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Research Article

UV Absorbing Property of *Ageratum conyzoides* Linn Leaves: Effects of Temperature and Time on Extraction Process

Tanaya Ghosh¹, Prasenjit Mitra² and Prasanta Kumar Mitra^{1*}

¹Department of Medical Biotechnology, Sikkim Manipal University, Sikkim Manipal Institute of Medical Sciences, Gangtok, Sikkim, India ²Department of Biochemistry, All India Institute of Medical Sciences (AIIMS),

Jodhpur, Rajasthan, India

*Corresponding Author: Prasanta Kumar Mitra, Professor and Head, Department of Medical Biotechnology, Sikkim Manipal University, Sikkim Manipal Institute of Medical Sciences, Gangtok, Sikkim, India. Received: February 19, 2020 Published: February 27, 2020 © All rights are reserved by Prasanta Kumar Mitra., *et al.*

Abstract

Recently we have noted anti solar activity of ethanol extract of *Ageratum conyzoides* Linn (*A. conyzoides* L.) leaves. We did extraction of the plant leaves to establish this pharmacological activity. As it is known that pharmacological activity of a plant depends on temperature and time of extraction process, aim of the present work was to see effects of temperature and time of the extraction process on anti solar activity of ethanol extract of *A. conyzoides* L. leaves. *A. conyzoides* L. leaves were collected and identified by the taxonomist. Ethanol extracts of the leaves were prepared separately at temperatures 20, 37, 55, 70 degree centigrade for 10, 15, 30, 50 minutes. Anti solar activity of different extracts was checked by a spectrophotometer taking absorption in UV region (200 - 400 nm) at 10 nm intervals. Results showed that ethanol extract of *A. conyzoides* L. leaves at 370C for 15 minutes showed maximum anti solar activity. These conditions may be maintained for isolation work of the anti solar compound from *A. conyzoides* L. leaves. *Keywords: Ageratum conyzoides* Linn. Leaves; Solvent Extractions; Temperature; Time; Anti Solar Activity

Introduction

Extraction process is carried out to demonstrate pharmacological activity of plants and/or to isolate active ingredients from plants and other sources. There are several techniques of extraction process. These include hot continuous extraction (Soxhlet), digestion, decoction, infusion, counter current extraction, maceration, percolation, sonication, microwave-assisted extraction etc. In all these techniques temperature has a considerable role on the rate of extraction. Hamzah., et al. studied effect of extraction temperature on the extraction of phenolic compounds from Orthosiphon stamineus leaves. It was found out that a temperature of 160°C is required for optimum extraction of phenolic compounds from *O. stamineus* leaves [1]. While studying effect of temperature on extraction process of total phenolic content of henna (Lawso*nia inermis*) stems Tan., *et al.* observed that 55°C temperature was most suitable for extraction process to collect maximum amount of phenolic compounds from henna [2]. Wingard and Phillips studied effect of temperature on the rate of extraction of crude oils from vegetable oil seeds with solvents and noted that rate of extraction varied with temperature [3].

Sulaiman., *et al.* studied effect of time on extraction of phenolic compounds vis-a-vis anti-radical activity of *Clinacanthus nutans* Lindau leaves. Authors observed that 120 min extraction is needed to get maximum phenolic compounds thereby maximum anti-radical activity of *C. nutans* Lindau leaves [4]. Effect of duration time of maceration on nitrate content of *Vernonia cinerea* (L.) was studied by Chaowalit and Chitradee. Authors showed that sixty min macer-

ation was needed to get maximum nitrate from *V. cinerea* [5]. Mahmoud., *et al.* showed that forty five min extraction was needed to get maximum antioxidant activity of one species of Lavandula [6].

A. conyzoides L. (family, *Asteraceae*) is a medicinal plant. The plant is native to tropical America, especially Brazil and considered an invasive weed in many other regions. *A. conyzoides* L. is found throughout India in garden areas, waste places and in the middle hill up to the height of 6000 ft [7]. The plant is known by different names - goat weed (English), jangli pudina (Hindi), helukasa (Kanada), kattappa (Malayalam), vosadi (Marathi), visamustih (Sanskrit), pumpul (Tamil), pokapanthi (Telugu) etc. Throughout year the plant gives flower, purple white flower appears [8].

