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ANTIOXIDANT ACTIVITY OF *SENECIO AMPLEXICAULIS* KUNTH. ESSENTIAL OIL GROWING IN HIGH-ALTITUDE HIMALAYAN REGION

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ABSTRACT

Objective: *Senecio amplexicaulis* Kunth. Syn *Ligularia amplexicaulis* DC (Asteraceae), a deciduous shrub found in north-west (Garhwal) Himalayas of Uttarakhand (India) was collected at an altitude of around 3000 m. Light pale yellow essential oil (0.66% w/v) of *S. amplexicaulis* Kunth., obtained by hydro-distillation of roots was evaluated for antioxidant potential. **Methods:** The antioxidant and free radical activity of *S.amplexicaulis* essential oil was investigated by assessing its effect on linoleic acid peroxidation and ability to scavenge DPPH and ABTS free radicals. **Results:** The essential oil exhibited noticeable antioxidant activity in all the assays. The scavenging ability for DPPH free radicals was dose dependent with IC_{50} value of 81.6µg/ml. The oil inhibited 44.8% of the ABTS radical cations at 100µg/ml. The oil also exhibited concentration dependent effect on inhibition of lipid peroxidation. After 30h, essential oil produced 71% inhibition of lipid peroxidation at 100µg/ml, when compared to control. The antioxidant potential of essential oil was however low when compared to synthetic antioxidant, butylated hydroxy toluene. **Conclusion:** The present study reveals noticeable antioxidant potential of essential oil extracted from *Senecio amplexicaulis* Kunth. and suggests a need for further exploration.

KEYWORDS: Senecio, lipid peroxidation, free radical, Asteraceae.

1. INTRODUCTION

The genus Senecio is distributed throughout the world and consists of more than 1500 plant species. The plants of this genus have been studied extensively because of the traditional medicinal uses associated with them. Phytochemical investigation and biological evaluation of the plants of this genus have revealed compounds with diverse biological activities viz. cytotoxic, antihyperglycemic, antimicrobial, anti-inflammatory and antioxidant.[1,2] Chemical investigation and biological evaluation of the essential oils of plants of this genus has also been reported.[3-10]

Essential oils are liquid blends of volatile compounds obtained from plants.[11] Essential oils are defined as the products obtained from different parts of plants through distillation by steam distillation, hydro distillation and pressing the citrus fruit pericarp techniques.[12] These are aromatic oily liquids, volatile, characterized by a strong odour, rarely coloured, and generally with a lower density than that of water. They can be synthesized by all plant parts (flowers, buds, seeds, leaves, twigs, bark, herbs, wood, fruits and root) and therefore extracted from these parts, where they are stored in secretory cells, cavities, canals, epidermic cells or glandular trichomes.[13] Many essential oils have antioxidant properties and there is a growing interest in essential oils as natural antioxidants because some synthetic antioxidants like butylated hydroxyl anisole (BHA) and butylated hydroxyl toluene (BHT) are suspected to harm human health.[14]

Senecio amplexicaulis Kunth. Syn *Ligularia amplexicaulis* DC (Asteraceae) is a deciduous shrub found in north-west (Garhwal) Himalayas of Uttarakhand (India).[15, 16] The chemical composition and biological activity of the essential oil from the roots of *S. amplexicaulis* have been previously reported in our paper.[17] The traditional claim associated with *Senecio* species and lack of scientific studies regarding antioxidant potential of *Senecio amplexicaulis* essential oil prompted us to perform this study. The objective of the present study was to evaluate antioxidant potential of essential oil from the roots of *Senecio amplexicaulis*. To the best of our knowledge, this is first report of antioxidant activity of *Senecio amplexicaulis* essential oil collected from north-west Himalayas, India.

2. MATERIAL AND METHODS

2.1 Chemicals

Anhydrous sodium sulfate, DPPH, ABTS, potassium persulfate, linoleic acid, tween 20, BHT were obtained

from Hi Media Chemicals or Sigma. All other reagents used were of analytical grade.

2.2 Plant material and essential oil extraction

Senecio amplexicaulis Kunth. was collected (May-Jun) from north-west Himalayas (Uttarakhand, India) and identified by Botanical Survey of India, Dehradun, Uttarakhand, India with voucher no. 42099. Freshly collected roots were shade dried and cut into small pieces. The chopped roots were subjected to hydrodistillation for 3h using a Clevenger apparatus. The extracted light pale yellow essential oil (0.66% w/v) was dried over anhydrous sodium sulphate to remove traces of water. The oil was stored in sealed vial, stored under refrigeration (4°C), protected from light; until used for analysis.

