



**ISOLATION OF ONE COMPOUND FROM *AMARANTHUS SPINOSUS* L. LEAVES
RESPONSIBLE FOR UV RADIATION ABSORPTION**

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ABSTRACT

Since long *Amaranthus spinosus* linn. (*A. spinosus* L.) is being used for treatment of several diseases. In traditional medicine the plant leaves are used to cure burns, wounds, eczema, boils, gastroenteritis, gall bladder inflammation, ulcerated mouths, colic menorrhagia, abscesses, arthritis and for the treatment of snakebites. Modern researchers evaluated pharmacological activity of the plant and found that the plant has several pharmacological activities like anti cancer, anti inflammatory, anti diabetic, anti microbial, anti gastric ulcer, anti oxidant, hepato protective, gastro protective etc. Recently we have noted UV absorption property of *A. spinosus* L. leaves and maximum absorption was found in autumn. Aim of the present work was to isolate the active compound from the plant leaves responsible for UV radiation absorption. *A. spinosus* L. leaves were processed for isolation work by standard methodologies. After solvent extraction and acid hydrolysis followed by solvent treatment, chromatographic experiments were done. Finally a compound was crystallized. UV absorption property of the isolated compound was studied. The compound showed maximum absorption at 200 nm. The compound, therefore, may be used in the preparation of sun screen lotion.

KEYWORDS: *Amaranthus spinosus* Linn. leaves, UV absorbing property, isolation of active compound, sun screen lotion.

1. INTRODUCTION

A. spinosus L. (family, Amaranthaceae), a medicinal plant, is now widely distributed in different parts of the globe like Bangladesh, Nepal, Indonesia, Maldives, Japan, Australia, Ghana, Myanmar, Sri Lanka and Philippines. The plant is also present in fields, road sides and waste places of India.^[1]

Commonly known as 'Prickly amaranthus'.^[2] *A. spinosus* L. shows high content of phenolic acids, steroids, catechuic acid, saponin, flavonoids, glycosides, betalain, alkaloids, amino acids, rutin, xylofuranosil, uracil, beta-sitosterol glucoside, b-sitosterol, stigmaterol, beta – D-ribofuranosyl adenine, betaxanthin, betacyanin, amaranthine and isomaranthine, stigmaterol, hydroxycinnamates, terpenoids, lipids, 7-p-coumaroyl apigenin 4-o-beta-D-glucopyranoside, linoleic acid, tannins, betanin, quercetin, kemferol glycoside as well as carotenoids.^[3]

In traditional medicine, the plant is used as nutritive vegetable.^[4] It is also used as an antidote to snake poison and to treat different ailments like arthritis, diarrhea,

gastroenteritis, gall bladder problem, burns, wounds, toothaches, abscesses, boils, ear ache etc.^[5]

Modern researchers claimed that *A. spinosus* L. has a wide range of pharmacological properties. These include anti-helminthic, antifertility, antimalarial, antipyretic, antipeptic ulcer, antidiabetic, anti-hyperlipidemic, antibacterial, antioxidant, anti-nociceptive, antigenic and allergenic activity, antidiarrhoeal etc. Different parts of the plant also act as bronchodilator and spasmolytic, diuretic, analgesic and hepato-protective. They possess immuno-modulatory properties as well as gastrointestinal and haematological activity.^[6]

Recently we have seen that *A. spinosus* L. leaf has UV absorption activity and maximum activity was found during autumn (results are under communication). It is, therefore, thought worthwhile to isolate the active compound present in leaves of *A. spinosus* L. responsible for UV absorption property. The present study was thus carried out in this direction.

2. MATERIALS AND METHODS

2.1 Collection of plant material

A. spinosus L. leaves were purchased from the local market during autumn. Leaves were authenticated by the taxonomist of the department of Botany of the University

of North Bengal, Dist. Darjeeling, West Bengal, India. A voucher specimen was kept in the department of Medical Biotechnology, Sikkim Manipal Institute of Medical Sciences of the Sikkim Manipal University, Gangtok, Sikkim, India for future references.



