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Review on Antipyretic And Antioxidant Activity Of Methanolic Extract Of Little Ironweed Cyanthillium Cinerum

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ABSTRACT

In this study, the ameliorative antipyretic and antioxidant potential of methanolic extract of Cyanthillium cinerium (L.) H. Rob. leaf extract (MECC) was investigated. Methanol was used, to extract the biologically active components from the plant sample by adopting hot continuous soxhlet extraction method. Phytocompounds such as alkaloids, carbohydrates, saponins and phenolic compounds were chiefly identified in the methanolic extracts. The in vivo antipyretic activity was determined by brewer's yeast induced pyrexia method. The quantity of total phenolic and flavonoid content was found to be in methanol extract 76.59 mg/g and 31.67 mg/g. Antioxidant radical scavenging activity conducted with DPPH (2,2-diphenyl-1-picrylhydrazyl), ABTS (2,2' azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)) and phosphomolybdenum assay for the crude solvent extracts showed potent results. The DPPH % inhibition expressed by methanol extract was IC50 44.35 µg/mL and ABTS % inhibition was found to be IC50 3.52 µg/mL. The free radical scavenging potential of methanol extract with the phosphomolybdenum assay showed highest absorbance of 0.794nm at 60 µg/mLThe antipyretic result showed that the methanolic extract of Cyanthillium cinereum are non-toxic and possess significant antipyretic effect which may attribute in the presence of flavonoid and saponin in the extract..

KEYWORDS: Phytochemical Analysis, DPPH, ABTS, Free Radical Scavenging, Anti Pyretic Activity.

1.INTRODUCTION

The Cyanthillium cinereum (L.) H. Rob. is a weed crop and commonly referred as little ironweed. It belongs to the Angiosperm family Compositae (Asteraceae). The plant is extensively found throughout the world's tropical and subtropical climates. Leaves are rich in medicinal properties and used as analgesic, anti-pyretic, anti-bacterial and anti-fungal agents (Yusoff et al., 2020). Due to its enormous medicinal value, it is traditionally used in Ayurveda to treat fever. The plant decoction or infusion provides remedy for spasms of the urinary bladder and strangury. Additionally, it may be used as a treatment for impotency, asthma, cancer, cholera, colic pain, cough, diarrhea, and dysentery. (Joshi et al. 2021). Iwalewa et al. (2003) reported that Cyanthillium cinereum (L.) H. Rob. plant contains antioxidant compounds like tannins, catechins, and flavonoids which shields 2,2′-azobis(2-amidinopropane) dihydrochloride (AAPH) oxidation in human red blood cells.

Certain phytocompounds, like phenolic, vitamin, amino acid, and mineral compounds, may be employed as possible treatments for chronic illnesses like cancer and the heart. Methanol is commonly used for phenolic compounds extraction. Naturally occurring antioxidants are used in foods or medicinal materials to replace synthetic antioxidants, which are being restricted due to their carcinogenicity. Antioxidants prevent certain types of chemical damages caused in cells by an excess of free radicals, charged molecules produced due to smoking, exposure to pesticide and fumes from exhaust. Free radicals annihilation gives protection against cancer, heart diseases, stroke and other immune compromising diseases (Yi-Fang et al. 2002; Arouma, 2003).

The final step in preliminary validation of medicinal drugs involves induction of disease or disease−like conditions in suitable model animals and use of the extracted compounds to ascertain their effect on treatment. To validate the antipyretic effects of plant derived medicinal compounds, the brewer's yeast induced pyrexia has been extensively used (Alam et al., 2008) Hence, the aim of the present work was to analyse the phytocompound characters and antipyretic activity of Cyanthillium cinereum (L.) H. Rob. plant extracts obtained using methanol solvents, and evaluate its chemical constituents for antioxidant and antipyretic activity.

1.1 FEVER

Fever is a complex physiologic response triggered by infectious or aseptic stimuli, normally caused as a secondary impact of infection, tissue damage, inflammation, graft rejection, malignancy or other diseased states. It is the body's natural defense to create an environment where infectious agent or damaged tissue cannot survive. The infected or damaged tissue initiates the enhanced formation of pro-inflammatory mediators (cytokines like interleukin 1β, α, β and TNF-α), which increase the synthesis of prostaglandin E2 (PGE2) near preoptic hypothalamus area and thereby triggering the hypothalamus to elevate the body temperature (Purssell et al., 2007).

