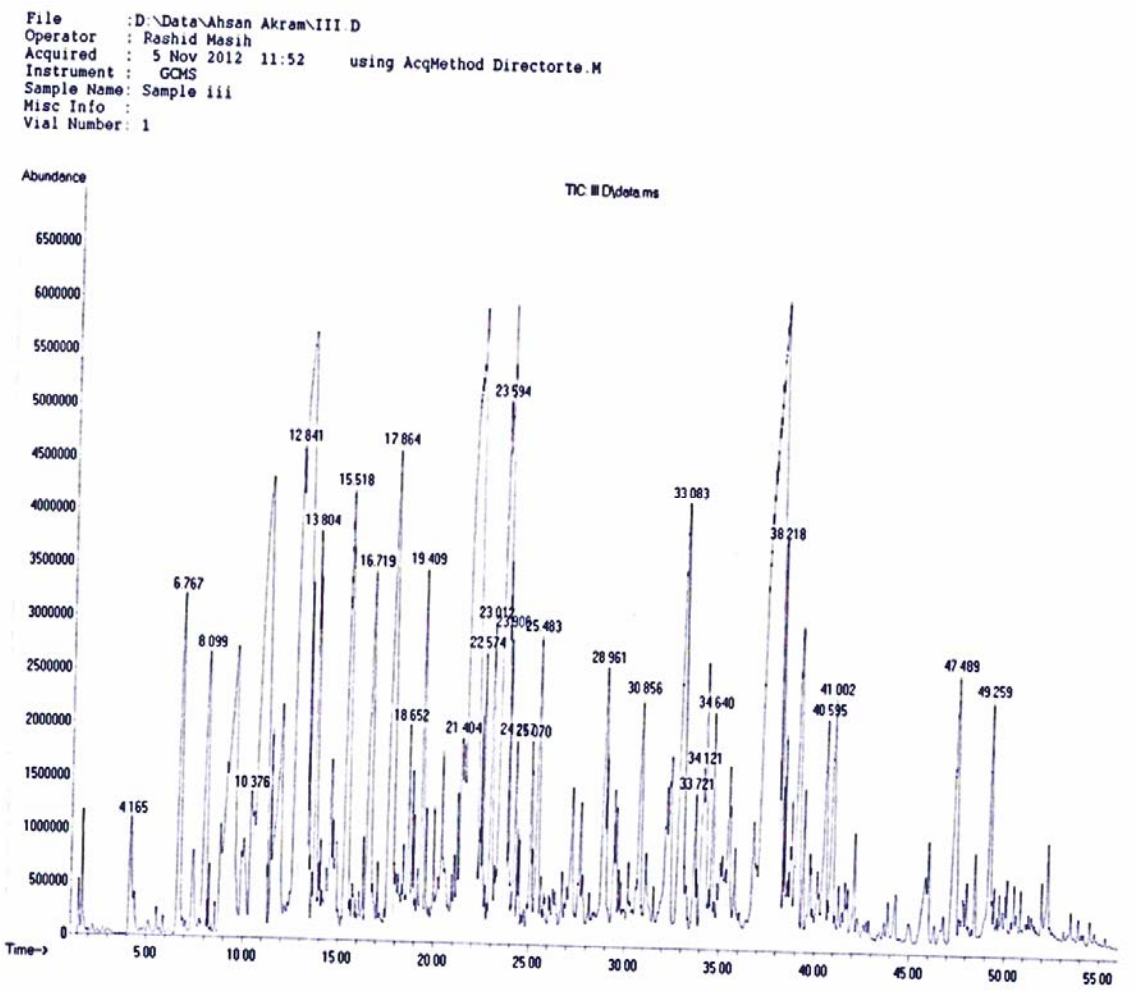

Fig. 2: SCFE showing chemical components in GC-MS chromatogram

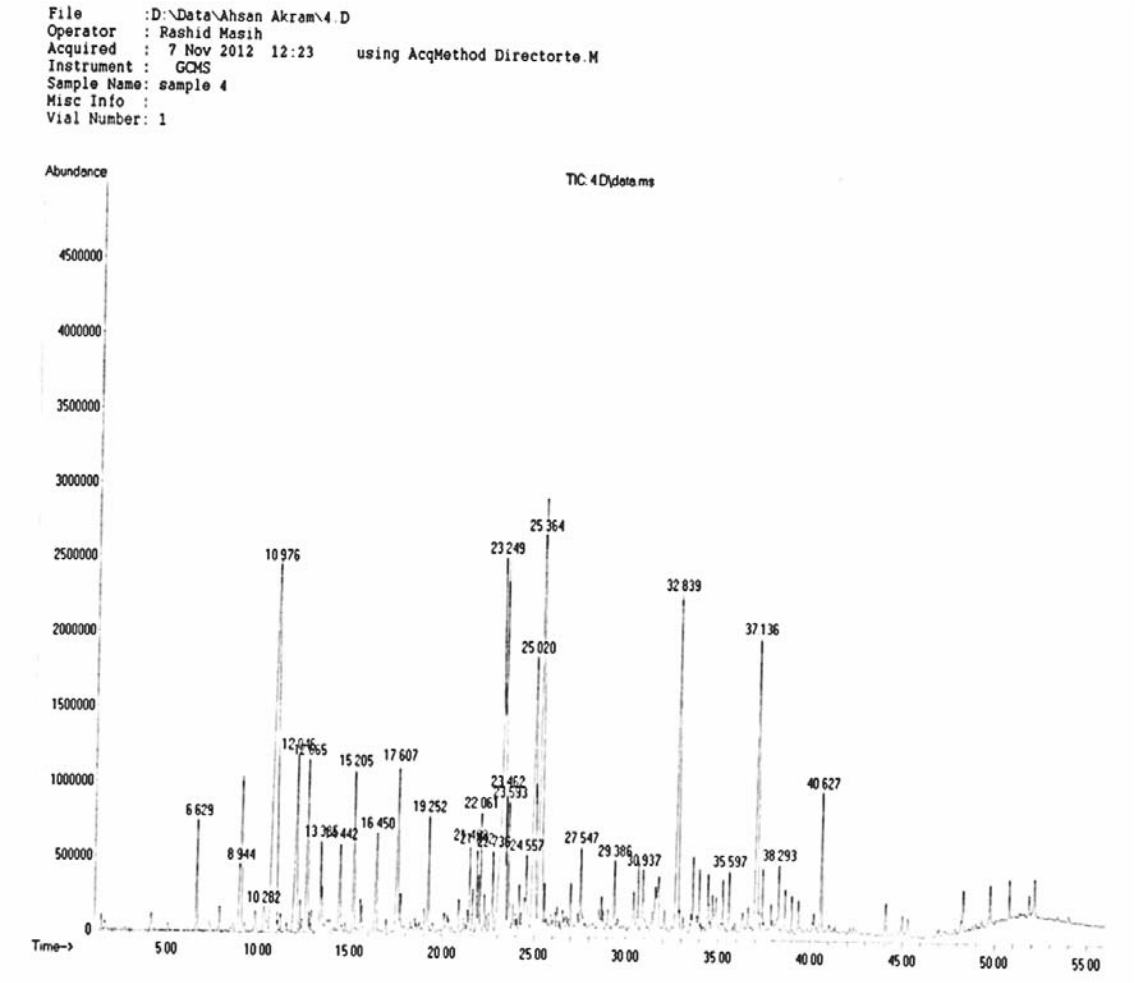

Fig. 3: Hydro-distillation showing chemical components in GC-MS chromatogram

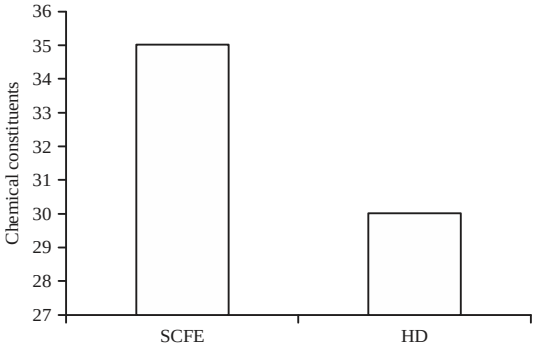

Fig. 4: SCFE and HD showing chemical constituents obtained from *Jasminum sambac* essential oil through GC-MS chromatograms

Data regarding chemical components of *Jasminum sambac* essential oil obtained in SCFE and hydro-distillation extraction techniques. The results regarding chemical components are also represented graphically in Fig. 4.

