FREE RADICAL SCAVENGING ACTIVITY OF JASMINUM SAMBAC

Krishnaveni. A*
Santh Rani Thaakur1

*Department of Pharmacognosy, College of Pharmacy, Madurai Medical College, Madurai – 625 020. Tamilnadu.India.

1Professor, Institute of Pharmaceutical Technology, Sri Padmavathi Mahila Visvavidyalayam (Women’s University), Tirupati, Andhra Pradesh, India - 517502.

INTRODUCTION

Jasminum sambac is a sub erect shrub with young shoots of ovate or elliptic glabrous simple leaves, entire margin, and acute apex with opposite arrangement, grown as an ornamental shrub in gardens and cultivated throughout the tropical and subtropical parts of India (1). Leaves, roots and flowers are used as lactifuge. The whole plant is used as diuretic, emmenagogue, antihelminthic and deobstruent. Otto from flowers is used as deodorant and leaf preparations are used to treat insanity (2,3,4). The earlier literature of Jasminum sambac revealed the presence of dotricontanol, oleanolic acid, daucosterol and hesperidin and dotriacontanic acid isolated from the roots (5). In addition, the presence of glycosidic precursors such as benzyl 6-O-ß-D-xylopyranosyl ß-glucopyranoside(beta–primeveroside),2-phenyl ethyl ß primeveroside, phenyl ethyl 6-O-ß-D-rhamnoside were reported(6). Preliminary phytochemical screening reported the presence of alkaloids, glycosides and tannins from the leaves of Jasminum sambac(7,8). The plant also exhibited antilactation effect, antibacterial, antiviral, antiproliferative, anti acne and anti inflammatory effect(9-13). The present study is an attempt to investigate the antioxidant potential of Jasminum sambac.

ABSTRACT

Oxidative stress is the major role involved in the aetiology of depression. The concentrations of antioxidants and some pro-oxidative enzymes in the human brain may be involved in depression. Reduced oxidative stress correlates with the antidepressant treatment and brings the moderate clinical recovery of depression. Natural antioxidants that are present in herbs and spices are responsible for inhibiting or preventing the deleterious consequences of oxidative stress. Jasminum sambac belongs to the family Oleaceae, found in Indian gardens especially in the Southern parts of India. Essential oil from the flowers used as perfumes and antilactation. Leaves preparations are used to treat insanity hence the leaves have been selected to evaluate the antioxidant potential. The objective of the present study is to investigate the antioxidant potential of hydroalcoholic extract of leaves by various in vitro methods. Different concentrations of Jasminum sambac was used to evaluate the antioxidant effect. Antioxidant screening methods such as DPPH assay, scavenging of nitric oxide and hydrogen peroxide were determined. Total reducing power and antioxidant capacity of the hydroalcoholic extract were evaluated. Jasminum sambac showed moderate scavenging effect in the order towards the DPPH radicals (122 µg/mL), nitric oxide (173.94 µg/mL) and hydrogen peroxide (125µg/mL) when compared to ascorbic acid. The results indicate that the total antioxidant capacity (155.40 µg/mL) and its reducing power (44.28 µg/ml) was found to be activity of the crude extract of Jasminum sambac is slightly higher than that of ascorbic acid. The phytochemical tests indicated the presence of alkaloids, glycosides, tannins, and flavonoids in the hydroalcoholic extract. The antioxidant effect may be due to the phytochemicals present in it. These are known to possess potent antioxidant activity. Thus, the present research study added scientific evidence to the antioxidant potentiality of Jasminum sambac. Further formulations may be prepared and evaluated by in vivo studies.

Keywords: antioxidant, Jasminum sambac, Oleaceae

Chemicals used

1. 1-diphenyl, 2-picryl hydrazyl (DPPH), sulphanilamide, O-phosphoric acid, naphthyl ethylene diaminedihydrochloride, potassiumferricyanide, ferric chloride,, hydrogen peroxide and sodium nitroprusside.

Plant material

The leaves were collected from the foot hills of Tirumala, Tirupati, Andhra Pradesh. The leaves were authenticated by Dr. Madhav Shetty, Taxonomist, Department of Botany, S.V.University, Tirupati, Andhra Pradesh, India.

Plant Extract

The collected plant material was dried, coarsely powdered and passed through the sieve no 40. About 50 g of the powder was packed in Soxhlet extractor, defatted with petroleum ether, residue was dried and further extracted with 70% hydroalcohol until the complete exhaustion of the drug was done. The extract was evaporated and concentrated to semisolid mass (14).

