

**CHARACTERIZATION AND EVALUATION OF ANTIPYRETIC ACTIVITY OF  
ALTERNANTHERA BRASILIANA (L.) KUNTZE ETHANOLIC LEAVES EXTRACT**Unnimaya I. S.<sup>1\*</sup>, Dr. G. N. Pramodini<sup>2</sup>, Dr. D. Vijay Kumar<sup>3</sup> and Dr. Sapna Shrikumar<sup>4</sup><sup>1,2,4</sup>Department of Pharmacognosy and Phytochemistry, <sup>3</sup>Department of Pharmaceutical Chemistry,  
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**ABSTRACT**

*Alternanthera brasiliana* (L) Kuntze *Amaranthaceae* is a herbaceous plant used against inflammation, cough, and diarrhea in Brazilian popular medicine. It is a well-known plant rich in phytoconstituents. In the present work deals with the spectroscopic characterization and evaluation of *in-vivo* Antipyretic activity of ethanolic leaves extract of *Alternanthera brasiliana* (L) Kuntze. Spectroscopic analysis by FTIR revealed the presence of flavonoids especially quercetin, through the appearance of intense bands at specific areas. Antipyretic activity of ethanolic extract of *Alternanthera brasiliana* (L) Kuntze leaves were evaluated using yeast induced pyrexia model. At the dose 200 mg/kg of ethanolic extract shows significant reduction in body's temperature ( $p > 0.05$ ) was observed in the temperature of rats tested.

**KEYWORDS:** *Alternanthera brasiliana* (L.) Kuntze, Flavonoids, Ethanolic extract, FTIR, Antipyretic activity.**INTRODUCTION**

*Alternanthera brasiliana* (L) Kuntze, *Amaranthaceae* is evergreen, perennial herbs, native to tropical and sub-tropical regions of Australia & South America. *Alternanthera brasiliana* (L) Kuntze are grown for edging beds and for bedding.<sup>[1]</sup> They hardy plants which stand trimming, and can be easily propagated by cutting or by division. *Alternanthera brasiliana* (L) Kuntze is a brazilian plant occurring in several regions, being known as "penicilina" or terramicina, widely used by rural communities as medicinal agent to cure different disease, such as inflammation, and dolorous or infection processes, wound healing, analgesic, antitumor activity, immunomodulator and lymphocyte proliferation.<sup>[2,3]</sup>

**METHODS AND METHODOLOGY****Preparation of *Alternanthera brasiliana* (L) Kuntze leaf extract**

The plant materials (leaves) of *Alternanthera brasiliana* (L) Kuntze were collected and then shade dried and coarsely powdered. 30 g of coarsely powdered leaves was packed in Soxhlet apparatus and extracted by using 1000ml ethanol (80%) (Approx. 2 days). The extract was collected. The extract was then filtered through Whatmann No. 1 filter paper and concentrated.<sup>[4]</sup>

**CHARACTERIZATION OF *ALTERNANTHERA BRASILIANA* (L) KUNTZE EXTRACT  
FOURIER TRANSFORM INFRARED (FTIR)  
SPECTROSCOPIC ANALYSIS**

- Fourier transform infrared (FTIR) was used to identify the characteristic functional groups in the extract. It provides the information about the structure of a molecule could frequently be obtained from its absorption spectrum.
- The functional groups present in the chemical constituents of plants are identified by FTIR.
- When the extract was passed into the FTIR; the functional groups of the components were separated based on its peaks ratio.<sup>[5,6,7]</sup>

**Procedure**

- 1mg of the dried powder of ethanolic extract of *Alternanthera brasiliana* (L.) Kuntze mixed with KBr salt, using a mortar and pestle, and compressed into a thin pellet at a pressure of 6 bars for 2 min, in order to prepare a translucent sample disc.
- Then the sample was loaded in FTIR- spectroscope (Shimadzu, Japan).
- The IR spectrum was obtained using Bruker, Germany Vertex 70 infrared spectrometer.
- The sample was scanned from 4000 to 400  $\text{cm}^{-1}$  with a resolution of 4 $\text{cm}^{-1}$  The peak values of the FTIR were recorded.<sup>[8,9,10,11]</sup>

## IN-VIVO PHARMACOLOGICAL ACTIVITY IN-VIVO ANTIPYRETIC ACTIVITY

### Methodology and experimentation

#### Test System

Species: Wistar rats

Strain: Wistar rats

Age: 6-8 weeks at the time of dosing

Total no. of rats: 24

Sex: Male

#### Experimental Animals

In-bred Wistar rats ((180–220 g) grams) were selected for these studies. The animals were housed in polypropylene cages with stainless steel top grills having facilities for holding pellet food and drinking water in bottle with stainless steel sipper tube. Each cage contained 6 rats. All mice had free access to potable water and standard pelleted laboratory animal diet *ad libitum*. Paddy husk was used as bedding material. Throughout the acclimatization and treatment period, animal room temperature and relative humidity was maintained at 22±3°C and 30% to 70% RH respectively. Illumination was controlled to give 12 hours light and 12 hours dark cycle during the 24-hour period.

The experimental protocol is approved by the institutional Animal Ethics committee proposal no: SLIMS/53/IAEC/2024-25. Following C.P.C.S.E.A guideline. All the experimental works were carried out in the KMCH College of Pharmacy, Coimbatore.

#### Acclimatization

The rats were acclimatized for 3 days to the laboratory conditions and were identified by a unique tail marking using permanent red marker pen. During the acclimatization, individual animal was subjected to daily general observation and prior to final assignment to the study the animals were subjected to a detailed clinical examination to ensure the selected rats were in a good state of health.

#### PROCEDURE

- The Antipyretic activity of *Alternanthera brasiliensis* (L) Kuntze extract was evaluated by using wistar rats. The animals were divided into 4 groups (Wistar rats group) containing six animals in each. Give water but denied food for 6 hrs. before starting experiment.
- One day before the experiment, three basal readings of normal rectal body temperature in each rat were recorded by using digital thermometer.
- Pyrexia was induced in all rats by injection of 20% aqueous suspension of brewer's yeast (*Saccharomyces cerevisiae*) 10 ml/kg. Subcutaneous (SC) into the dorsum region.
- All animal groups (4 groups) were fasted with access to only water, after injection of yeast for 18 h.
- After 18 hrs. of yeast administration; initial body temperature was recorded.

- After that, the rectal temperature of each rat was recorded and pyrexia was confirmed by increase in temperature more than 1°C, while animals showing less than 1°C rise in temperature were excluded from the experiment.
- The group I received saline (10 ml/kg), group II received paracetamol (100 mg/kg) as a standard drug, while group 3 and 4 received extract 100,200 mg/kg b. w, p. o. The rectal temperature of the groups was recorded at 1 h intervals for 6 h.<sup>[12,13,14]</sup>

#### Randomization & Grouping

- Following completion of acclimatization, the rats were randomized into 4 groups, each group consisting of 6 animals.
- **Group I-** Vehicle (distilled water)<sup>a</sup> p. o.
- **Group II-** Received a standard paracetamol (100 mg/kg b. w.) p. o. + brewer's yeast (*Saccharomyces cerevisiae*) 10 ml/kg.
- **Group III-** received extract 100 mg/kg b. w p. o. + brewer's yeast (*Saccharomyces cerevisiae*) 10 ml/kg.
- **Group IV-** received extract 200 mg/kg b. w p. o. + brewer's yeast (*Saccharomyces cerevisiae*) 10 ml/kg.
- The rectal temperature of the animals was measured at 0, 1, 2, 3, 4, 5 and 6 hours after the drug administration.

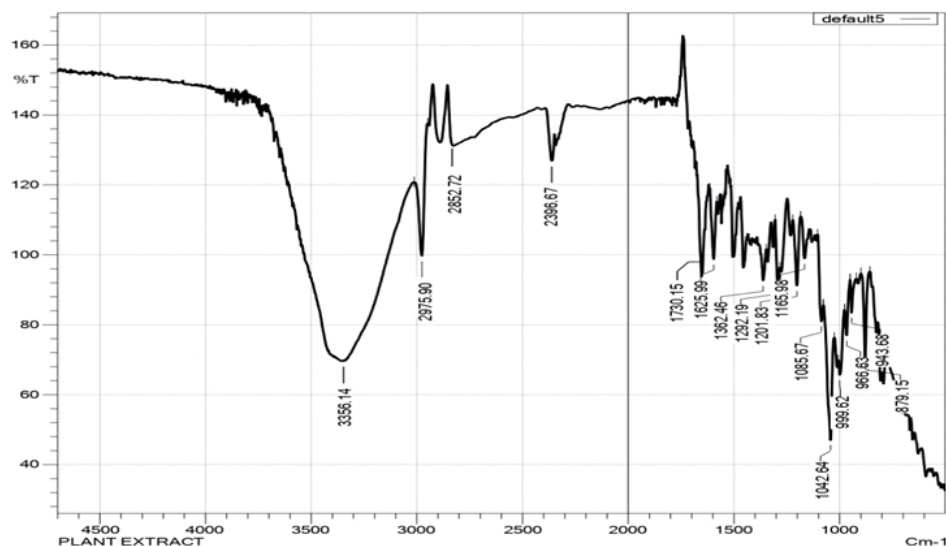
#### Dose Formulation Preparation

The test and standard drug of Paracetamol were prepared freshly on the day of dosing. The compounds were dissolved in distilled water.

#### Parameters Assessed

Rectal body temperature.

## RESULT AND DISCUSSION

CHARACTERIZATION BY FTIR SPECTROSCOPIC ANALYSIS OF ABEE  
FTIR SPECTRAL DATA INTERPRETATIONGraph No. 2: FTIR spectrum of leaf extract of *Alternanthera brasiliana* (L) Kuntze.

The data on the peak values and the probable functional groups (obtained by FTIR analysis) present in the leaf extract of *Alternanthera brasiliana* (L) Kuntze.

Table 23: The data on the peak values and the probable functional groups present in the leaf extract of *Alternanthera brasiliana* (L) Kuntze.

SL No:	Wave no: (cm <sup>-1</sup> ) test	Wave no: (cm <sup>-1</sup> ) standard	Functional group assignment	Expected phytochemical identified
1	3356.14	3570-3200	H-bonded, O-H stretch, N-H stretch	Hydroxyl group, alcohols, phenols, amines, amides
2	2975.90	2400-3500	O-H stretch, H-bonded	Carboxylic acid, alkanes (carboxylic acid hydroxy compounds)
3	2852.72	2865-2845	Symmetric stretching of CH (CH <sub>2</sub> ) vibration, O-H stretch, C-H stretch	Fatty acids, lipids, proteins (carboxylic acid, alkanes)
5	1730.15	1820-1670	C=O stretch	Carbonyls (general), carboxylic acids, aldehydes, saturated aliphatic
6	1625.99	1680-1620	C=C stretch	alkanes
7	1362.46	1410-1310	O-H bend/ alcoholic group	Phenol/tertiary alcohol
8	1292.19	1320-1210	C-O stretch, C-N stretch	Carboxylic acids, aliphatic amines
10	1165.98	1360-1080 1300-1150	C-N stretch, C-H stretch	Amine, alkyl halides
11	1085.67	1100-1000	PO <sub>3</sub> stretch	Phosphate ion
12	1042.64	1100-1000	PO <sub>3</sub> stretch	Phosphate ion
13	999.62	1100-1000	C-H bending	Alkenes
14	966.63	1100-1000	C-H bending	Alkenes

- The results of FT-IR spectroscopy confirm the presence of various chemical constituents such as flavonoids, phenols, alcohol, alkanes, aromatic carboxylic acid, halogen compound and alkyl halide in the ethanolic extract of *Alternanthera brasiliana* (L) Kuntze leaves (are given in the table).
- The result revealed the presence of the flavonoids and polyphenols due to the presence of O-H stretching alcohols due to N-H stretching, terpenes due to C-H groups, and also the presence of alkanes,

alkenes, aldehydes, alkyl halides, carboxylic acids, aliphatic amines, amides and phosphate ion in the plant extract.

- The strong instance peak are identified at 3356.14 cm<sup>-1</sup> due to the presence of flavonoids especially the functional group of quercetin is phenol with a hydrogen bonded O-H stretch between 3600-3100 cm<sup>-1</sup>.

- So we can conclude that the presence of functional groups that responsible for the various medicinal properties of the test plant.
- There was no absorbance in the between the region 2220-2260  $\text{cm}^{-1}$  indicates that there was no cyanide group in this extract and doesn't contain any toxic substance in this extract.

### IN-VIVO PHARMACOLOGICAL ACTIVITY OF ABEE

#### IN-VIVO ANTIPYRETIC ACTIVITY

Antipyretic activity of *Alternanthera brasiliana* (L) Kuntze was evaluated by using brewer's yeast induced pyrexia. The leaves extract of *Alternanthera brasiliana* (L) Kuntze revealed pyrexia in rats by using brewer's yeast in a dose dependent manner.

### OBSERVATIONS

#### BODY TEMPERATURE

Table 24: Body temperature.

