



Interspecies variation of chemical constituents and antioxidant capacity of extracts from *Jasminum sambac* and *Jasminum multiflorum* grown in Malaysia

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ABSTRACT

The chemical constituents from various process of extraction of *Jasminum sambac* and *Jasminum multiflorum* were identified by gas chromatography–mass spectrometry to depict their interspecies differences. Major compounds identified from methanol extract of *J. sambac* were β -farnesene (52.52%), nerolidol (19.85%) and benzyl alcohol (17.56%) whereas, headspace solid phase microextraction (HS-SPME) showed linalool (35.92%), benzaldehyde (17.92%) and benzyl alcohol (10.87%) while hydrodistillation yielded β -farnesene (45.13%), α -cadinol (26.21%) and linalool (9.96%). The methanol extract of *J. multiflorum* yielded nerolidol (42.44%), benzyl benzoate (39.00%) and jasmolactone (12.02%) whereas, HS-SPME eluted nerolidol (76.56%), jasmone (15.31%) and hexyl benzoate (4.40%) while hydrodistillation yielded hexenyl benzoate (35.89%), β -farnesene (24.62%) and α -cadinol (14.30%). The methanol extracts of both *J. sambac* (0.48%) and *J. multiflorum* (1.38%) showed DPPH free radicals scavenging activities with an IC_{50} value of 208 μ g/mL and 81 μ g/mL respectively. The antioxidant properties of *Jasminum* provided a natural preservative ingredient for food and pharmaceutical products.

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1. Introduction

The genus *Jasminum* is a genus of small trees and vines of the Family Oleaceae with 600 known species. *Jasminum sambac* (*J. sambac*) and *Jasminum multiflorum* (*J. multiflorum*) are small shrubs, which can grow up to roughly 1–1.5 m in height and are widely cultivated in Asia and some parts of Europe and Africa (Rao and Rout, 2003).

J. sambac and *J. multiflorum* are the two most widely cultivated jasmine species in Malaysia and both species bloom abundantly all year round reaching its peak in March–July. The flowers are commonly used for religious purpose and its fragrance has the characteristic jasmine aroma which has demand especially in the tea industries. The perfumery industries use the absolutes extensively for its fine, sweet, fruity, and elegant notes (Edris et al., 2008). Generally, jasmine scented tea is produced by allowing tea leaves to absorb the fragrance of fresh jasmine flowers. Among the scented teas, jasmine green tea, which is reprocessed from green tea scented of *J. sambac* flowers, is the most popular around the world. In some parts of Asia, a poultice of crushed *J. sambac* flowers

are used as a lactifuge on women's breast while an infusion of the flowers are used in the treatment of pulmonary catarrh, bronchitis, and asthma (Samy et al., 2014).

Crude extracts of plants, fruits, herbs, and vegetables are becoming popular due to the presence of phytochemicals which contain considerable antioxidant capabilities and health benefits. Such benefits include prevention of heart diseases and cancer (Lörliger, 1991). Antioxidants are compounds which are capable of slowing down or hindering the oxidation of lipids and other molecules by inhibiting the initiation or propagation of oxidative chain reactions (Velioglu et al., 1998). The usage of extracts from natural sources as functional ingredients in foods, drinks, toiletries and cosmetics has gained a wide interest in consumers due to the increasing concern of potentially harmful synthetic additives (Reische et al., 1998). Natural extracts which possess a combination of pleasant taste or smell and preservative action can prevent the oxidation of lipids and spoilage by microorganisms. These unwanted reactions usually occur when a lipid or perishable organic substrate is present. These reactions will promote undesirable off-flavors, produce toxicity, loss of color, flavor and aroma that affect the shelf-life of goods (Farag et al., 1989; Hiras and Takemasa, 1998).

Few studies have been carried out on Malaysian grown *J. sambac* and *J. multiflorum*. The value of an isolate and composition of jasmine flowers is highly dependent on the techniques used for

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extraction. The objective of this study is to identify the extractable compounds from *J. sambac* and *J. multiflorum* by various extraction techniques: solvent extraction, hydrodistillation and headspace solid phase microextraction. In addition, we presented the genetic variation between the two species from different extraction techniques. The antioxidant activity of methanol extracts was also presented to show their different levels of free radical scavenging activities.

2. Materials and methods

2.1. Plant materials

Fresh blooms of *J. sambac* and *J. multiflorum* were obtained from a nursery in Sungai Buloh, Kuala Lumpur, Malaysia. The voucher specimen was deposited at the Chemistry Department Herbarium with voucher no: URL/JM/28 for *J. multiflorum* and voucher no: URL/JS/88 for *J. sambac*.

The flowers were washed with distilled water to remove dirt on the petals, and the cleaned flowers were aired at room temperature.