Figure 1: *A. spinosus* L. leaves.

2.2 Chemicals

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

2.3 Preparation of the plant leaves

A. spinosus L. leaves were washed thoroughly under running tap and then by distilled water. Leaves were shade dried and powdered. The powder was kept for isolation study.

2.4 Isolation of active compound

This was carried out by the following steps involving principles of standard isolation procedures of chemical compounds from plant sources.^[7-10]

Powdered leaves of *A. spinosus* L. (100 g)

SOLVENT EXTRACTION

Extracted with 500 ml of ethanol in soxhlet at 37°C for 15 minutes.



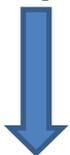
It was then centrifuged. Supernatant collected and evaporated to dryness.



Active brown mass

ACID REFLUX

Refluxed with 100 ml of 1(N) HCl for 10 min on a water bath at 100 °C. It was then cooled and centrifuged. Supernatant was evaporated to dryness.



Active brown mass

TREATMENT WITH n-BUTANOL

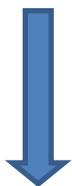
Extracted with 100 ml of n-butanol on a rotary shaker for 30 min. It was then centrifuged. Supernatant was evaporated to dryness.



Active brown mass

ALUMINA COLUMN CHROMATOGRAPHY

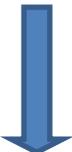
Extracted with 20 ml of ethanol for 10 min. It was then filtered. With filtrate alumina column chromatography was performed. Elution was done by ethanol – n butanol mixture (1:1 v/v).



Third band was found active

SILICA GEL G CHROMATOGRAPHY

Eluent of active third band was evaporated to dryness. The dry mass was extracted with 20 ml of ethanol for 10 min. It was then filtered. With filtrate silica gel G column chromatography was done. Elution was by chloroform : ethanol mixture (1:1 v/v).



Fourth band was found active

POLYAMIDE COLUMN CHROMATOGRAPHY

Eluent of active fourth band was evaporated to dryness. The dry mass was extracted with 20 ml ethanol for 10 min. It was then filtered and the filtrate was subjected to column chromatography using polyamide as adsorbent. Elution was done by ethanol, acetone mixture (1:1 v/v).



Second band was found active

CRYSTALLIZATION

Eluent of the active second band obtained from the above step was evaporated to dryness. Repeated crystallization was done from ethyl acetate, chloroform mixture (40:60, v/v).



Crystals obtained (18.8 mg)

2.5 UV absorption property of the isolated compound

To 10 mg of the isolated compound distilled water (100 ml) was added. The solution was filtered and the filtrate was processed in a spectrophotometer for UV ray absorption at the ranges of 200-400 nm at 10 nm intervals.

3. RESULTS**3.1 Isolation of the compound**

A brown coloured compound was isolated.

3.2 UV absorption property of the isolated compound

Result, shown in figure – 2, indicates that the isolated compound absorbed UV ray in all wave lengths of UV region. Absorptions in respect of wave lengths were, 0.29 (400 nm), 0.48 (350 nm), 0.65 (300 nm) and 0.92 (250 nm). Maximum absorption, however, was noted at 200 nm (1.8).