DISCUSSION

Essential oil extraction was done by two methods, supercritical fluid extraction (SCFE) and hydro-distillation. As Jasmine essential oils are composed of heat-sensitive chemical compounds, the use of conventional steam distillation technique would inevitably inflict thermal degradation to the natural fragrance[20]. Techniques commonly used to extract the essential oils include steam distillation, hydro-distillation, enfleurage, dynamic and static headspace, solvent extraction and supercritical fluid extraction[21]. Comparison between hydro-distillation and supercritical fluid extraction reveals the superiority of the green solvent, SC-CO_2 to obtain a good yield and a flavor similar to plant material and to keep the native composition present in the genuine plant[22]. Now a days, research has focused on utilizing supercritical carbon dioxide technology in processing fine chemicals, pharmaceutical intermediates and nutraceuticals. In addition to being a

solvent for extraction and fractionation (purification) of organic compounds, carbon dioxide is also increasingly being utilized as a medium for reactions[23].

Supercritical fluids (SCF) has received much attention especially in food, pharmaceutical and cosmetic industries, because it presents an alternative for conventional processes such as organic solvent extraction and steam distillation[24,25]. Supercritical fluids (SCF), is an extraction method which have high pressure but low temperature condition. This low temperature separation process prevents the degradation of the chemical compounds of the extract due to heat, as in steam distillation[26,27].

Study on the supercritical CO_2 fractionation of jasmine concrete concluded that it avoids thermal degradation of the product and prevents pollution of the environment with organic solvents. Also CO_2 density was increased in three successive steps from 0.28-0.84 g cm^{-3}, for selective extraction of different compounds and the resultant products were paraffin-free and showed a lower fatty acid methyl ester content than the traditional products derived by hexane extraction[28]. Jasmine concrete oil extracted with liquid carbon dioxide is artifact-free and solvent-residue-free. It also has a natural aroma, golden yellow colour and good storage life[29].

Each method has its own merits and demerits. The most widely practiced method for recovery of most of essential oils is hydro-distillation which requires extracting raw materials by various solvents such as hexane, foranes etc. for obtaining many sensitive and expensive oils. After extraction the solvent is removed by distillation leaving a resionid or concrete. An increasing popularity of essential oils and extracts of numerous flowers like rose, jasmine, tuberose, champa etc. can be viewed in high grade perfumes and aroma therapy[30].

Essential oil yield of *Jasminum sambac* under study varied from 0.334-0.021% in supercritical fluid extraction. The absolute oil yield obtained from this concrete was 0.021-0.16%. It means that absolute oil recovery from concrete was about 54%, i.e., almost half of concrete. The findings support the results of Younis *et al.*[31], who observed about half absolute oil recovery from the concrete of *J. sambac*. The results are also in confirmatory with those of Weiss[32], who reported the concrete yield of 0.33% for *J. grandiflorum* and 0.17% for *J. sambac*. The results of present study do not support the findings of Waheed-ur-Rehman[33] who reported much higher percentage of 0.304% of absolute oil recovery in *J. sambac* from n-hexane solvent extraction and 0.233% absolute oil recovery when SCFE was used for oil extraction.

Hydro-distillation is a widely used method for the extraction of essential oils, particularly for delicate flowers like those of jasmine and roses. In the present study this method gave oil yield of 0.008%. Essential oil yield obtained by hydro-distillation is usually low as compared to solvent extraction. Waheed-ur-Rehman[33] also recorded low essential oil recovery of 0.207% by hydro-distillation as compared to solvent extraction. Researchers have gained quite variable results with respect to oil production. This is so because many factors may effect the oil yield from flowers. These factors include the variety it self, topography, time of harvest, month of harvest, skill of labour engaged in harvesting of flowers, climatic conditions, cultural practices and administration control over the whole process of extraction.

