ISSN No:-2456-2165

Evaluation of Antipyretic Activity of Ethanolic and Methanolic Extract of Leaves of *Clerodendrum infortunatum*

Abhijit V. Nikam[1]
[1]MRes Animal Science., Scholar., Life Science Department,
Aberystwyth University, Aberystwyth, Wales, United Kingdom(UK).

Ramprasad P. Mandade[2]
[2]Department of Veterinary Surgery & Radiology,
Nagpur Veterinary College, Maharashtra Animal and
Fishery Sciences University,
Seminary hills, Nagpur-440006, India

Govind V. More[3]
[3]Department of Veterinary Pharmacology & Toxicology,
Nagpur Veterinary College, Maharashtra Animal and
Fishery Sciences University,
Seminary hills, Nagpur-440006, India

Abstract:- The present study was planned to investigate possible antipyretic activity of ethanolic and methanolic extracts of leaves of the plant *Clerodendruminfortunatum* in Wistar rats. The plant leaves were collected, dried, powdered and defatted with petroleum ether and then subsequently subjected for 50% ethanolic and 50% methanolic extracts in Soxhlet's apparatus. Total 30 Wistar rats of either sex weighing 250 to 300 gm were used for the study. The pyrexia was induced with subcutaneous injection of 20% Brewer's yeast in the neck region. A stable pyrexia was achieved 18 hrs post injection. The rats were divided into 5 groups comprising 6 rats each. The group T1 and T2 were normal and pyretic control respectively, treated with 1ml of normal saline orally. The group T3 was treated with standard antipyretic drug, paracetamol at the rate of 10mg/kg body weight orally. Groups T4 and T5 were treated with 50% ethanolic and 50% methanolic extracts respectively at the rate of 400mg/kg body weight orally. The body temperature was recorded at an interval of 30 min, 60min, 180min an 360min post treatment. Both the extracts significantly restored the elevated body temperature at all the time interval and the result obtained was comparable with standard drug paracetamol. The findings of the study indicate that the plant *Clerodendruminfortunatum* possesses antipyretic activity and further investigations are necessary to confirm the activity.

Keywords:- *Clerodendruminfortunatum, Brewer's yeast,methanolic extract.*

I. INTRODUCTION

For centuries, Individuals in Bharat have embraced the utilization of organic substances derived from flora and fauna to address wounds, a practice deeply rooted in the ancient wisdom ofIntegrative Medicine, a widely recognized Indian medicinal tradition. The utilisation of natalmerchandise has transcended geographical boundaries, gaining significance globally. These legitimate remedies are emerging as pivotal components of substitute medicine due to their notably minimal adverse effects. Consequently, the scientific community is actively exploring the potential of traditional medicines, seeking to enhance animal health. These remedies are applied in their raw or unrefined state to combat chronic ailments effectively.This ethno pharmacology approach helps guide drug discovery and development based on centuries of experiential wisdom.

Elevated body temperature, commonly known as pyrexia or fever, represents a sophisticated exempt-corporal state activated by a deluge of intricate immunologicalresponses in reaction to contagion or provocative incentives within the organism. Pyrexia, in anatomic stint, denotes the elevation of the body's core temperature in response to the invasion of infective factors (Blatteis, 2006). As a result, inflammatory and infectious agents such as cytokines, interleukin-6, endothelin-1, interleukin-1β, chemokines, and especially prostaglandin E2 (PGE2) contribute to the generation of various autogenouspyrectic(Roth *et al.*, 2014). PGE2 is the key eicosanoid component involved in fever generation in the central nervous system of mammals (Gaetano *et al*., 2010). In summary, fever is an immune response mediated by biochemical factors like PGE2 that signal the body to raise temperature to combat infection.

