



ANTITUBERCULAR ACTIVITY OF *ACTINIOPTERIS RADIATA* LINN

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ABSTRACT

The Antitubercular effect of n-Hexane, Chloroform, Ethanol extracts was prepared from whole plant of *Actiniopteris radiata* Linn was evaluated against *Mycobacterium tuberculosis* using Microplate Alamar Blue assay (MABA). Minimum inhibitory concentration (MIC) was taken to assess antitubercular activity. The results shown that Chloroform extract have more significant antitubercular activity as compared to n-Hexane, Ethanolic extracts. Pyrazinamide and Streptomycin was taken as standard drugs.

Keywords: *Actiniopteris radiata* Linn, minimum inhibitory concentration, antitubercular activity.

INTRODUCTION:

Actiniopteris radiata Linn more commonly known as *Nemaliadugu* in telugu which belongs to the family Actiniopteridaceae, it is a fern widely distributed throughout Africa and adjacent Islands, Madagascar, Arabia, Iran, Afghanistan, Nepal, India, Burma and Australia.^{1,2,3,4} The plant is claimed to possess anti-histaminic activity, anti-cholinergic, anti-microbial activity, anti-inflammatory activity, anti-helmenthic activity, analgesic activity and used as styptic.¹⁻²¹ The aim of our present study was to investigate the Antitubercular activity of n-Hexane, Chloroform and Ethanolic extracts of *Actiniopteris radiata* Linn by using Microplate Alamar Blue assay.

MATERIALS AND METHODS

Plant Materials: The whole plant of *Actiniopteris radiata* Linn were collected from Tirumala Hills, Tirupati and Chittoor district of

Andhra Pradesh, in the month of July which were identified by Dr. K. Madhava Chetty, Associate Professor, Hebarium keeper, Department of Botany, Sri Venkateshwara University, Tirupati, Chittoor (Dist.), A.P.

Preparation of Extract: The powder of whole plant of *Actiniopteris radiata* Linn was extracted with n-Hexane, Chloroform, Ethyl acetate and Ethanol successively by Soxhlation method and concentrated over water bath and evaporated under reduced pressure. The n-Hexane, Chloroform and Ethanolic extracts was chosen for Anti tubercular activity.⁴⁻⁶

Chemicals: n-Hexane, Chloroform, Ethanol, 96 well plate, Middlebrook 7H₉ broth with *M. tuberculosis* H37 RV strain, Deionized water, Almar Blue reagent, 10% Tween 80, Incubator, Micropipettes.

EXPERIMENTAL PROCEDURE:¹⁴

1. Briefly, 200µl of sterile deionized water was added to all outer perimeter wells of sterile 96 wells plate to minimized evaporation of medium in the test wells during incubation.
2. The 96 wells plate received 100 µl of the Middlebrook 7H₉ broth and serial

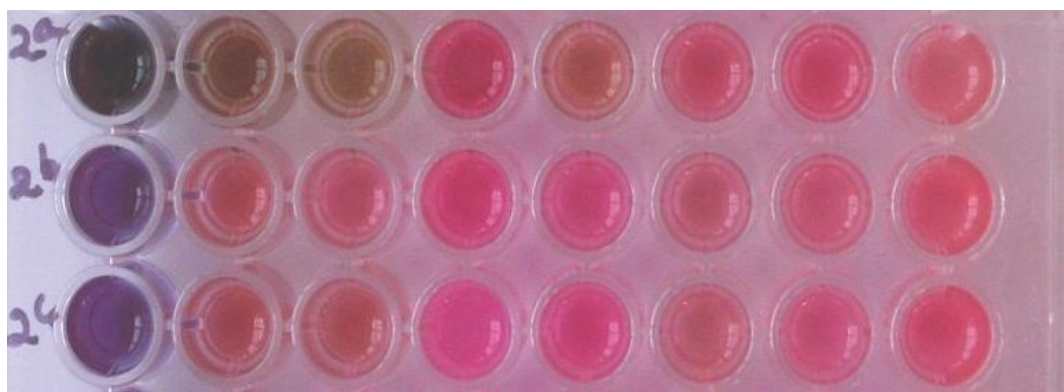
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- dilution of extracts were made directly on plate.
- The final drug concentrations tested were 100 to 0.2 µg/ml.
 - Plates were covered and sealed with para film and incubated at 37°C for five days.
 - After this time, 25µl of freshly prepared 1:1 mixture of Almar Blue reagent and 10% Tween 80 was

- added to the plate and incubated for 24 hrs.
- A blue color in the well was interpreted as no bacterial growth, and pink color was scored as growth.
- The MIC was defined as lowest drug concentration which prevented the color change from blue to pink shown in fig No: 1.



Microplate Almar Blue Assay

Phytochemical analysis: 10-12

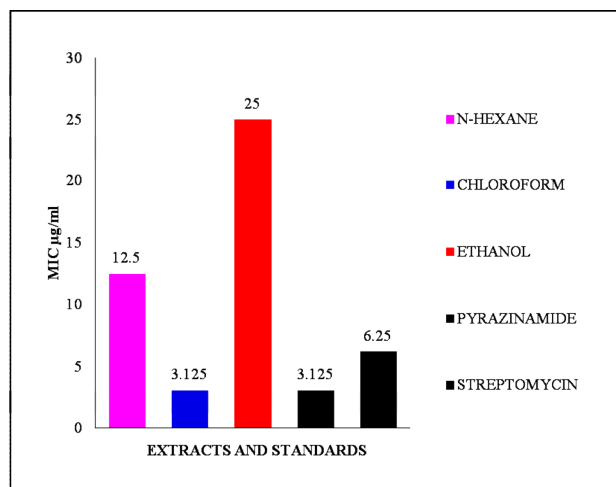
The n-Hexane, Chloroform and Ethanol extracts of *Actiniopteris radiata* Linn were subjected to Thin Layer Chromatography using TLC plates (0.1 mm thick silica gel) eluted with n-hexane: Ethyl acetate (8:2) and Chloroform: Benzene (6:1) respectively. The spots were identified under long UV light by using UV cabinet.

RESULTS AND DISCUSSION: 15-20

Traditionally medicinal plants have been used as a folk medicine throughout the world to treat various diseases; especially tuberculosis.^{11,12} We evaluated preventive effects of n-hexane, Chloroform and Ethanolic extracts of using Microplate Almar Blue assay method. The minimum inhibitory concentration of n-Hexane, Chloroform and Ethanolic extracts was 12.5, 3.125, 25µg/ml respectively and compared with standard drug Pyrazinamide- 3.125µg/ml and Streptomycin- 6.25µg/ml. The obtained results were shown in Table 1.

Table 1: MIC of different Extractives

S. No	Type of Extract	MIC (µg/ml)
1.	n-Hexane (2b)	12.5
2.	Chloroform (2a)	3.125
3.	Ethanol (2c)	25



CONCLUSION:

This study reveals that the plant shown significant antitubercular effect of n-Hexane, Chloroform and Ethanol extracts from plant *Actiniopteris radiata* Linn. Further studies using more specific methods are required to explore the constituents responsible for the activity and the mechanism of this activity which might prove important and improved therapies for the treatment and prevention of tuberculosis.

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