In the present study the above two methods were used for the extraction of essential oil from *Jasminum sambac*. Both the methods have their own advantages and disadvantages. Handa *et al.*[34] proposed that supercritical fluid extraction was suitable for expansive, delicate and thermally unstable materials like jasmine, tuberose and hyacinth. Solvent extraction is a costly method which involves the use of costly apparatus like rotary vacuum evaporator and centrifuge apparatus. Solvent extraction requires complicated and expensive apparatus and the running expenses are high[16]. However, this method initially yields concrete oil from which absolute is recovered. This concrete is of great importance for the industry because in many industrial products concrete is used as such. Huge quantities of concrete are being prepared from jasmine and their flowers in many countries. Concrete exports steadily increased from countries like Egypt, Spain, Morocco and Algeria in the 1970s[32]. Hydro-distillation is still being used as the principal method for the preparation of essential oils. This is a cheap method and does not involve costly apparatus. The units of hydro-distillation can be set up in close proximity to the production fields. Although, SCFE has more initial cost but it pays long time. Major advantage of SCFE over steam and hydro-distillation is that it avoids thermal degradation of chemical compounds which is not possible in conventional extraction methods.

Certain physical analysis was conducted for the determination of the quality of essential oil of jasmine. These included color, specific gravity, congealing point and refractive index.

The color of *Jasminum sambac* was light brown was dark brown. There is variation of results reported by different researchers. It may be due to the reason that factors responsible for affecting yield, as noted above in this discussion, also affect the physical and chemical qualities of essential oil. Therefore, minor variation in results is not uncommon in the available literature. Even some have mentioned the color and yield, etc.) without mentioning the name of variety. For example Guenther[35] reported the color of

jasmine absolute as a clear yellow-brown liquid. Our results are more or less similar to those who have already reported their findings. For instance, Weiss[32] reported the colour of concrete of *Jasminum sambac* to be deep red. Younis *et al.*[31] reported the color of oil of *J. sambac* to be off-whitish yellow. Anonymous[36] reported the color of concrete of *J. grandiflorum* to be light yellow to brown. The color of the absolute oil of jasmine flowers was clear yellow in case essential oil obtained through SCFE and it was very light brown in case hydro-distillation which isin contrast to the findings of Gilbert *et al.*[37], who found jasmine oil to be reddish brown in color. The method of extraction and environmental conditions can influence the color of oil. The color of oil obtained through SCFE was off-whitish yellow while it was light brown in case hydro-distillation essential oil extraction.

Determination of specific gravity is also an important physical parameter of essential oils. Like other physical parameters the results of this property of essential oil also vary to some extent from those of other researchers. In the present study refractive index at 20°C for oil extracted through SCFE was 1.49, while for essential oil extracted hydro-distillation it was 1.37 (Table 1). Usually oil extracted through different methods has different congealing points[38], in the present study two extraction methods were used to extract oil, so the different congealing point was observed in both extraction techniques. In comparison with the Weiss[32] reported specific gravity of concrete of *J. sambac* to be 0.8794, respectively at 30°C, Eid *et al.*[39] reported the specific gravity of concrete of *J. grandiflorum* to be 0.82 at 20°C and Younis *et al.*[31] reported specific gravity of absolute of *J. sambac* to be 0.9850 at 20°C. Thus it can be seen that the results of various researchers differ from one another to some extent.

Determination of congealing point of concrete was also made for *Jasminum sambac* under study. The highest temperature at which an essential oil is solidified is called its congealing point. The congealing point of essential oil obtained through SCFE was 20°C while it was 57°C in case hydro-distillation. Other researchers have reported their results of congealing point which vary from one an other to some extent and vary to large extent for concrete and absolute. The congealing point of concrete is always higher because it contains waxes and other odourless substances. Congealing point of absolute of *J. sambac* was 17.25°C as reported by Younis *et al.*[31]. Anonymous[36] reported congealing point of concrete of *J. grandiflorum* to be 54-55°C. In the present study the congealing point of concrete of *J. sambac* was taken.