Address for correspondence

Krishnaveni. A*
College of Pharmacy, Madurai Medical College, Madurai – 625 020. Tamilnadu.India.
Email: akrishnaveni72@rediffmail.com
Mobile no: 09789795091
FREE RADICALS SCAVENGING ACTIVITY

**Determination of DPPH assay**
Various concentration of the extract was added 4 ml of DPPH solution and finally made up to 5 ml with methanol, incubated for 30 minutes in dark. The absorbance of the incubated solution was recorded at 517nm (15) The experiments were conducted in triplicates and the percentage of inhibition was expressed in terms of ascorbic acid equivalents. The experiment was repeated in triplicate. The DPPH scavenging activity is calculated as:

\[
\text{1}\% = \frac{A_{\text{blank}} - A_{\text{sample}}}{A_{\text{blank}}} \times 100
\]

Where \(A_{\text{blank}}\) is the absorbance of the control \(A_{\text{sample}}\) is the absorbance of the sample

**Determination of Nitric oxide scavenging effect**
Nitric oxide was generated from sodium nitroprusside in aqueous physiological pH generates nitric oxide which interacts with oxygen to produce nitrite ions. Scavenging effect of nitric oxide with oxygen leads to the reduced production of nitrite ions. This was measured by spectrophotometrically at 546 nm (16). Various concentration of the extract of range from 40-200µg/mL prepared from saline was incubated with at 25°C for 5 hours. Control experiments were performed without test compounds but with the equivalents amount of buffer. After 5 hours, 0.5 ml of the incubated solution was removed and treated with 0.5 ml Greiss reagent containing (1% sulphanilamide, 2% O-phosphoric acid and 0.1% napthyl ethylene diaminedihydrochloride. The absorbance of the chromospheres forma during diazotization of nitrite with sulphanilamide and its subsequent coupling with napthyl ethylene diamine was read at 546 nm. The experiment was repeated in triplicate. The nitric oxide radical scavenging activity is calculated as:

\[
\text{1}\% = \frac{A_{\text{blank}} - A_{\text{sample}}}{A_{\text{blank}}} \times 100
\]

Where \(A_{\text{blank}}\) is the absorbance of the control \(A_{\text{sample}}\) is the absorbance of the sample

**Determination of hydrogen peroxide radical scavenging activity**
Different concentrations of the extract range from 40-200 µg/mL were incubated with 0.6 ml of 4mM hydrogen peroxide solution for 10 minutes. The absorbance of the solution was measured spectrophotometrically at 230nm against blank solution (17). The experiments were repeated in triplicate. The scavenging of hydrogen peroxide radical was calculated as follows:

\[
\text{1}\% = \frac{A_{\text{blank}} - A_{\text{sample}}}{A_{\text{blank}}} \times 100
\]

Where \(A_{\text{blank}}\) is the absorbance of the control \(A_{\text{sample}}\) is the absorbance of the sample

**Reducing power assay**
The reducing power of the extract was evaluated according to Oyaizu method(18). Different concentrations of the extract range from 40-200 µg/mL were treated with 2.5ml of 0.2 phosphate buffer (pH 6.6) and 2.5 ml of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 minutes, 2.5ml of trichloroacetic acid was added and centrifuged at 2000rpm for 10 minutes. About 0.5ml of the upper solution was pipette out and mixed with 2.5ml of methanol, 0.5ml of 0.1% ferric chloride and the absorbance was measured at 700nm. Increase in absorbance indicated the increased reducing power The experiments were conducted in triplicates and the reducing power was expressed in terms of ascorbic acid equivalents (µg/mg of extract).

**Total antioxidant capacity**
Different concentrations of the extract range from 40-200 µg/mL was combined with 1 ml of reagent containing 0.6M sulphuric acid, 28mM sodium phosphate and 4mM ammonium molybdenum. The tubes were capped and incubated at 95°C for 90 minutes and cooled. The absorbance of the solution was measured spectrophotometrically at 695 nm(19) The experiments were conducted in triplicates and the reducing power was expressed in terms of ascorbic acid equivalents.

**RESULTS AND DISCUSSION**
The present article draws the potentiality of *Jasminum sambac*. The antioxidant activity was determined using the free radicals released by DPPH, nitrous oxide and hydrogen peroxide released from sodium nitroprusside solution in buffer and hydrogen peroxiderespectively. In DPPH assay the inhibitory concentration was found to be122 µg/mL in comparison with ascorbic acid. The minimum inhibitory concentration of nitric oxide and hydrogen peroxide was found to be 173.94 µg/mL and 125µg/mL respectively when compared with ascorbic acid. The total antioxidant capacity and reducing power were found to be 155.40 µg/mL µg/mL and 44.28µg/mL respectively in accordance with ascorbic acid. The sequence of free radical scavenging activity was in the order: hydrogen peroxide > nitric oxide>DPPH. The extract was showed moderate reducing power and total antioxidant capacity. The extract containing phytoprinciples such as tannins, alkaloids, flavonoids,phenolic compounds, reducing sugars and proteins may be responsible for the antioxidant effect. However, the chemical constituents present in the extract, need to be investigated. The crude methanolic extract merits further experiments in vivo. Thus, the present research study added a scientific credit to the antioxidant potentiality of *Jasminum sambac* which may be due to the phytoprinciples present in it. Further research may envisaged towards the isolation of the isolation of the phytoprinciples in it.
Figure 1: DPPH scavenging of *Jasminum sambac*

Figure 2: Nitric oxide scavenging of *Jasminum sambac*

Figure 3: Hydrogen peroxide scavenging activity of *Jasminum sambac*

Figure 4: Reducing power of *Jasminum sambac*

Figure 5: Total antioxidant capacity of *Jasminum sambac*

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