Clerodendrum constitutes a vast assemblage of blossoming vegetation encompassing herbage, bush, and diminutive trees within the Lamiaceae family, thriving in equatorial and semi-equatorial zones across the globe. Originally delineated by Linnaeus, the genus unfolds a diverse botanical panorama. The blossoms of these plants attract flying insects and apodiform birds, while specific *Clerodendrum* species serve as sustenance for insect larvae. Indigenous medical traditions in numerous countries have harnessed the therapeutic potential of numerous types within this genus. Several *Clerodendrum* species have undergone comprehensive scrutiny, unveiling a wealth of Bioactive Compounds and unveiling noteworthy therapeutical attributes. These encompass antibiotic, antitumour, anti-miasma,Flavonoids, flavones, catechins, polyphenols, biguanides, biocide, and antiflux activities, as evidenced by research findings (Shrivastava *et al.*, 2007). the *Clerodendrum* genus contains medicinal flowering plants native to tropical regions that produce bioactive compounds exhibiting a range of pharmacological properties.

ISSN No:-2456-2165

Clerodendrum infortunatum Linn. is a species within the diverse *Clerodendrum* genus of flowering plants. Regionally, it is called “peruvelam” in Kerala and “Bhant” in Hindi. *Clerodendrum* species are present across tropical countries. *C. infortunatum* leaves are used in traditional medicine as a bitter libation, anti-episodic, anthelmintic, anodyne, and purgative and stimulate the contraction of the gallbladder. The plant extract are also applied topically in the form of a paste to treat malignancy and certain skin diseases (Kokate *et al.*, 2022). C. infortunatum is an Ayurvedic medicinal plant used to treat a variety of conditions and exhibits bioactive properties.

Clerodendrum infortunatum possesses useful medicinal properties due to presence of bioactive phytoconstituents like Phenols, Flavonoids, Steroids, Terpenoids, Fixed oil and Sugars.(Limsay*et al.*, 2019)

II. MATERIALS AND METHODS

A. Plant material

The harvest(Plant) material were procured from the local suppliers and duly authenticated from Department of Botany, RTM Nagpur University, Nagpur.(Voucher specimen no. 084, 28/11/2022) The powdered material was subjected to methanolic and ethanolic extraction used as herbal medicine.