The optical rotation of *Jasminum sambac* essential oil from SCFE was +3.30, while for hydro-distillation essential oil extraction, it was +2.34. The specific gravity at 30°C for essential oil from SCFE was recorded as 0.964, while for oil extracted through hydro-distillation it was recorded as 0.895 (Table 1).

The last physical parameter observed was refractive index. Measurements were taken with the help of refractometer. The results show that this index for *J. Sambac*, 1.375. These findings although differerent, but are in close proximity to those of other researchers. Guenther[35] observed refractive index of jasmine to be 1.4822 at 2°C, Weiss[32] reported refractive index of concrete of *J. grandiflorum* and *J. sambac* to be 1.4750 and 1.4665, respectively. The varying results of the *Jasminum* varieties may be seen in the context of different climatic and geographical conditions as concluded by early research workers.

The acid number for *Jasminum sambac* essential oil extracted for both SCFE and hydro-distillation essential oil extraction was 0.950 and 0.913, respectively. It was observed that the ester number of essential oil of *Jasminum sambac* essential oil extracted through SCFE and hydro-distillation essential oil extraction was 213 and 204, respectively (Table 1). Esters are very important in regard to the odor of the oil. In the present study, oil extracted through SCFE had the highest ester number, meaning essential oil had a strong odor. Rose that had been slightly fermented prior to distillation yield oil with a much higher number than the oil extracted from freshly picked flowers.

The physical and chemical properties of essential oils are strongly influenced by variety, degree of maturity, seasonal variations, seasonal rainfall and method of extraction and yield of oil.

This study proved to helpful for essential oil industry to opt the best extraction technique. Pakistan as an agricultural country is rich in medicinally or economically important flora. The current status of Pakistan's essential oil industry is not encouraging and productive. The essential oil industry in the country has not so far been established on the sound footings due to lack of adoption of modern agricultural and horticultural practices and better understanding for their uses as value added products .

CONCLUSION

In present study, it was concluded that the SCFE extraction technique proved to be an efficient method of essential oil extraction. It was also proved that contents of *Jasminum sambac* were significantly affected by harvesting stage of flowers. There were variations in constituents of essential oil at different extraction conditions of SCFE. The

quantity of essential oil obtained differed in both extraction techniques as more oil was obtained SCFE and less oil in quantity was obtained in hydro-distillation. These results showed that the composition of essential oils in the plant at different extraction conditions changes and the production of a specific component of these essential oils depend on which extraction technique and condition we use for extraction of *Jasminum sambac* essential oil.

SIGNIFICANCE STATEMENTS

This study determine the best extraction technique for essential oil of *Jasminum sambac.* Supercritical fluid extraction (SCFE) proved to be the best among both extraction techniques. Physio-chemical analysis of jasmine essential oil were also conducted to determine color, specific gravity, refractive index and congealing point, acid and ester number. The number of chemical components in *Jasminum sambac* essntial oil along with their retention time was identified through GC-MS technique. Successful cultivation of *Jasminum sambac* can paved the way to boost perfumery and essential oil industry in the country. This study will help the researcher to reveal the best optimized extraction technique.

ACKNOWLEDGMENTS

This study was conducted at Institute of Horticultural Sciences, University of Agriculture, Faisalabad, Pakistan and supported by HEC, Pakistan under 5000 Indigenous Ph.D Scholarship. This study was supported and published by Floriculture Biotechnology Laboratory, Department of Horticultural Science, Kyungpook National University, 41566 Daegu, Republic of Korea.

REFERENCES

1. Irulappan, I., 1994. Breeding Jasmine for High Yield of Flowers and Floral Oil. In: Floriculture Technology, Prakash, J. and K.R. Bhandary (Eds.). Oxford and IBH Press, India.
2. Mitsui, T., 1997. New Cosmetic Science. Elsevier, New York, ISBN: 9780080537498, pp: 101-106.
3. Vaze, S.V., 2000. Indian essential oil industry: Present and future. J. Med. Aromatic Plant Sci., 22: 186-191.
4. Younis, A., A. Riaz, M.A. Khan and A.A. Khan, 2009. Effect of time of growing season and time of day for flower harvest on flower yield and essential oil quality and quantity of four *Rosa* cultivars. Floriculture Ornamental Biotechnol., 3: 98-103.
5. Younis, A., A. Riaz, M.A. Khan, A.A. Khan and M.A. Pervez, 2008. Extraction and identification of chemical constituents of the essential oil of *Rosa* Species. Acta Hortic., 766: 485-492.
6. Anonymous, 2009. Essential oils: A short guide. http://www.Herbs2000.com/miss/essential_oils.htm
7. Jia, Q., W. Su, W. Peng, P. Li and Y. Wang, 2008. Anti-Diarrhoea and Analgesic Activities of the Methanol Extract and Its Fractions of *Jasminum amplexicaule* Buch.-Ham. (Oleaceae). J. Ethnopharmacol., 119: 299-304.
8. Friedman, M., P.R. Henika and R.E. Mandrell, 2002. Bactericidal activities of plant essential oils and some of their isolated constituents against *Campylobacter jejuni*, *Escherichia coli*, *Listeria monocytogenes* and *Salmonella enterica*. J. Food Prot., 65: 1545-1560.
9. Fang, S.H., G.Q. Xu, T. Xia, L.X. Li and X.C. Wan, 2003. Study on the correlation between the principal factors during scenting process for jasmine tea. J. Tea Sci., 23: 21-26.
10. Nakayama, A., K. Shinohara, S. Nakao, K. Adachi, H. Hamazaki, S. Kubo and H. Sakurai, 2004. Reactive oxygen species-scavenging activity and biological response of herb essential oils. Aroma Res., 18: 123-128.
11. Pakistan Bureau of Statistics, 2010. Pakistan statistical yearbook, 2010. Pakistan Bureau of Statistics, Islamabad, Pakistan.
12. Heath, H.B., 1981. Source Book of Flavors. Springer, New York, ISBN: 9780870553707, pp: 81-122.
13. Younis, A., 2006. Phyto-chemical analysis and potential for exploitation of hetrosis of essential oil of *Rosa* species. Ph.D. Thesis, Institute of Horticultural Sciences, University of Agriculture Faisalabad, Pakistan.
14. Paroul, N., L. Rota, C. Frizzo, A.C.A. dos Santos and P. Moyna *et al*., 2002. Chemical composition of the volatiles of angelica root obtained by hydrodistillation and supercritical CO_2 extraction. J. Essent. Oil Res., 14: 282-285.
15. Pourmortazavi, S.M. and S.S. Hajimirsadeghi, 2007. Supercritical fluid extraction in plant essential and volatile oil analysis. J. Chromatogr. A, 1163: 2-24.
16. Gunkel, W., L.C. Fraser and S.C. Bhatia, 2010. Concrete and Absolute of Jasmine and Lilac. In: Handbook of Essential Oils, Volume 1, Gunkel, W. (Ed.). CBS Publishers and Distributors Pvt. Ltd., New Delhi, ISBN: 9788123918204, pp: 31-43.
17. Grolier Inc., 1987. Academic American Encyclopedia, Volume 5. Grolier International Inc., USA., pp: 112-113.
18. Paqout, C., 1986. Determination of Refractive Index. In: Standard Methods for the Analysis of Oils, Fats and Derivatives, Paqout, C. (Ed.). Blackwell Scientific Publications, USA., pp: 23-24.
19. Steel, R.G.D., J.H. Torrie and D.A. Dickey, 1997. Principles and Procedures of Statistics: A Biometrical Approach. 3rd Edn., McGraw-Hill Co., New York, USA., ISBN: 9780070610286, pp: 134-135.
20. Thavaselvan, A. and L. Superamaniam, 2008. Extraction of Essential Oils from Jasmine Flower Using Solvent Extraction Method: A Study on Feed Ratio Effects. Universiti Malaysia Pahang, Malaysia, Pages: 64.

21. Kovacevic, M. and M. Kac, 2001. Solid-phase microextraction of hop volatiles: Potential use for determination and verification of hop varieties. J. Chromatogr. A, 918: 159-167.
22. Pop, E.G. and D. Barth, 2001. Supercritical fluid extraction of Z-sabinene hydrate-rich essential oils from Romanian mentha hybrids. Pure Applied Chem., 73: 1287-1291.
23. Marentis, R., J.T. Hsu and K. James, 2001. Supercritical fluid extraction of nutraceutical products. Proceedings of the 4th Brazilian Meeting on Supercritical Fluids EBFS 2001, October 9-11, 2001, Salvador-Bahia, Brazil, pp: 1-4.
24. Ferreira, S.R.S., Z.L. Nikolov, L.K. Doraiswamy, M.A.A. Meireles and A.J. Petenate, 1999. Supercritical fluid extraction of black pepper (*Piper nigrun* L.) essential oil. J. Supercrit. Fluids, 14: 235-245.
25. Eikani, M.H., I. Goodarznia and M. Mirza, 1999. Supercritical carbon dioxide extraction of cumin seeds (*Cuminum cyminum* L.). Flavour Fragr. J., 14: 29-31.
26. Gamiz-Gracia, L. and M.D.L. de Castro, 2000. Continuous subcritical water extraction of medicinal plant essential oil: Comparison with conventional techniques. Talanta, 51: 1179-1185.
27. Raeissi, S. and C.J. Peters, 2005. Application of double retrograde vaporization as an optimizing factor in supercritical fluid separations. J. Supercrit. Fluids, 33: 115-120.
28. Reverchon, E., G.D. Porta and D. Gorgolione, 1995. Supercritical CO_2 fractionation of jasmine concrete. J. Supercrit. Fluids, 8: 60-65.
29. Gopalakrishnan, N. and C.S. Narayanan, 1991. Carbon dioxide extraction of Indian jasmine concrete. Flavour Fragr. J., 6: 135-138.
30. Sahoo, S., 2001. Conservation and Utilization of Medicinal and Aromatic Plants. Allied Publishers Limited, India, ISBN: 9788177642216, pp: 396.
31. Younis, A., A. Mehdi and A. Riaz, 2011. Supercritical carbon dioxide extraction and gas chromatography analysis of *Jasminum sambac* essential oil. Pak. J. Bot., 43: 163-168.
32. Weiss, E.A., 1997. Essential Oil Crops. CAB International, Wallingford, UK., ISBN: 0-85199-137-8, pp: 342-361.
33. Waheed-ur-Rehman, 2006. Extraction and gas chromatographic analysis of essential oil of *Jasmine sambac*. M.Sc. Thesis, Department of Chemistry, University of Agriculture, Faisalabad.
34. Handa, S.S., S.P.S. Khanuja, G. Long and D.D. Rakesh, 2008. An Overview of Extraction Techniques for Medicinal and Aromatic Plants. In: Extraction Technologies for Medicinal and Aromatic Plants, Handa, S.S., S.P.S. Khanuja, G. Longo and D.D. Rakesh, (Eds.). International Centre for Science and High Technology ICS-UNIDO, Trieste, Italy, pp: 21-54.
35. Guenther, E., 1952. The Essential Oils. D. van Nostrand Co. Inc., Princeton, New Jersey, pp: 325-332.
36. Anonymous, 2012. Jasmine properties. M.K. Exporters, India.
37. Gilbert, A.N., R. Martin and S.E. Kemp, 1999. Cross-modal correspondence between vision and olfaction: The color of smells. Am. J. Psychol., 109: 335-351.
38. Joy P.P., J. Thomas, S. Mathew, G. Jose and J. Joseph, 2001. Aromatic Plants. In: Tropical Horticulture Vol. 2. Bose, T.K., J. Kabir, P. Das and P.P. Joy (Eds.). Naya Prokash, Calcutta, pp: 633-733.
39. Eid, R.A., L.S. Taha and M.M.S. Ibrahim, 2010. Physiological properties studies on essential oil of *Jasminum grandiflorum* L. as affected by some vitamins. Ozean J. Applied Sci., 3: 87-96.