2.2. Chemicals and materials

All chemicals were of analytical grade, obtained from Merck (Darmstadt, Germany). Sodium sulfate was purchased from System (Shah Alam, Malaysia). The chemical reagents, namely, 1,1-diphenyl-2-picrylhydrazyl (DPPH) and butylated hydroxy toluene (BHT) were purchased from Sigma Chem. Co. (St. Louis, USA). A SPME fibre holder with a 1 cm fibre assembly coated with a 100 μ m polydimethylsiloxane (PDMS) was purchased from Supelco (Bellefonte, PA, USA).

2.3. Solvent extraction

An amount of 0.9 g of cleaned flowers were placed into a 20 mL scintillation vial (in triplicates) and extracted by maceration with 5 mL of methanol at room temperature for 24 h. After 24 h, each extract was filtered using filter paper and dried by passing through a 5 cm in length and 1 cm in width sodium sulfate column. A 1.5 mL of the extract was subjected to GC–MS analysis.

2.4. Hydrodistillation

An amount of 380 g of fresh flowers were placed in a 12 L round bottom flask which contained 6 L of distilled water and connected to a Clevenger apparatus. The mixture was heated to 100 °C for four hours. The distillate was extracted twice using 2 mL of dichloromethane. The organic layer was collected and drawn into a beaker containing anhydrous sodium sulfate for drying purposes. The concentrated extracts were analyzed using GC–MS.

2.5. Headspace solid phase microextraction (HS-SPME)

An amount of 0.7 g of fresh flowers were placed into a 20 mL vial (with triplicates) fitted with a pre-cut septum cap. The vial was then left for about 2 min at 80 °C to allow equilibration. A SPME fibre holder with a 1 cm fibre assembly coated with a 100 μ m polydimethylsiloxane (PDMS) was used. The SPME needle was then injected through the cap into the headspace of the vial, and the fibre was exposed to carry out sampling for 15 min. The SPME fibre was immediately placed and left for 7 min in the injector port of the GC–MS system.

2.6. Gas-chromatography mass-spectroscopy (GC–MS)

A Hewlett Packard HP 6890 series mass selective detector linked to a GCMS-QP 2010 Plus Shimadzu gas chromatograph was used for the identification of chemical constituents. A sample volume of 2 μ L was injected in a splitless mode into the gas chromatograph fitted with a DB-5 ms column coated with 5% phenyl 95% dimethyl arylene siloxane with film thickness of 0.25 μ m, length of 30.0 m and a diameter of 0.25 mm. Helium was used as a carrier gas with a flow rate of 58.2 mL per minute. The injector temperature was set at 250 °C. Electron impact mass spectra with ionization energy of 70 eV was recorded at the 50–600 amu mass range. The initial oven temperature was set at 50 °C and held for 2 min. This temperature was then programmed to 120 °C at 3 °C per minute and held for 3 min. Then, the temperature was programmed to 180 °C at 3 °C per minute and held for 3 min. Finally, the temperature was programmed to 280 °C at a rate of 8 °C per minute.

2.7. Kovats indices

The compounds were identified by comparing its mass spectra with NIST library and they were further confirmed with Kovats retention index (Adams, 2001). The retention index of a component is calculated by interpolation (usually logarithmic) that relates the adjusted retention times of a component to the adjusted retention times of two standards which elutes before and after the peak of interest. In the gas chromatography separation, *n*-alkanes are employed as standards where the retention index, *I* is expressed as:

$$\text{Retention index, } I = 100 \left(\left(\frac{\log X_i - \log X_z}{\log X_{(z+1)} - \log X_z} \right) + Z \right)$$

where *X* refers to the adjusted retention times, *z* is the number of carbon atoms of the *n*-alkane eluting before and (*z* + 1) is the number of carbon atoms of the *n*-alkane eluting after the peak of interest. The Kovats index uses the number of carbon atoms of a theoretical alkane which would have an adjusted retention time similar to that of the peak of interest which was evaluated under identical conditions (McNaught and Wilkinson, 1997). This enables Kovats indices to be used to confirm the peaks which were previously identified by GC–MS and by comparison of its relative retention indices with literature values (Adams, 2001).

2.8. Genetic distances

A measure of genetic differences between species or between populations within a specie is known as genetic distance and this can be determined by using Nei's (1987) standard genetic distance. This method explains that if the rate of genetic change is constant per year or generation then Nei's standard genetic distance (*D*) raises in proportion to divergence time. This method assumes that mutation and genetic drift contribute to the causes of genetic differences. The formula of genetic distances described by Nei is as follows:

$$D = - \frac{(\ln \sum X_i Y_i)}{(\sqrt{\sum X_i^2 \sum Y_i^2})}$$

where *X* and *Y* represent two different populations which have been studied and in this study *J. sambac* and *J. multiflorum*, respectively.

2.9. Methanolic extract for antioxidant assay

J. sambac and *J. multiflorum* flowers (11.08 g and 10.01 g, respectively) were extracted by maceration with 30 mL of 99.8% methanol at room temperature for 72 h. The mixture was then filtered

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