Image 1: Clerodendrum infortunatum plant

Image 2: Drying of C. infortunatum Leaves

B. Preparation of methanolic and ethanolic extract

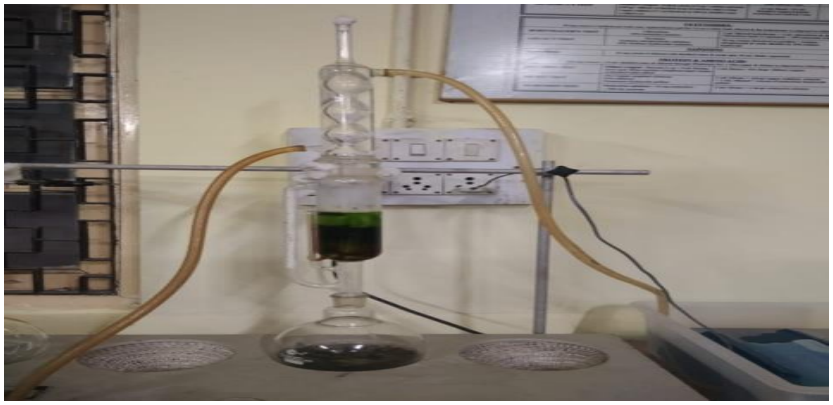


Image 3: Extraction of C. Infortunatum Leaves by using Soxlet’s assembly

ISSN No:-2456-2165

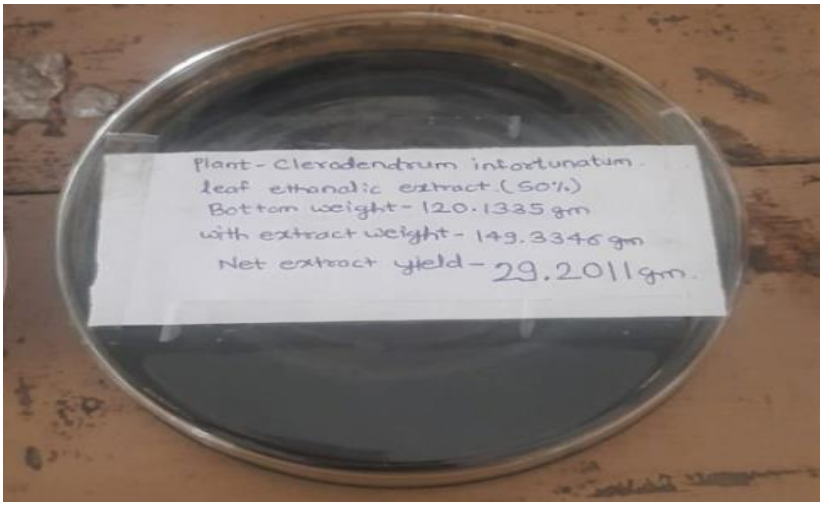


Image 4: 50% hydroethanolic extract

The 50% methanolic extract was prepared and used in the proposed study. The foliagewas thoroughly bathedwith demineralizationwater and shade scorched. After drying leaves were powdered in fine particles. A known quantity of powder of plant material first defatted with petroleum ether and subsequently extracted with 50% methanol and 50% ethanol using Soxhlet apparatus.

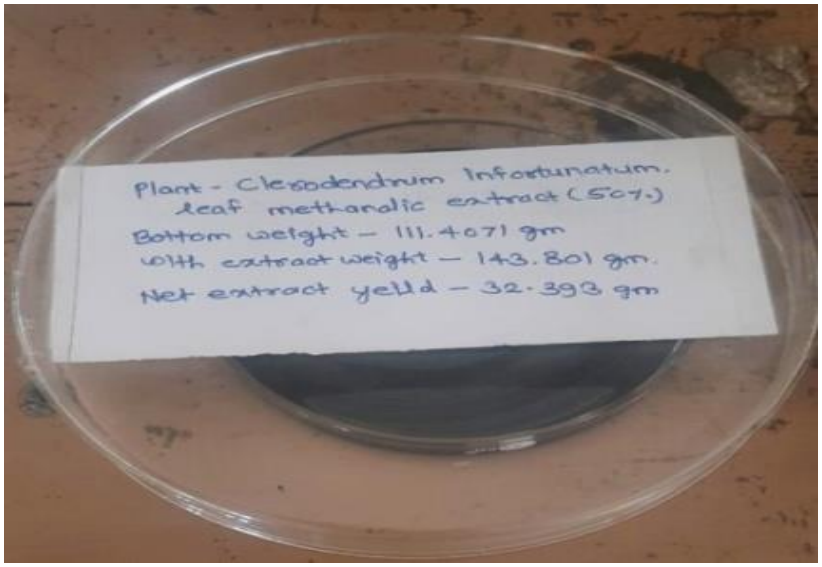


Image 5: 50% Hydromethanolic extract


Image 6: Rotatary vaccum evaporater

ISSN No:-2456-2165

C. Experimental Protocol:

According to the rules of Committee for the Purpose of Control and Supervision of Experimental Animals (CPCSEA), Ministry of Social Justice and Empowerment, Government of India, the protocol was submitted to Institutional Animal Ethical Committee (IAEC) for necessary approval. Total 30healthyeither sex wistar rats of 150-200g used for the study, They originated by the Laboratory Animal Breeding Centre accredited by CPCSEA.

D. Housing

All the animals were kept under standard management conditions as per the norms of CPCSEA. Animals had access to a 12-hour light and dark cycle. Every animal was kept in cages made of polypropylene.

E. Feeding

The animals were maintained on balanced hygienic pellet nourishment and fresh water for consumption for the duration of the trial.

F. Bedding Material

Clean dried rice husk was used as a bedding material.

G. Antipyretic activity (Brewer's yeast method)

Fever elicitation was done by using the Brewer's yeast method was implemented in experimental subjects by administering a 20% yeast suspension subcutaneously, with a dosage of 10 mL/kg, following an overnight fasting regimen (Emon *et al*., 2021). The baseline temperature was meticulously documented through the rectal route using a digital thermometer. Subsequently, after an elapsed period of 18 hours, the animal models exhibiting a rectal temperature elevation within the range of 0.2 – 0.4 °C (32.45 to 32.85 °F) were selected for antipyretic activity assessment.

To conduct the test, the plant extract at varying concentrations (50, 100, 200, and 400 mg/kg body weight) was administered, alongside paracetamol (10 mg/kg) as the benchmark drug. Following this intervention, rectal temperatures were systematically recorded at intervals of 30 minutes, 60 minutes, 120 minutes, and 180 minutes (Abena *et al.*, 2003), with each reading representing the mean value derived from three measurements.

Image 7: Subcutaneous administration of Brewer's yeast

Image 8: Oral administration of drug

ISSN No:-2456-2165

Image 9: Rectal temperature of rat

H. Statistical analysis

The data was analyzedfor statistical significance by using ICAR's WASP 2.0 software and Microsoft excel for appropriate designs. P-values less than 0.01 were considered significant.

III. RESULTS AND DISCUSSION

During receiving brewer's yeast suspension subcutaneously, the internal temperature of the rectal area significantly increased after eighteen hours. After giving the rats that were medicated an oral dosage of paracetamol (10 mg/kg, i.p.), 50% methanolic, and 50% ethanolic (400 mg/kg), they experienced a significant ($p < 0.01$) reduction in pyrexia. 50% ethanolic and 50% methanolic at a dose rate of 400 mg/kg have been shown to reduce pyrexia to the greatest extent possible; this is comparable to the recommended dosage of paracetamol (10 mg/kg).

Table 1: Effect of ethanolic and methanolic extract of *Clerodendrum infortunatun* Linn. On Brewer's yeast induced Pyrexia in Wistar Rats

Sr no	**Drug**	**Dose (mg/kg**	**Temp .of Apyretic rats(F)**			
			30 min	**60 min**	**180 min**	**360 min A.V.**
1	Normal control (T1)		97.083	97.4167	97.4667	97.7
2	Pyretic control (T2)		97.9	97.2667	97.4	97.4667
3	Std treatment (T3)	10	97.816	98.1667	97.9	97.0833
4	50% Ethanolic (T4)	400	97.2666	97.0833	97.5333	97.95
5	50% methanolic(T5)	400	97.0833	98.0333	97.6167	97.7833

(this table shows temperature change in four intervals of time mice of the control group and other experimental groups)

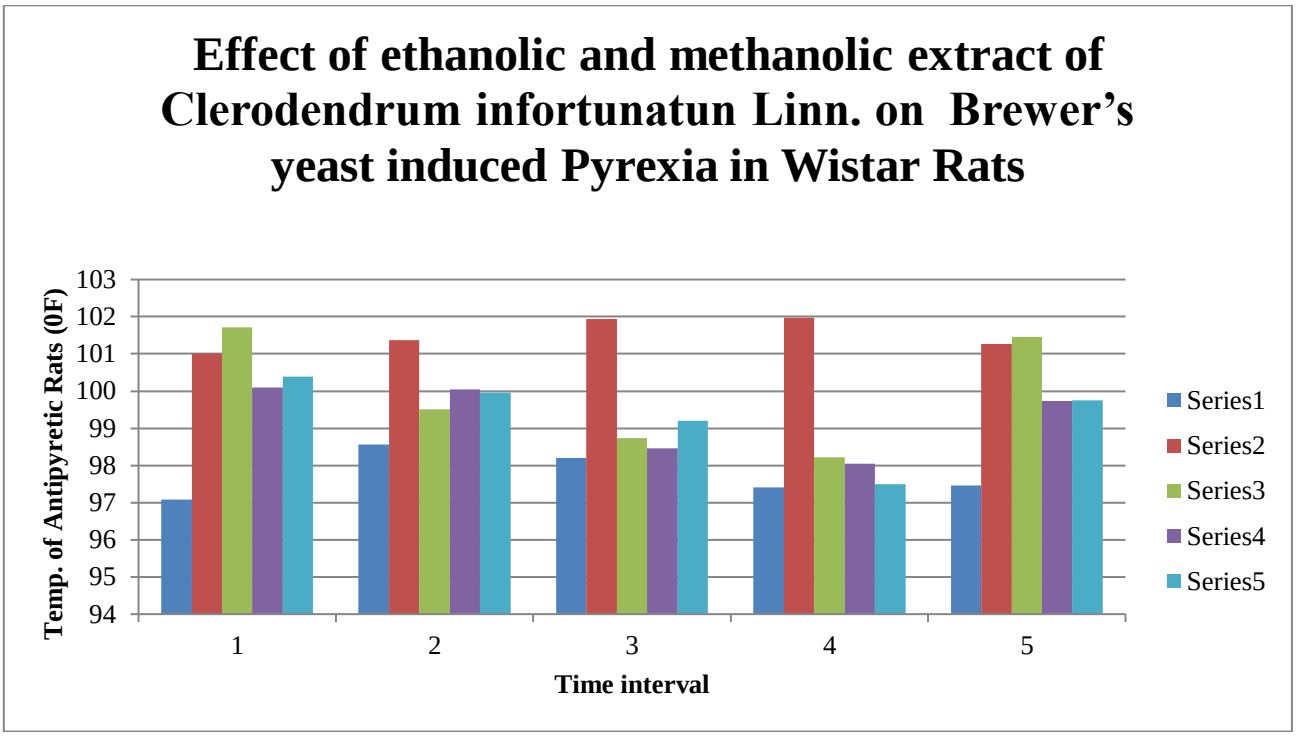
Table 2: Comparison of Antipyretic Activity: Normal Control, Pyretic Control, Standard Treatment, 50% Methanolic Extract, and 50% Ethanolic Extract.