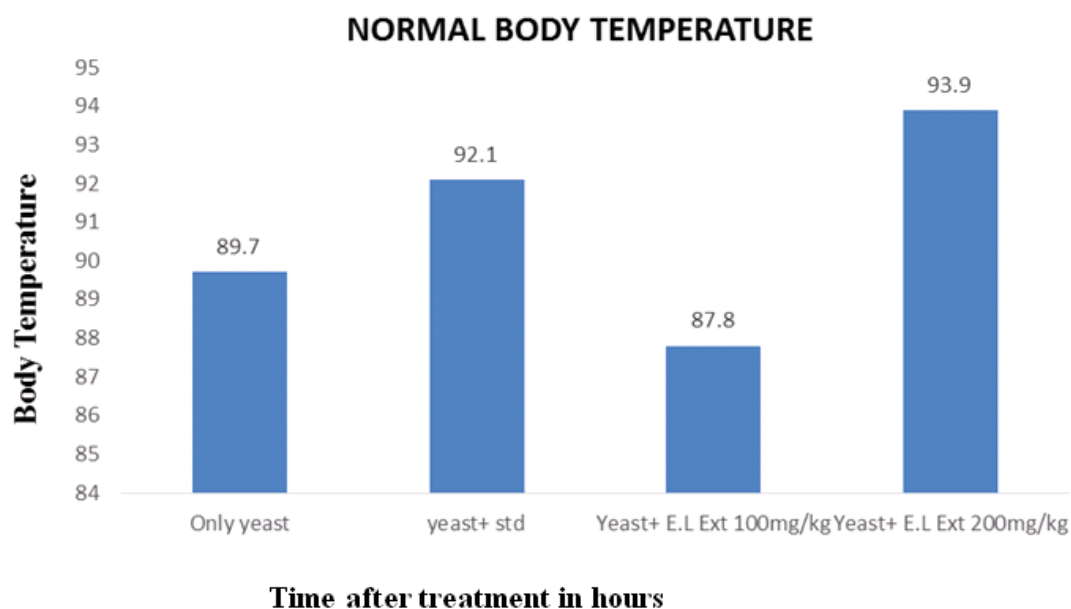
Group	Normal Body Temperature	Body Temperature 18 Hrs After Induction With Yeast						
		Initial Body Temperature After 18 Hrs.	Body Temperature after Treatment with Test & Std Drug					
			1hr	2 <sup>nd</sup> hr.	3 <sup>rd</sup> hr.	4 <sup>th</sup> hr.	5 <sup>th</sup> hr.	6 <sup>th</sup> hr.
Only Yeast	89.7±2.3	105± 1.24	106±0.937	106±1.11	105±0.421	104±1.71	101±1.62	98.7±1.01
Yeast + Std Paracetamol 100mg/kg	92.1±2.06	102±0.486 <sup>ns</sup>	101±0.814***	95.8±1.64***	94±1.77***	97.9±1.48*	95±2.25 <sup>ns</sup>	89.9±0.818 <sup>ns</sup>
Yeast + Ethanolic Leaf Extract 100mg/kg	87.8±2.08	101±0.285**	103±0.338*	98.9±0.564***	97.6±1.29**	98.9±1.78 <sup>ns</sup>	96.4±1.65 <sup>ns</sup>	97.2±1.75 <sup>ns</sup>
Yeast + Ethanolic Leaf Extract 200mg/kg	93.9±1.64	102±0.191 <sup>ns</sup>	102±0.74**	97.6±1.03***	98.9±1.06**	97.6±1.44*	93.1±1.86*	73.7±12.9*

Values are expressed as the mean ± S. D. Statistical significance (P) calculated by one way ANOVA followed by dunnett's \* $P < 0.05$  calculated by comparing