2.3 DPPH (2, 2-diphenyl-1-picrylhydrazyl) free radical scavenging activity

A DPPH methanolic solution was prepared at a concentration of 40µg/ml. Essential oil (0.3ml of each dilution of oil in methanol) was added to stock DPPH solution (2.7ml) in a test tube. After 60min, the absorbance was measured at a wavelength of 517nm.[18] The antioxidant activity was calculated as percentage inhibition of DPPH, using the following equation.

% Inhibition of DPPH free radical =

$$\frac{Ablank - Asample}{Ablank} * 100$$

2.4 ABTS [2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid] scavenging activity

ABTS free radical was produced by reacting 2mM ABTS aqueous solution with 2.45mM potassium persulfate, stored in the dark for 4h. Prior to assay, the ABTS stock solution was diluted in ethanol and equilibrated at 30°C to give an absorbance of 0.700 ± 0.02 at 734 nm. Then, to 1 ml of diluted ABTS solution was added 3ml of essential oil solution in ethanol at different concentrations (20-100µg/ml). After thirty minutes, absorbance was read at 734 nm for each concentration. Solvents blanks were run in each assay.[19] Decrease in absorbance indicates ABTS radical scavenging ability of the essential oil or BHT.

2.5 Total antioxidant activity determination

The antioxidant activity of essential oil was determined according to the ferric thiocyanate method (FTC) using linoleic acid emulsion. Different concentrations of essential oil (50 and 100µg/ml) in 2.5ml sodium phosphate buffer were added to 2.5ml of linoleic acid emulsion in sodium phosphate buffer. For this, 5ml of the linoleic acid emulsion was prepared by mixing and homogenising 15.5µl of linoleic acid, 17.5 mg of tween-20 and 5 ml phosphate buffer (pH 7.0). On the other hand, 5ml of control consisted of 2.5ml of linoleic acid emulsion and 2.5ml sodium phosphate buffer (pH 7.0). The mixed solution (5ml) was incubated at 37°C in dark. After the mixture was stirred for 3 min, the peroxide value was determined by reading the absorbance at 500 nm in a spectrophotometer, after reaction with $FeCl_2$ and thiocyanate at intervals during incubation.[20] High absorbance value indicates greater linoleic acid oxidation. The inhibition percentage of lipid peroxidation in linoleic acid emulsion was calculated by the following equation.

$$\% \text{ Inhibition} = \left[\frac{Ac - As}{Ac}\right] * 100$$

Where, Ac is the absorbance of control and As is the absorbance of sample.

2.6 Statistical Analysis

Experimental results are expressed as mean ± SD of three measurements. *P* values <0.05 were regarded as significant and *P* values <0.01 very significant.

3. RESULTS AND DISCUSSION

Plants are well known to have naturally occurring substances possessing antioxidant activity. Essential oils of many higher plants have been evaluated for antioxidant activity. The antioxidant and free radical activity of *S.amplexicaulis* essential oil was investigated by assessing its effect on linoleic acid peroxidation and ability to scavenge DPPH and ABTS free radicals.

DPPH free radical scavenging method is widely used for evaluation of antioxidant activity of natural products. Scavenging of DPPH radical involves hydrogen atom transfer process.[21] The study revealed the antioxidant potential of essential oil as evidenced by the decolourization of DPPH free radicals to the yellow coloured diphenylpicrylhydrazine. The essential oil was evaluated for scavenging ability in the range of 20-100µg/ml. At 100µg/ml, the essential oil exhibited 59% inhibition of DPPH free radicals (Fig. 1). The free radical scavenging effect was dose dependent and the IC_{50} value was found to be 81.6µg/ml. A low IC_{50} value is associated with high antioxidant activity.

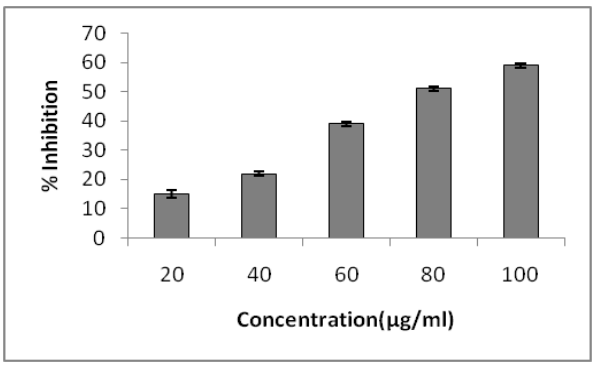

Fig. 1: DPPH radical scavenging ability of *S. amplexicaulis* essential oil.