Temp. of pyretic rats (F)	**Temp after drug administration**				
	30 min	**60 min**	**180 min**	**360 min**	**P value**
97.0833	98.56	98.2	97.41	97.46	0.01
101	101.367	101.933	101.967	101.267	0.01
101.717	99.5167	98.7333	98.2167	101.45	0.01
100.1	100.05	98.46667	98.05	99.73333	0.01
100.38	99.95	99.2	97.5	99.75	0.01

(Table 2: This ANOVA table would help determine whether there are statistically significant differences in body temperatures among the treatment groups at various time points after drug administration)

ISSN No:-2456-2165

Graph 1: Temporal Comparison of Antipyretic Activity in Wistar Rats Treated with Clerodendrum infortunatum Extracts.



Graph 1: Comparision of a antipyretic activity with control,standard drug and treatment groups.(on X axis indicates series 1-normal control, series 2-Pyretic control, series 3-Standard treatment, series 4-50% methanolic extract, series 5-Ethanolic extract;1,2,3,4,5 indicates time intervals 30 min, 60 min, 120 min, 180 min resp. Y axis indicates rectal temperature in oF)

IV. CONCLUSION

The purpose of the research investigation was to support *C. infortunatum's* traditional use as a potential antipyretic. Both the extracts significantly restored the elevated body temperature at all the time interval and the result obtained was comparable with standard drug paracetamol. The findings of the study indicate that the plant *Clerodendrum infortunatum* possesses antipyretic activity and further investigations are necessary to confirm the activity. To guarantee the effectiveness and secure usage characteristics of certain phytocompounds, more in-depth biology and therapeutic investigation on the tested compounds is necessary.

Furthermore, more research is necessary to confirm *C. infortunatum*'s potential as an antipyretic drug for the well-being of animals and to identify the precise process behind its antipyretic effect.

ACKNOWLEDGEMENT

The authors are highly thankful to the Nagpur Veterinary College, Nagpur for providing necessary facilities for this work.

- **How to cite this article:** Nikam, A., Mandade, R. P., More, G. V. (2023). Evaluation of antipyretic activity of Ethanolic and Methanolic extract of leaves of *Clerodendrum infortunatum*
- **Source of support**: Nil
- **Conflict of interest:** None

REFERENCES

[1]. Abena, A. A., Diatewa, M., Gakosso, G., Gbeassor, M., Hondi-Assah, T. H., & Ouamba, J. M. (2003). Analgesic, antipyretic and anti-inflammatory effects of essential oil of *Lippia multiflora.Fitoterapia*, *74*(3), 231-236.

[2]. Al-Snafi, A. E. (2016). Medicinal plants possessed anti-inflammatory antipyretic and analgesic activities (part 2)-plant based review. *Sch Acad J Pharm*, *5*(5), 142-158.

[3]. Bhattacharjee, D., Das, A., Das, S. K., & Chakraborthy, G. S. (2011). *Clerodendrum infortunatum* Linn.: A review. *J Adv Pharm Healthcare Res*, *1*(3), 82-85.

[4]. Bhattacharya, A., Behera, R., Agrawal, D., Sahu, P. K., Kumar, S., & Mishra, S. S. (2014). Antipyretic effect of ethanolic extract of *Moringa oleifera* leaves on albino rats. *Tanta Medical Journal*, *42*(2), 74.

[5]. Blatteis, C. M. (2006). Endotoxic fever: new concepts of its regulation suggest new approaches to its management. *Pharmacology & therapeutics*, *111*(1), 194-223.

[6]. Emon, N. U., Alam, S., Rudra, S., Haidar, I. K. A., Farhad, M., Rana, M. E. H., & Ganguly, A. (2021). Antipyretic activity of *Caesalpinia digyna* (Rottl.) leaves extract along with phytoconstituent's binding affinity to COX-1, COX-2, and mPGES-1 receptors: *In vivo* and *in silico* approaches. *Saudi journal of biological sciences*, *28*(9), 5302–5309. https://doi.org/10.1016/j.sjbs.2021.05.050

[7]. Gaetano, L., Watanabe, K., Barogi, S., & Coceani, F. (2010). Cyclooxygenase-2/microsomal prostaglandin E synthase-1 complex in the preoptic-anterior hypothalamus of the mouse: involvement through fever to intravenous lipopolysaccharide. *Acta physiologica*, *200*(4), 315-324.
[8]. Kokate, A., Limsay, R. P., & Dubey, S. A. (2022). The phytochemistry and antioxidant activity of methanolic extract of *Clerodendrum infortunatum* leaves.
[9]. Limsay, R. P., Somkuwar, A. P., Bonde, S. W., Kakde, V. K., Shirankar, S. S., Borekar, V. I., & Jagdale, R. A. (2021). In vitro anti-inflammatory activity of *Clerodendrum infortunatum* L. leaves.
[10]. Muhammad, N., Saeed, M., & Khan, H. (2012). Antipyretic, analgesic and anti-inflammatory activity of *Viola betonicifolia* whole plant. *BMC complementary and alternative medicine*, *12*, 1-8.
[11]. Pal, D., Sannigrahi, S., & Mazumder, U. K. (2009). Analgesic and anticonvulsant effects of saponin isolated from the leaves of *Clerodendrum infortunatum* Linn. in mice.
[12]. Ram, P., Limsay, R. P., Somkuwar, A. P., Adey, K. S., & More, G. Y. (2023). Studies on Phytochemistry and Wound Healing Activity ofLeaf Extract Ointment of *Clerodendrum infortunatum* inCanines. *Indian Journal of Veterinary Sciences & Biotechnology*, *19*(5), 37-41.
[13]. Roth, J., & Blatteis, C. M. (2014). Mechanisms of fever production and lysis: lessons from experimental LPS fever. *Compr Physiol*, *4*(4), 1563-604.
[14]. Safari, V. Z., Ngugi, M. P., Orinda, G., & Njagi, E. M. (2016). Anti-pyretic, anti-inflammatory and analgesic activities of aqueous leaf extract of Urtica dioica L. in albino mice. *Med Aromat Plants*, *5*(237), 2167-0412.
[15]. Shrivastava, N., & Patel, T. (2007). *Clerodendrum* and healthcare: an overview. *Medicinal and aromatic plant science and biotechnology*, *1*(1), 142-150
[16]. Shukla, S., & Mehta, A. (2015). In vivo anti-inflammatory, analgesic and antipyretic activities of a medicinal plant, *Caesalpinia bonducella* F. *Pakistan journal of pharmaceutical sciences*, *28*.
[17]. Sultana, S., Asif, H. M., Akhtar, N., & Ahmad, K. (2015). Medicinal plants with potential antipyretic activity: A review. *Asian Pacific Journal of Tropical Disease*, *5*, S202-S208.
[18]. Uddin, M. J., Çiçek, S. S., Willer, J., Shulha, O., Abdalla, M. A., Sönnichsen, F., & Zidorn, C. (2020). Phenylpropanoid and flavonoid glycosides from the leaves of *Clerodendrum infortunatum* (Lamiaceae). *Biochemical Systematics and Ecology*, *92*, 104131.