A. conyzoides L. is described in Ayurveda, Charaka Samhita and Sushruta Samhita as emetic, purgative, febrifuge, antispasmodic and antiasthmatic. In different folkloric system of medicine the plant is used to treat pneumonia, wounds, chronic ulcer, boils, frontal headache, malaria, stomach ailments, rheumatism, colic, sore throat, skin disorders, sleeping sickness etc. [9]. In traditional medicine leaves and root of *A. conyzoides* L. are widely utilized - root juice as antibiotic, leaf juice as eye lotion [10].

Many phytochemicals are identified and characterized in different parts of *A. conyzoides* L. Few of them are, 6-(1-ethoxyethyl)-7-methoxy-2,2-dimethylchromene, 5,6,7,8,3',4',5'-heptamethoxyflavone (5'-methoxynobiletin), 5,6,7,3',4'5'-hexamethoxy flavone, 6-(1-hydroxy ethyl)-7-methoxy-2,2-dimethylchromene, 4'-hydroxy-5,6,7,8,3'-pentamethoxyflavone, coumalic acid, pro-

tocatechuic acid, 4'-hydroxy-5,6,7,8,3',5'-hexamethoxyflavone, 5,6,7,8,3'-pentamethoxy-4', 5,6,7,5'-tetramethoxy-3',4'-methylenedioxyflavon, etc. [11].

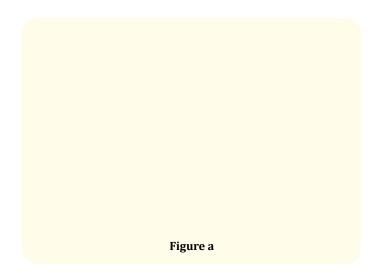
A. conyzoides L. possesses a wide range of pharmacological activities. These include, antioxidant, antimicrobial, anti-inflammatory, antidiabetic, antitumor, anthelmintic, antiulcerogenic, hepatoprotective, anticonvulsant, radioprotective, gastroprotective, antiprotozoal, analgesic, cytotoxic allelopathic, insecticidal, haematopoietic, wound healing, anti-cataleptic etc. [12].

Recently, we have noted anti solar activity of ethanol extract of *A. conyzoides* L leaves. Paper is under communication. Aim of the present work was to see effects of temperature and time on the extraction process to get maximum anti solar activity of *A. conyzoides* L leaves.

Methodology

Plant material

A. conyzoides L. leaves were collected from the medicinal plants garden of the University of North Bengal, Siliguri (26041'30.9984" N, 88027'4.5756" E, elevation, 410 ft), Dist. Darjeeling, West Bengal, India sometimes in the month of April, 2019 in morning hours (9 - 10 AM). Leaves were authenticated by the taxonomist of the department of Botany of the University of North Bengal, Siliguri. A voucher specimen (No. SM-MB-08) was kept in the department of Medical Biotechnology, Sikkim Manipal Institute of Medical Sciences of Sikkim Manipal University, Gangtok, Sikkim, India for future references.



Test drug

Leaves of *A. conyzoides* L. were washed thoroughly under tap followed by distilled water. Leaves were then shed dried and powered. The powder, used as test drug, was stored desiccated at 40C until further use.

Solvent extraction

Test drug (50g) was extracted with 500 ml of ethanol. Ethanol was chosen as solvent because in our earlier experiments we found

that ethanol extract of *A. conyzoides* L. leaves had maximum anti solar activity.

Effect of temperature on extraction process

In separate experiments extraction processes were carried out at 20, 37, 55, 70 degree centigrade.

Effect of time on extraction process

Extraction processes were done separately for 10, 15, 30, 50 minutes.

The extract was filtered and the filtrate evaporated to dryness in vacuo with rotary evaporator. This was applied separately for all extracts. Brown masses obtained.

Anti solar activity

10 mg of this mass was dissolved in 100 ml distilled water. The solution was processed in a spectrophotometer for UV ray absorption at the range of 200 - 400 nm at 10 nm intervals

Chemicals

Chemicals required for the study were purchased from Loba Chem. Lab, Himedia Lab, India and from Merck, Germany

Statistical analysis

All experiments were conducted thrice. Data were analysed statistically by SPSS 20. The statistical significance between UV absorption spectra of different extracts was evaluated with Duncan's multiple range test (DMRT). 5% were considered to be statistically significant [13].

Results and Discussion

Effect of temperature on extraction process for determination of anti solar activity of *A. conyzoides* L. leaves is shown in table 1. UV ray absorptions of ethanol extract of *A. conyzoides* L. leaves at 20°C for 15 min (as in earlier studies we kept 15 minutes as extraction time) were 0.95, 0.71, 0.63, 0.55, 0.46 at 200 nm, 250 nm, 300 nm, 350 nm and 400 nm respectively. For temperature 370C under same condition values came 1.3 (200 nm), 0.98 (250 nm), 0.88 (300 nm), 0.72 (350 nm) and 0.60 (400 nm). UV ray absorptions of ethanol extract of *A. conyzoides* L. leaves at 550C for 15 min were 1.29, 0.99, 0.86, 0.73, 0.58 at 200 nm, 250 nm, 300 nm, 350 nm and 400 nm respectively. For temperature 700C under same conditions values came 1.31 (200 nm), 0.95 (250 nm), 0.82 (300 nm), 0.74 (350 nm) and 0.61 (400 nm).

Effect of time on extraction process for determination of anti solar activity of A. conyzoides L. leaves is shown in table 2. UV ray absorptions of 10 minutes at 37°C (as in this experiment 37°C was found optimum) ethanol extract of A. conyzoides L leaves at 200 nm, 250 nm, 300 nm, 350 nm and 400 nm were 1.0, 0.81, 0.76, 0.70, 0.52 respectively. For 15 min extraction time under the same condition values came 1.4 (200 nm), 0.95 (250 nm), 0.86 (300 nm), 0.75

Solvent	Temperature (Degree centigrade) Time: 15 minutes (Fixed)	Anti solar activity (absorptions at 200/ 250/ 300/ 350/ 400 nm)
Ethanol	0	0.95/ 0.71/ 0.63/ 0.55/ 0.46
	37	1.3/ 0.98/ 0.88/ 0.72/ 0.60
	55	1.29/ 0.99/ 0.86/ 0.73/ 0.58
	70	1.31/ 0.95/ 0.82/ 0.74/ 0.61

Table 1: Anti solar activity of ethanol extract of *A. conyzoides* L.leaves : Effect of temperature on extraction process.

(350 nm) and 0.64 (400 nm). UV ray absorptions of 30 minutes at 370C ethanol extract of *A. conyzoides* L. leaves at 200 nm, 250 nm, 300 nm, 350 nm and 400 nm were 1.4, 0.92, 0.85 0.76, 0.63 and for 50 min at 370C extraction time the values came 1.4 (200 nm), 0.95 (250 nm), 0.87 (300 nm), 0.75 (350 nm) and 0.65 (400 nm).

Solvent	Time (minutes)Tem- perature: 37ºC (Fixed)	Anti solar activity (absorp- tions at 200/ 250/ 300/ 350/ 400 nm)
Ethanol	10	1.0/ 0.81/ 0.76/ 0.70/ 0.52
	15	1.4/ 0.95 0.86/ 0.75/ 0.64
	30	1.4/ 0.92/ 0.85/ 0.76/ 0.63
	50	1.4/ 0.95/ 0.87/ 0.75/ 0.65

Table 2: Anti solar activity of ethanol extract of *A. conyzoides* L.leaves : Effect of time on extraction process.

Effect of temperature of extraction process on anti solar activity of ethanol extract of *A. conyzoides* L. leaves is shown in figure 1. *A. conyzoides* L. leaves when extracted with ethanol at 20°C, 37°C, 55°C and 70°C separately absorbed UV radiations at 200 nm, 250 nm, 300 nm, 350 nm and 400 nm wave lengths. Maximum absorption for all extracts was found at 200 nm. Ethanol extract of *A. conyzoides* L. leaves done at 37°C had more UV ray absorption capacity than that of 20°C extract but the results were not statistically significant. Ethanol extracts of *A. conyzoides* L. leaves done at 55°C and 70°C had more or less same UV ray absorption capacity when compared to that of ethanol extract done at 40°C (Figure 2). **Figure 2:** Comparison of anti solar activity of ethanol extract of *A. conyzoides* L. leaves: Effect of temperature. on extraction process.

Effect of time of extraction process on anti solar activity of ethanol extract of *A. conyzoides* L. leaves is shown in figure 3. A. conyzoides L. leaves when extracted with ethanol for 10 min, 15 min, 30 min and 50 min separately exert anti solar activity. All extracts absorbed UV rays at 200 nm, 250 nm, 300 nm, 350 nm and 400 nm wave lengths. Maximum absorption, however, was found at 200 nm. It was also found that UV ray absorption values of 15 min ethanol extract of *A. conyzoides* L. leaves was comparatively higher than that of 10 min extraction but the values were not statistically significant. Ethanol extract of A. conyzoides L. leaves done for 30 min and 50 min had more or less same UV ray absorption capacity when compared to that of ethanol extract done for 15 minutes (Figure 4).

Figure 3: Anti solar activity of ethanol extract of *A. conyzoides* L. leaves : Effect of time of extraction process.

Figure 1: Anti solar activity of ethanol extract of A. conyzoides L.leaves: Effect temperature on extraction process.

Figure 4: Comparison of anti solar activity of ethanol extract of *A. conyzoides* L. leaves: Effect of time on extraction process.

Reports are available on effect of time and temperature on extraction process. Terblanche., et al. while conducting screening of variables influencing extraction yield of Cotyledon orbiculata found that the optimum conditions for the aqueous extraction design were extraction temperature 30°C, pH 8.99 and extraction time 240 minutes [14]. Hossain., et al. studied effect of temperature and extraction process on antioxidant activity of various leaves crude extracts of Thymus vulgaris and noted that the yield of extraction was increasing with increasing temperature [15]. Jahangiri., et al. while studying effect of temperature and solvent on the total phenolic compounds extraction from leaves of Ficus carica observed that percentage extraction could be increased using a higher temperature of 80°C [16]. Norshazila., et al. studied effect of time and temperature on extraction of pumpkin carotenoids and found that optimum time and temperature for the extraction process are 12 hours and 30°C respectively [17]. While experimenting on the effect of ethanol concentration, extraction time and extraction temperature on the recovery of phenolic compounds and antioxidant capacity of Centella asiatica extracts Chew., et al. noted that optimal conditions for phenolic recovery were 40% ethanol for 60 min at 65°C [18]. Mitra., et al. showed that anti solar activity of Costus seciosus leaves of Sikkim Himalayas depends on time and temperature of the extraction process [19]. In the present work we found that ethanol extract of A. conyzoides L leaves for a period of 15 min at temperature 370C had maximum UV ray absorption capacity.

It is known that biological activity of medicinal plants varies with season [20-22]. We are now interested to see the effect of season on solar UV radiation absorption by *A. conyzoides* L leaves. Work is now going on in our laboratory in this direction.

Conclusion

Present study confirmed extraction time and temperature for preparation of ethanol extract of *A. conyzoides* L leaves required for maximum UV radiation absorption. The conditions may be applied to get maximum UV radiation absorption by *A. conyzoides* L leaves.

Recommendation

Ethanol extract of *A. conyzoides* L. leaves for a period of 15 min at temperature 37^oC may be used in preparation of sun screen lotions to protect humans from UV radiation.

Acknowledgement

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Conflict of Interest

Nil.

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