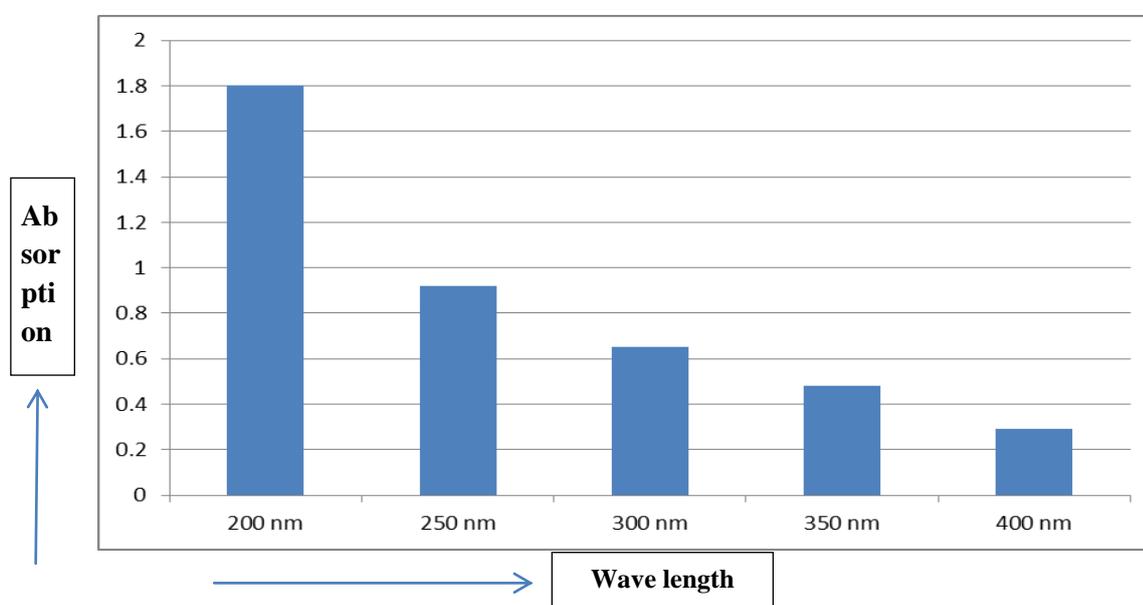


Figure 2: UV radiation absorption by the isolated compound from *A. spinosus* L. leaves.

4. DISCUSSION

In electromagnetic spectrum ultraviolet radiation falls under 180 – 400 nm wavelength region. Ultraviolet radiation is non-ionizing radiation. Based on region ultraviolet radiation is known as, black light (wave length, 315-400 nm), erythral (wave length, 281-314 nm) as well as germicidal (wave length, 180-280 nm). Common source of UV radiation is sunlight. UV generates in the laboratory through lasers, trans illuminators, germicidal lamps, biological safety cabinets and cross linkers.

Solar UV-radiation has both good and bad effects. It is required for cutaneous synthesis of vitamin D and this covers almost 90% of the vitamin D requirement of human body. On the other hand there are plenty bad effects of UV radiation. These include photosensitivity reactions to ingested drugs, skin injury, eye injury, genetically determined photo sensitivities etc. Excessive exposure to UV rays has profound health risks, including pigmentary changes, atrophy, wrinkling and malignancy. Skin is affected. Over exposure of UV may develop any one of three types of skin cancer viz. squamous cell carcinoma, basal cell carcinoma and malignant melanoma may develop. UV ray also affects eye. It can cause injury to cornea, the outer protective coating of the eye. There is a painful inflammation of the eye caused by UV radiation-induced lesions. Chronic UV exposure to eye can lead to the formation of cataracts. Over-exposure to UV radiation can change the distribution and function of disease-fighting white blood cells in humans thereby has a harmful suppressing effect on the immune system.^[11]

Therefore, efforts are going on to identify the sources which can absorb UV radiation from the environment for human protection. In this context research has been extended even in the field of medicinal plants.^[12] Several medicinal plants like *Mentha piperita*, *Azadirachta indica*, *Carica papaya*, *Aloe vera*, *Lycopersicon esculantum*, *Oscimum sanctum*, *Phyllostachys pubescens*, *Calotropis gigantea* L. etc. are known to have anti solar activity.^[13,14]

In the present study we have isolated a compound from *A. spinosus* L. leaves. The compound can absorb rays in all wave length of UV region but maximum absorption was at 200 nm. The compound needs characterization. Work in this direction is now going on in our laboratory.

5. CONCLUSION

In the present study we found UV radiation absorption property of the isolated compound from *A. spinosus* L. leaves. The property may be utilized in future to protect humans from UV radiation.

Recommendation

Isolated compound from *A. spinosus* L. leaves may be used in sun screen lotion preparation as UV absorbing material.

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Conflict of interest

Nil.

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