The essential oil was also evaluated for the ability to scavenge ABTS free radicals. ABTS radical cation, a blue/green chromophore is generated by the reaction of ABTS and potassium persulfate. Addition of antioxidant reduces the blue/green ABTS radical cation chromophore into a colourless form. This method is

suitable for assessing the antioxidant activity of essential oil. ABTS cation radicals are more reactive than DPPH radicals and the quenching reaction involves electron transfer process.[21] The antioxidant activity of essential oil was evaluated in the range of 20-100µg/ml. The ABTS radical scavenging ability increases with increase in concentration of essential oil (Fig. 2). At 100µg/ml, the essential oil inhibited 44.8% of the ABTS radical cations. The scavenging effect was however very low when compared to synthetic antioxidant butylated hydroxyl toluene (BHT).

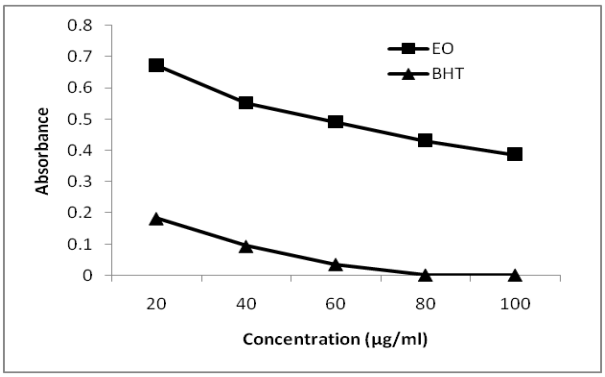

Fig. 2: ABTS radical scavenging ability of *S. amplexicaulis* essential oil.

The essential oil was also evaluated for total antioxidant activity in linoleic acid by using ferric thiocyanate (FTC) method at concentration of 50 and 100µg/ml. In this assay, hydroperoxide produced by oxidation of linoleic acid added to the reaction mixture was indirectly measured.[19] In this method, during the linoleic acid peroxidation, peroxides are formed and this leads to oxidation of Fe^{+2} to Fe^{+3} ions, which form a complex with thiocyanate that can be estimated by measuring absorbance at 500nm. The oil exhibited noticeable effect on lipid peroxidation as evidenced by lower absorbance in comparison to control (Fig. 3). The oil also exhibited concentration dependent effect on inhibition of lipid peroxidation. After 30h, essential oil produced 71% inhibition of lipid peroxidation at 100µg/ml, when compared to control.

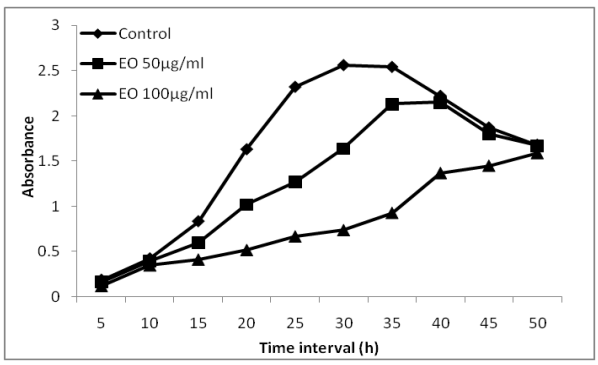

Fig. 3: Total antioxidant activity of *S. amplexicaulis* essential oil as determined by FTC method.

4. CONCLUSION

Essential oil of *Senecio amplexicaulis* Kunth. was evaluated for antioxidant activity in linoleic acid using ferric thiocyanate method, DPPH and ABTS free radicals scavenging ability. The GC-MS analysis of essential oil of *S.amplexicaulis* collected from north-west Himalayas has reported abundance of monoterpene hydrocarbons with marker compounds as α-phellandrene (48.57%), o-cymene (16.80%) and β-ocimene (7.61%).[17] The present study revealed noticeable antioxidant potential of *S.amplexicaulis* essential oil that can be attributed to its phytoconstituents. The essential oil was able to scavenge the DPPH free radicals to a fair extent as evident by its IC_{50} of 81.6µg/ml. Similarly, the oil inhibited 44.8% of ABTS radical cations at 100µg/ml. The total antioxidant activity determination in linoleic acid using ferric thiocyanate method also revealed antioxidant potential and ability of essential oil to inhibit lipid peroxidation. In conclusion, the essential oil exhibited antioxidant potential which was low when compared to synthetic antioxidant, BHT. Despite this, the results are encouraging as obtained by naturally occurring essential oil wherein the combined effect of its constituents may have produced deterring or synergistic effect on the antioxidant activity.

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CONFLICTS OF INTEREST

Declared none.