Fever is a common medical sign reflected by an elevation of temperature. In human it means a temperature above the normal range of 36.5–37.5°C. Fever occurs with the increase in the concentrations of prostaglandin E2 (PGE2) in the hypothalamus thus altering the firing rate of neurons that control thermoregulation in the hypothalamus. Fever leads to the disturbance of metabolism including: increase in blood pressure, pulse rate, cardiac output, respiration rate among others hence the need to eliminate the fever using the antipyretic agents (Kumar et al., 2011)

Pyrexia is the Greek word, pyr means fire. Febrile is the Latin word, febris meaning fever. Due to the increase in body temperature, there is also an increase in the muscle contractions which cause a feeling of cold. Due to this, there is a production of a large amount of heat and efforts to conserve heat (Rajasekaran et al., 2010). When the temperature of the body returns to its normal range, then a person feels hot and may begin to sweat. Sometimes in the case of young children, the fever may trigger a febrile

1.1.1. Range For Normal Temperatures

A wide range for normal temperatures is found. Central temperatures, such as rectal temperatures, are much more precise than peripheral temperatures (Fodouop Chegaing et al., 2020). Fever is mostly agreed to be present if the elevated temperature is caused by a raised set point and:

Temperature in the anus (rectum/rectal) is at or over 37.5–38.3 °C (99.5–100.9 °F)

Temperature in the mouth (oral) is at or over 37.7 °C (99.9 °F)

Temperature under the arm (axillary) or in the ear (tympanic) is at or over 37.2 °C (99.0 °F)

1.1.2 Hyperyrexia

It is an extreme elevation of body temperature. Hence it is considered as a medical emergency condition. It may indicate a serious underlying condition or lead to problems including permanent brain damage, or death. The most common cause of hyperpyrexia is an intracranial hemorrhage. Other possible causes include Kawasaki syndrome, sepsis, neuroleptic malignant syndrome, drug overdose, serotonin syndrome, and thyroid storm (B. P. Devi et al., 2003).

1.1.3 . Hyperthermia

It is an example of a high temperature. It is not a fever. It occurs due to several causes including heatstroke, neuroleptic malignant syndrome, malignant hyperthermia, stimulants such as substituted amphetamines and cocaine, idiosyncratic drug reactions, and serotonin syndrome. Infections are the most important cause of fever

1.2 Types Of Fever

Types of fever with temperature condition

1. **Continuous fever:** When the body temperature above the normal one throughout the day and it does not fluctuate more than 1ºC in 24 hours.
2. **Remittent fever:** When the body temperature above the normal one throughout the day and it fluctuates more than 2ºC in 24 hours.
3. **Intermittent fever:** The temperature elevation is present only for a certain period, after that cycling back to normal.
4. **Hectic or Septic fever:** When the body temperature variation is more than 5ºC
5. **PelEbstein fever:** When the body temperature may take three days to rise and remains at a higher level for three days and remits in three days, followed by apyrexia for nine days.
6. **Low-grade fever:** When the body temperature increases a little bit in the evening for some days but does not exceed 37.8ºC. This is the indication of tuberculosis.
7. **Stepladder fever:** When the body temperature increases step by step to a higher range.
8. **Relapsing fever:** The pyrexial condition is separated by intervals of normal temperature and it may remain from 2 - 9 days.
9. **Tertian fever:** When the temperature of the body increases once in 48 hours.
10. **Quartan fever:** When the temperature of the body increases once in 72 hours.
11. **Double quotidian:** When the temperature of the body rises twice in a day, once in the morning and once in the evening.
12. **Inverse fever:** When the temperature of the body rises early in the morning.

1.2.1 Pathophysiology of Fever

Fever is recognized as a complex, coordinated, autonomic, behavioral response and neuroendocrine which occurs due to acute phase reaction to immune challenge. Natural defence system of the human body is activated whenever body finds any infectious agent in order to create an unfavourable environment for the survival of infectious agent. The infectious agent or damaged tissues initiate the increase production of proinflammatory mediators cytokines such as interleukin 1β, β, α and TNF-α which enhance the formation of prostaglandin E2 (PGE2) near the peptic hypothalamus area and the prostaglandin in turn act on the hypothalamus to elevate the body temperature(Mirrasekhian et al., 2020; Mota-Rojas et al., 2021).

1.2.2 Molecular Mechanism Of Fever

Distinct members of Toll receptors in macrophages receptor family recognize different and specific microbial components, but biosynthesis and releases same endogenous pyrogens, such as IL-1β, TNF, and IL-6.

Cytokines which are transported by the bloodstream could act at sites lacking a tight blood–brain barrier, the so-called circumventricular organs (Evans et al., 2015). Alternatively, circulating cytokines could interact with their receptors on brain endothelial cells or perivascular cells and thereby stimulate these cells to release pyrogenic mediators into the abluminal brain tissue. It has been proposed that fever-promoting cytokines are transported from the blood into the brain via specific carriers. An assumed manifestation of a febrile response produced by these mechanisms is termed as the humoral hypothesis of fever induction (Flier et al., 1994).

These pyrogenic cytokines acts on organum vasculosum area of the brain known as laminae terminalis leading to activation of the enzyme cyclo-oxygenase-2 (COX-2) that results in release of prostaglandin E2 (PGE2), which binds to receptors in the hypothalamus leading to an increase in heat production and a

decrease in heat loss until the temperature in the hypothalamus reaches an elevated set point.

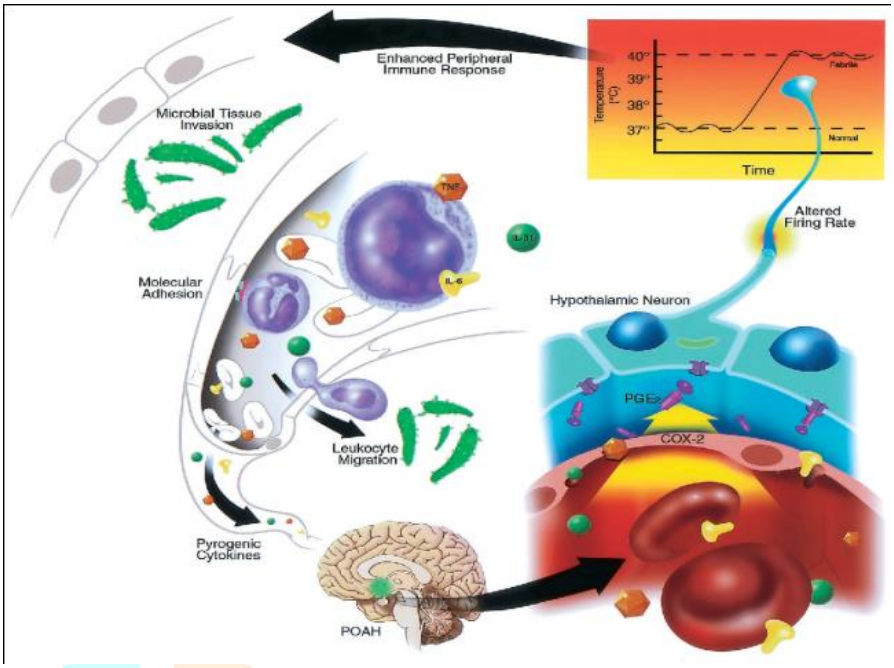

Fig 1 **Molecular mechanism of fever**

1.2.3 The pathogenesis of fever

Many of the mediators underlying pyrexia have been described in recent years (Figure 4). The critical "endogenous pyrogens" involved in producing a highly regulated inflammatory response to tissue injury and infection are polypeptide cytokines. Pyrogenic cytokines, such as interleukin- 1β (IL-1β), tumor necrosis factor (TNF), and interleukin-6 (IL-6), are those that act directly on the hypothalamus to effect a fever response (Mota-Rojas et al., 2021). Exogenous pyrogens, such as microbial surface components, evoke pyrexia most commonly through the stimulation of pyrogenic cytokines. The gram-negative bacterial outer membrane lipopolysaccharide (endotoxin), however, is capable of functioning at the level of the hypothalamus, in much the same way as IL 1β

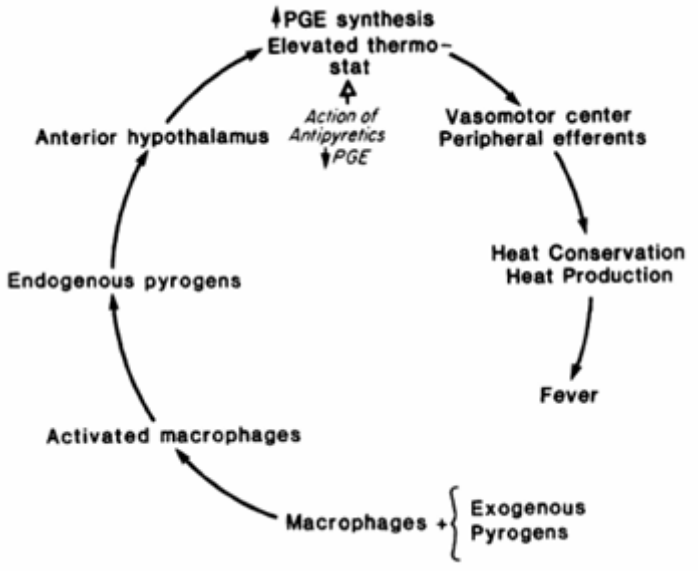

Fig 2: Pathogenesis of fever

2. MATERIALS AND METHODS

2.1. Plant material

The healthy leaves of Cyanthillium cinereum (L) H. Rob. Belongs to the family Asteraceae were collected from the roadsides of Nagpur, Maharashtra, India. In this study we use the leaf of Cyanthillium cinerum Fresh, healthy leaves samples were washed twice under running tap water and then rinsed with distilled water, to remove the external debris adhered on its surface. The washed plant material was shade dried at room temperature to get rid of residual moisture. The dried plant sample was cut into smaller pieces and pulverised into a fine powder. The powdered plant sample was sieved and was stored in an air tight sterile container and later used for the preparation of plant extracts.

Fig.3.a. habitat of Cyanthillium cinerium

3.b. Leaves of Cyanthillium cinerium

All the chemicals used were of analytical grades, obtained from commercial suppliers. Paracetamol was obtained as gift sample from Mepro Pharmaceuticals Pvt. Ltd., Surendranagar, Gujarat, India. Brewer's yeast (Sigma-Aldrich). Double distilled water from all-glass still was employed throughout the study.

2.1.1Taxonomical classification

Kingdom: Plantae
Division: Mangnoliphyta
Class: Magnoliopsida
Subclass: Asteridae
Order: Asterales
Family: Asteraceae
Genus: Cyanthillium
Species: C. cinereum

Synonyms: Ironweed, common vernonia, ash-colored fleabane, goat weed, iron weed, purple fleabane, Vernonia cinerea, Vernonia abbreviata, Vernonia arguta, Conyza cinerea, Serratula cinerea

2.2.2 Botanical Description

Cyanthillium cinereum, also known as little ironweed, is an annual herb that can grow up to 120 cm tall (Suja et al., 2016) the following botanical description:

- **Growth form:** Herbaceous plant that can grow up to 1 m tall, but is usually found as a small herb about 0.3 m tall
- **Foliage:** Green leaves that are oval with shallow teeth along the leaf margin, and are arranged alternately with one leaf per node
- **Flowers:** Composite flowers with white to light purple disc florets and no ray florets
- **Fruit:** Small, brown fruits with fluffy white hairs called "pappus" that help to disperse the fruits by wind.
- **Stems:** Branching above, longitudinally ribbed, thinly puberulent above and coarsely pubescent below
- **Inflorescences:** Tubular capitula that are about 5 mm long, and arranged in loose and terminal corymbs

- **Corolla:** Tubular and develops five narrow lobes

2.2.2 Chemical composition

Phytochemical screening of this plant showed a presence of cardiac glycosides, alkaloids, phenols flavonoids, steroids, tannins, phlobatannins and saponins. Flavonoids and phenols are strong antioxidants and have an important role in the health care system. The flowers of Cyanthillium cinereum contain thymohydroquinone dimethyl ether (5.5%) and -cadinol (4.4%), while the leaves contain cadinol (23.2%), elemol (10.6%), cadinene (9.9%), -muurolol (8.2%), germacrene D-4-ol (6.1%), and terpinen-4-ol (4.9%). The ethanoic extract of Cyanthillium cinereum contains lupeol, lupeol acetate, luteolin-7-O-glucoside, stigmasterol-β-D-glucopyranoside, stigmasterol, and dotriacontanoic acid. Caryophyllene oxide (16.7 %) was the major compound followed by n-hexadecanoic acid (8.9 %) and phytol (7.1 %) (Bindu et al., 2018)

2.2.4 Pharmacological activity

(a) Antibacterial activity

Crude methanolic extract of Cyanthillium cinereum exhibits antibacterial activities against Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa and bacillus subtilis (Suja et al., 2016). The Cyanthillium cinereum plant extract showed significant antibacterial activity. The strong anti bacterial activity of Cyanthillium cinereum plant extract is due to the presence of phytochemical compounds such as flavonoids, phenols, tannins and saponins (Ramya et al., 2021).

(b) