ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Assessment of Sun Protection Factor From The Leaf Extract Of *Desmodium gangeticum* (L). DC.

Moumita Das[1], Sumita Mondal[2], Swastika Banerjee[1], and Abhijit Bandyopadhyay[1]*.

[1]UGC- CAS (Phase II) Department of Botany, The University of Burdwan,Burdwan- 713104, West Bengal, India.
[2]Vivekananda Mahavidyalaya, Burdwan- 713103, West Bengal, India.

ABSTRACT

The present study focused on evaluation of photo-protective activity of aqueous leaf-extract of *Desmodium gangeticum* (L). DC. The extract was subjected to UV Vis Spectrophotometer for obtaining spectral pattern. The spectral pattern was indicative of presence of such phenolic derivatives in the leaves which are known to have sun-protective property. FT IR result also speaks in the same way i.e. presence of wave numbers such as 3334.92, 3298.28 and 3290.56 cm^{-1}characteristic phenolic O-H stretched with the aliphatic groups. Moreover leaf extract possess high amount of vitamin C content (52.6 mg/l) which inhibits the sunburn. Presently in pharmaceutical research, sunscreen and sun blocker are given the top most priority to address increasing exposure to UV radiations. Extensive efforts are on the way to explore, isolate, characterize and market effective sunscreen compounds especially from plant sources. The potency of sunscreen is expressed in terms of SPF (sun protection factor) value. In the present study leaf extract of *Desmodium gangeticum* , a plant of the family Fabaceae, shows SPF value of 7.276, thus speaking of its ability to effectively block UV B by 83%- 88%. As such, the extract seems to be potent compound, which demands extensive study with clinical correlation for developing *Desmodium gangeticum* based sunscreen.

Keywords: Photoprotective activity, *Desmodium gangeticum*, Sunburn, SPF.

Corresponding author

ISSN: 0975-8585

INTRODUCTION

Our planet earth receives UVR which is classified into 3 groups namely UV A, UV B and UV C from the sun, all of the UV-C and the majority of UV-A and UV-B get filtered by the ozone layer. Ozone is made up by triatomic oxygen molecule found mainly in the stratospheric region of atmosphere, which is approximately 10.2–40.1 km above the Earth's surface. It is repeatedly being revived from O_2 through the UV dependent ozone-oxygen cycle. Free radicals such as chlorine and bromine atoms shift the cycle to develop more O_2 than O_3 which results ozone depletion. As we all know that emission of chlorofluorocarbons (CFCs) enormously increases the concentration of these free radicals which finally leads to the depletion of the ozone layer. Though the ozone depletion and climate change are separate entities but they are linked, both have the potential to increase the rate of skin cancer through different means. Ozone depletion leads to an increase in skin cancers and cause to feel anxious that it is still rising day by day. Warmer, drier weather is likely to encourage people to spend more time outdoors which leads to increase their exposure to UVR. The increasing effect results the rate of skin cancer brought about by behavioral change rather than environmental change [2].

Both the energy output of the sun and the transmission properties of the atmosphere regulate the quality and quantity of UV radiation (UVR) on the earth surface. From a biological perspective, UVB radiation has greater impact on terrestrial ecosystem and exposure levels of UVB radiation on the earth surface is largely controlled by ozone. Behavior associated with climate change is supposed to be largest determinant of sun exposure, which leads to skin cancer [4,5]. It has been postulated that the long-term elevation of temperature by 2°C, as an outcome of climate change, may increase the carcinogenic efficacy of solar UV by 10% [15].

The normal responses of the skin to UVR can be classified into two major groups i.e Acute effects includes Sunburn, Tanning, Immediate pigment darkening, Delayed tanning and chronic effects includes Photo aging, Carcinogenesis (Non melanoma skin cancer, Malignant melanoma) [7, 8].

Now a day's environmental pollutants and UV radiation causes oxidative skin damage by releasing free radicals in extreme level. Free radicals leads to premature signs of aging, sunburn, skin diseases and even skin cancer. But vitamin C acts as a potent antioxidant by neutralizing those free radicals as well as by enhancing the DNA damage repair system activity and stimulating collagen production. In another research, it has been established that both Vitamin C and E acts as complementary factors to each other and increase SPF's stability [14].

In view of this, the present work was undertaken to evaluate the Sun protection factor of leaf extract of *Desmodium gangeticum*. Shalparni is the common name of *Desmodium gangeticum*, a 60.96 – 121.90 cm high erect perennial herb with woody stem, branches covered with soft hairs. Frequently occurs in the forest and waste lands of India, from the plains and Western Ghats, and up to 1500 m in the north up to Sikkim. The plant show wide distribution throughout the world such as tropical Africa, Indian subcontinent, China, Japan, Myanmar, Thailand, Cambodia, Laos, Vietnam, Malaysia, Indonesia, Philippines, Australia.

The plant has numerous curative properties such as Antioxidant, Anti–inflammatory, Anti–nociceptive, Anti–rheumatic, Astringent, Antifebrile, Antihelmintic, Diuretic, Immunostimulant, Laxative, Nervine tonic, Tranquilizer.

Leaf paste is applied tropically to cure eczema. Leaves boiled along with leaves of neem, tulsi and black pepper in water, and then filtered to drink 1-2 times a day for a week to cure Skin related disease [10].

The literature speaks the immense use of leaves in curing different disease on the basis of this present work was undertaken. The leaf of *Desmodium gangeticum* being a polyvalent drug is likely to show synergy. In view of this, prevalence of SPF in leaves was tested which is likely to afford opacity to UV radiation. This attribute of leaves of different species is presently in the focus of scientists for appropriate selection of species for pharmaceutical preparation of sun-screen and lotions.

ISSN: 0975-8585

Fig 1: Fruit bearing twig [7]

Fig 2: Inflorescence [8]

MATERIALS AND METHODS

Mature disease free leaves of *Desmodium gangeticum* (Fabaceae) was collected from Golapbag campus of Burdwan University.

The plant is identified by Prof. Ambarish Mukherjee and voucher specimen was kept in the herbarium of Burdwan University (**BURD)** for future reference. The mature leaves were washed thoroughly under running tap water and distilled water, blotted dry and then dried by keeping inside hot air oven at 50°C temperature used for immediate extraction.

Sample preparation

The dried powdered leaves of the plant (10 g) were extracted in 100 ml of 95% ethanol. The resultant extract was filtered with the help of a vacuum filter and the filtrate was defatted using hexane. The ethanolic portion was dried and dissolved in distilled water for analysis [1, 3].

UV Visible spectrum analysis

The extract was scanned in the wavelength ranging from 200 to 700 nm using UV- 2600 Shimadzu Spectrophotometer and the characteristic peaks were detected and plotted [1, 3].

Analysis of FT-IR spectra

Fourier transform infrared spectroscopy (FT-IR) is one of the most powerful approaches to identify and characterize the type of chemical bonds (functional groups) present in compounds. The ethanol extract of the plant was mixed with KBr salt using a mortar pestle and compressed into thin tablets and IR spectra and peaks were recorded on a Perkin Elmer FT-IR (model RX1) spectrometer between 4000-400 cm^{-1}. Each analysis was twice done for confirmation [1, 3].

Determination of Sun Protection Factor (SPF) Value

SPF content of the extract was determined according to the method described by **Mansur et al. 1986.** The absorption spectra of sample were viewed in the range of 290 to 320 nm using 1 cm quartz cell, and taking ethanol as blank. The absorption data were obtained in the range of 290 to 320, every 5 nm, and 3 replica were made at each point, followed by the application of Mansur equation [11]. Here, EE (λ) is the erythemal effect spectrum, I (λ) represents the solar intensity spectrum, Abs (λ) denotes the absorbance of sunscreen product; CF is the correction factor (=10). The values of EE × I remain constant [13].

The efficacy of a sunscreen is usually expressed by the sun protection factor (SPF) = CF × $^{320}\Sigma_{290}$ EE (λ) × I (λ) × Abs (λ)

Ascorbic acid content (AA*)*

Colorimetric method was followed to determine the ascorbic acid content [12] of the collected sample species. 1 g of the fresh foliage leaves was homogenized with 10 ml of 6% trichloroacetic acid (TCA) with the help of mortar and pestle and centrifuged at 5000 rpm for 5 minutes. The supernatant was taken and a pinch of activated charcoal was added and filtered. The volume of the filtrate was made up to 100 ml with distilled water. 5 ml of supernatant was mixed with 3 ml of 2% 2, 4 - DNPH in 9 (N) H_2SO_4 and to it 1-2 drops of 10% thiourea solution in 70% ethanol was added and was boiled for 15 minutes in water bath and cooled in room temperature. To each sample 5 ml of 80% H_2SO_4 was added at 0°C. After 30 minutes the absorbance was measured at 530nm with a colorimeter. The concentration of unknown samples was extrapolated from a standard ascorbic acid solution of 50ppm using the following formulae:

Concentration of unknown solution = (Concentration of standard solution x $O.D_{530}$ of unknown) / $O.D_{530}$ of standard solution.

RESULT

UV-Vis spectrum pattern taken between 290 to 320 nm reveals that the characteristic absorbance (λ_{max}) centre is 295 nm for *Desmodium gangeticum* extract of leaves.

ISSN: 0975-8585

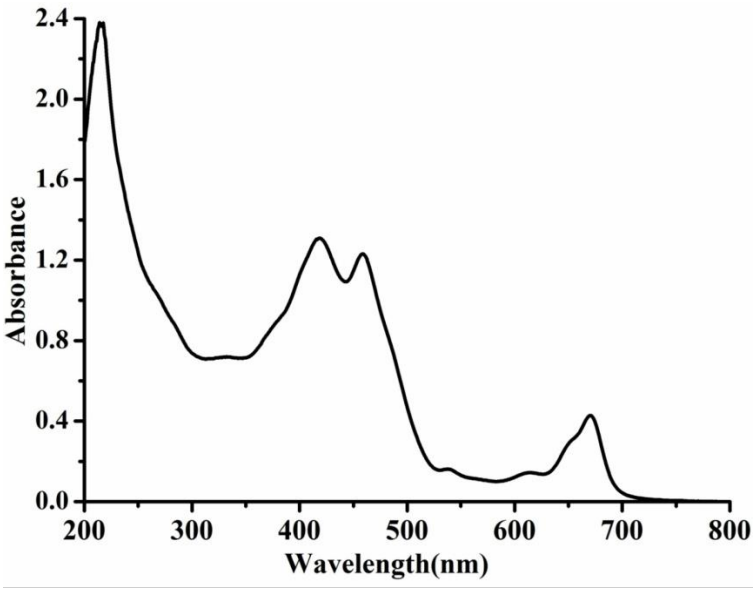

Fig 3: Ultraviolet-visible spectrum of water extracts of leaves of *Desmodium gangeticum*

The data of FT-IR analysis shows approximately similar pattern with characteristics wave numbers of phenolics antioxidants (presence of aliphatic group with characteristic wave number at 3334.92, 3298.28 and 3290.56 cm^{-1}) and peak of aromatic molecules conjugated with carbonyl groups (presence of carbonyl group with characteristic wave number at 1635.64 cm^{-1}) as well as multiple peaks of aromatic amine antioxidants (1315.45 cm^{-1}) that indicate the uniformity of spectral properties of aqueous extracts of across two different phytosources.