REFERENCES

1. Tundis R, Monica L, Statti GA, Brake AM, Menchini F, Houghton PJ. Bioactive extracts from Senecio samnitum Huet. Nat Prod Res, 2006; 20: 265-269.
2. Yang Y, Zhao L, Wang YF, Chang ML, Huo CH, Gu YC, Shi QW, Kiyot H. Chemical and pharmacological research on plants from the genus Senecio. Chem Biodiver, 2011; 8: 13–22.
3. Belaunde AG, Sandoval JG, De Martino L, Senatore F, De-Feo V. Chemical composition of essential oils of Senecio nutans Sch. Bip. (Asteraceae). J Essential Oil Bearing Plants, 2007; 10: 332-338.
4. Chalchat JC, Maksimovic ZA, Petrovic SD, Gorunovi MS. Essential oil of Senecio squalidus L., Asteraceae. J Essential Oil Res, 2004; 16: 227-228.
5. Kahriman N, Tosun G, Terzioglu S, Karaoglu SA, Yayli N. Chemical composition and antimicrobial activity of essential oil from flower, leaf, and stem of Senecio pandurifolius. Rec Nat Prod, 2011; 5: 82-91.
6. Maggio A, Venditti A, Senatore F, Bruno M, Formisano C. Chemical composition of the essential oil of Jacobaea maritima (L.) Pelser & Meijden and Jacobaea maritima subsp. bicolor (Willd.) B. Nord. & Greuter (Asteraceae) collected wild in Croatia and

Sicily, respectively. Nat Prod Res, 2015; 29: 857–863.
7. Nori-Shargh D, Raftari S, Deyhimi F. Analysis of the essential oil of Senecio vernalis Waldst. & Kit. from Iran. Flavour Frag J, 2008; 23: 357-359.
8. Sharma P, Shah GC. Composition and antioxidant activity of Senecio nudicaulis Wall. ex DC. (Asteraceae): a medicinal plant growing wild in Himachal Pradesh, India. Nat Prod Res, 2015; 29: 883–886.
9. Tundis R, Menichini F, Loizzo MR, Bonesi M, Solimene U. Studies on the potential antioxidant properties of Senecio stabianus Lacaita (Asteraceae) and its inhibitory activity against carbohydrate-hydrolysing enzymes. Nat Prod Res, 2012; 26: 393–404.
10. Usta A, Ucuncu O, Cansu TB, Terzioglu S, Yaylı N. Chemical composition of essential oil from flower of Senecio vernalis and Senecio platyphyllus var. platyphyllus. Asian J of Chem, 2009; 21: 6369-6374.
11. Sati SC, Sati N, Ahluwalia V, Walia S, Sati OP. Chemical composition and antifungal activity of Artemisia nilagirica essential oil growing in Northern hilly areas of India. Nat Prod Res, 2013; 27: 45-48.
12. Milene AA, Maria-das GC, Juliana-de A, Lucilene F, Silva MLT, Juliana M et. al. Chemical Composition and Antioxidant Activity of Essential Oils from Cinnamodendron dinisii Schwacke and Siparuna guianensis. Antioxid, 2013; 2: 384-397.
13. Maria GM. Antioxidant and Anti-Inflammatory Activities of Essential Oils: A Short Review. Molecules, 2010; 15: 9252-9287.
14. Amorati R, Foti MC, Valgimigli L. Antioxidant activity of essential oils. J Agriculture and Food Chem, 2013; 20: 10835-10847.
15. Gaur RD. Flora of district Garhwal north-west Himalayas, 1st ed. Srinagar Garhwal, India: Trans Media Publishers, 1999.
16. Naithani BD. Flora of the District Chamoli Garhwal, Dehradun, Botanical survey of India, 1984.
17. Rajendra S, Vivek A, Pratap S, Naresh K, Omprakash S, Nitin S. Antifungal and phytotoxic activity of essential oil from root of Senecio amplexicaulis Kunth. (Asteraceae) growing wild in high-altitude Himalayan region. Nat Prod Res 2015. http://dx.doi.org/10.1080/14786419.2015.1079910.
18. Tepe B, Daferera D, Sokmen A, Sokmen M, Polissiou M. Antimicrobial and antioxidant activities of the essential oil and various extracts of Salvia tomentosa Miller (Lamiaceae). Food Chem, 2005; 90: 333–340.
19. Ilhami G, Mahfuz E, Hassan Y, Aboul E. Antioxidant activity of clove oil–A powerful antioxidant source. Arabian J of Chem, 2012; 5: 489–499.
20. Ilhami G, Gungor SI, Sukru B, Mahfuz EO, Irfan K. Comparison of antioxidant activity of clove (Eugenia caryophylata Thunb) buds and lavender (Lavandula stoechas L.). Food Chem, 2004; 87: 393–400.
21. Kaviarasan S, Naik GH, Gangabhagirathi R, Anuradha CV, Priyadarsini KI. In vitro studies on antiradical and antioxidant activities of fenugreek (Trigonella foenum graecum) seeds. Food Chem, 2007; 103: 31–37.