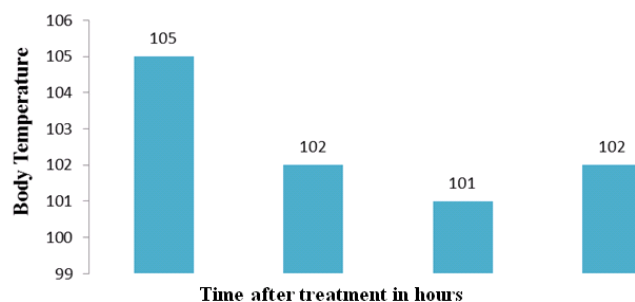
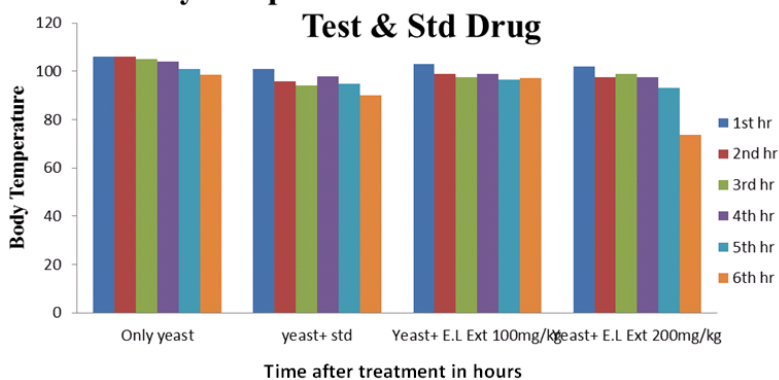
treated group with control group were considered to be significant.

### NORMAL BODY TEMPERATURE BEFORE YEAST ADMINISTRATION

Time after treatment in hours



Graph no 3: Normal body temperature before yeast administration.

**INITIAL BODY TEMPERATURE AFTER 18Hrs OF YEAST ADMINISTRATION****Initial Body Temperature After 18 Hrs****Graph no 4: Initial body temperature after 18 hrs of yeast administration.****BODY TEMPERATURE AFTER THE ADMINISTRATION OF TEST AND STANDARD DRUG****Body Temperature After Treatment With Test & Std Drug****Graph no 5: Body temperature after the treatment with test and standard drug.****Figure: Caging of rats.****Figure: Rectal Body Temperature Checking.**





**Figure: Pyrexia induced subcutaneously.**



**Figure: Plant extracts administration.**

### STATISTICAL ANALYSIS

The procedures were repeated three times and results expressed as mean  $\pm$  standard error of mean (SEM). Differences in observation were determined by Analysis of Variances (ANOVA) using Dunnett comparison method and difference between the treated group and control group were considered significant at \*  $p < 0.05$  levels.

### SUMMARY

- The overall summary of the thesis deals with the studies in the plant species of *Alternanthera brasiliana* (L) Kuntze belonging to the family, *Amaranthaceae*.
- Using a Fourier Transform Infrared (FTIR) Spectroscopy, *Alternanthera brasiliana* (L) Kuntze extract was characterized.
- The presence of numerous chemical components, including flavonoids, phenols, alcohol, alkanes, aromatic carboxylic acid, halogen compounds, and alkyl halides, is indicated by the strong bands that ABEE (*Alternanthera brasiliana* (L) Kuntze ethanolic extract) displayed. Furthermore, the O-H stretch is mostly caused by flavonoids. Furthermore, the strong peak at 3356.14, which is caused by flavonoids in particular because quercetin's functional group is hydrogen-bonded O-H stretch between 3600 and 3100  $\text{cm}^{-1}$ .
- The absence of absorbance in the range of 2220–

2260  $\text{cm}^{-1}$  suggests that this extract is free of hazardous substances and cyanide groups.

- *Alternanthera brasiliana* (L) Kuntze ethanolic extract was evaluated for its antipyretic properties. The results of this study demonstrated that *Alternanthera brasiliana* (L) Kuntze ethanolic extract effectively prevents mice from developing pyrexia caused by yeast.
- Additionally, the percent restraint is comparable while using Paracetamol, the usual medication. When ethanolic leaf extract is administered at a dose of 200 mg/kg, the body's temperature significantly drops in comparison to normal Paracetamol. The existence of flavonoids could be the cause of this.

### CONCLUSION

- In the present study FTIR analysis of Ethanolic extract of leaves showed the presence of flavonoids which are responsible for the medicinal properties of *Alternanthera brasiliana* (L) Kuntze plant.
- Further, This compound were evaluated for Antipyretic activity. The results of this study demonstrated that *Alternanthera brasiliana* (L) Kuntze ethanolic extract effectively prevents mice from developing pyrexia caused by yeast.
- Additionally, the percent restraint is comparable while using Paracetamol, the usual medication. When ethanolic leaf extract is administered at a dose of 200 mg/kg, the body's temperature significantly

drops in comparison to normal Paracetamol. This may be as a result of the presence of Flavonoids.

- Then we can conclude that the flavonoid enriched *Alternanthera brasiliana* (L) Kuntze shows significant reduction in pyrexia due to the inhibition of PGE2 synthesis by acting on hypothalamus.

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