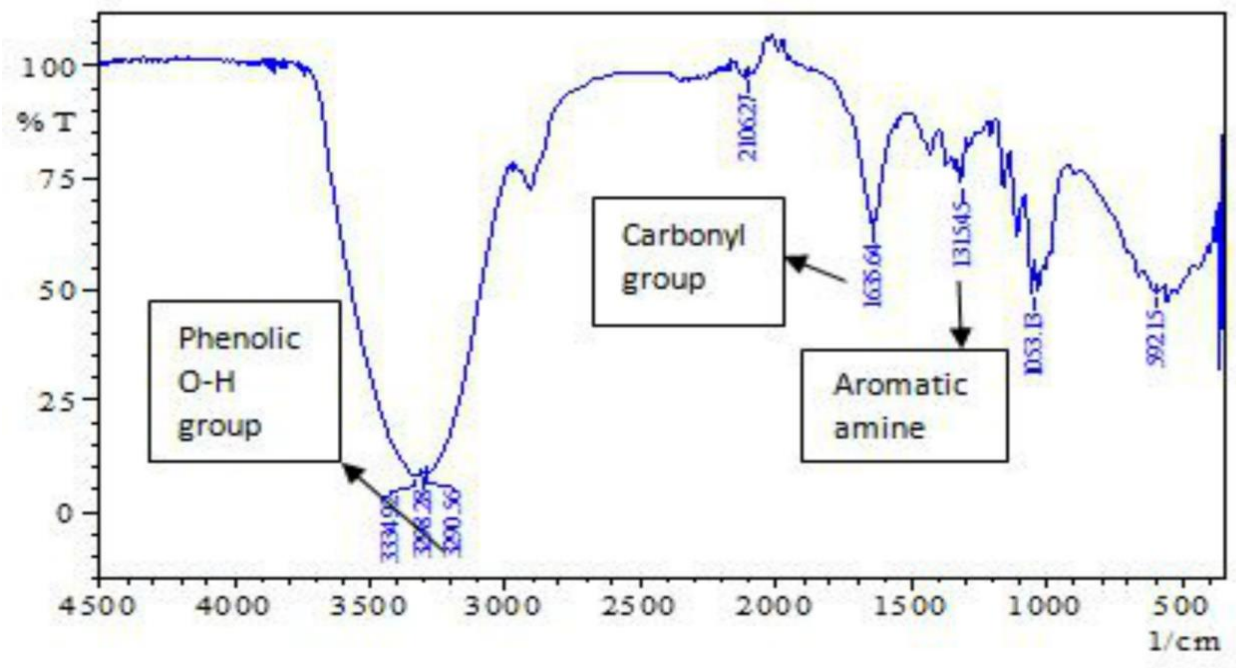

Fig 4: Ultraviolet-visible spectrum of water extracts of leaves of Desmodium gangeticum

ISSN: 0975-8585

Table 1: Determination of SPF value of the aqueous extract of *Desmodium gangeticum*

Wave Length (λ nm)	EE (λ) X I (λ) employed	Absorbance (A)	EE X I X A	SPF=ΣEE(λ) X I(λ) X A X 10*
290	0.0150	0.818±0.004	0.0122±0.00006	**7.276±0.04**
295	0.0817	0.769±0.004	0.0628±0.00033	
300	0.2874	0.738±0.004	0.2121±0.00115	
305	0.3278	0.720±0.003	0.2360±0.00098	
310	0.1864	0.710±0.003	0.1323±0.00056	
315	0.0837	0.710±0.003	0.0594±0.00025	
320	0.0180	0.711±0.003	0.0127±0.00005	
Σ EE(λ) X I(λ) X A			0.7276±0.0035	

DISCUSSION

FT-IR analysis reveals presence of distinct phenolic compounds (presence of aliphatic group with characteristic wave number at 3334.92, 3298.28 and 3290.56 cm^{-1}) which known to play critical role against sunray induced damage. The leading constituent in sunscreens are normally aromatic molecules conjugated with carbonyl groups (presence of carbonyl group with characteristic wave number at 1635.64 cm^{-1}) as well as multiple peaks of aromatic amine antioxidants (1315.45 cm^{-1}). The presence of such phenolic components validates the molecule to absorb high energy UV rays and release the energy as lower energy state, thereby reducing the adverse effect of skin-damaging UV rays. It was inspiring to note that the extract hold significant level of sun protection factor (SPF) i.e. 7.276 speaking of its effective capability to block UV B by 83%- 88% [9].

The UV-Vis spectral pattern actually corroborates the high phenol content of the extract. When observed closely, it is found that highest absorbance λ_{max} centre around UV region of the spectrum is 295 nm. It supports the expectation that most of these phenolics (including flavonoids) contribute significantly against sunburn.

The plant extract showed contents of SPF 7.276. Moreover, Vitamin C content of the extract was found to be 52.6 mg/l. The extract possesses remarkable combination of SPF and Vitamin C. SPF decrease UV induced skin damage while vitamin C check sunburn and skin cancer. Thus, the plant offers unrecognised source of dynamic sunscreen with virtue against photo-aging. Such a unusual combination may be used in future pharmaceutical.

CONCLUSION

It appears from this study that, the ethanol leaf extract of *Desmodium gangeticun* shows genuine sun protection ability. The promising SPF content fortified with natural vitamin C found in the extract is encouraging. Further study with rigorous clinical tests are required to develop as effective SPF emollient with dermato-protective activities.

ACKNOWLEDGEMENTS

This work was financially supported by UGC Centre for Advanced Study (phase II) Department of Botany, India.

REFERENCES

[1] Banerjee S, Das M, Mondal S, Bandyopadhyay A, Photoprotective property of three plant derived extracts. In Defining Modern Biology: Plants and Microbes. (Kolkata, 2016, Binary Banyan) p. 23.

[2] Bharath A.K, Turner R.J. Impact of climate change on skin cancer**.** Journal of the Royal Society of Medicine, 2009. 102: 215-218.

[3] Das M., Mondal S., Banerjee S., Bandyopadhyay A., Indian J.L.Sci., 6: 43, (2017). doi: http://ijls.in/upload/1202845891Chapter_7.pdf.
[4] Diffey B. L. Solar ultraviolet radiation effects on biological systems. Rev Physics Med Biol, 1991 36: 299-328.
[5] Diffey B. L.Climate change, ozone depletion and the impact on ultraviolet exposure of human skin. Phys Med Biol 2004. 49: R1- 11.
[6] Giacomoni P. U. Biophysical and Physiological Effects of Solar Radiation on Human Skin. Cambridge, UK: RSC Publishing; 2007.
[7] http://cookislands.bishopmuseum.org/species.asp?id=6047
[8] http://icwow.blogspot.in/2013/01/salpani-ful-desmodium-gangeticum.html
[9] http://msykes.com/things/sunscreen/
[10] http://www.bimbima.com/health/post/2016/06/11/shalaparni.aspx
[11] Mansur J. S, Breder M. N. R, Mansur M. C. A, Azulay R. D. Determinação Do Fator De Proteção Solar Por Espectrofotometria. An. Bras. Dermatol 1986. 61: 121-124.
[12] Roe J.H. Appraisal of methods for the determination of l-ascorbic acid**.** Annals of the New York Academy of Science 1961; 92: 277-283.
[13] Sayre R. M., Agin . P., Levee G. J, Marlowe E. A comparison of in vivo and in vitro testing of sunscreen formulas. Photochem. Photobiol 1979; 29: 559-66.
[14] Telang P. S. Vitamin C in dermatology. Indian Dermatol Online J 2013. 4: 143-146.
[15] Van der Leun J. C, de Gruijl F. R. Climate change and skin cancer**.** Photochem and Photobiol Sci 2002. 1: 324 – 326.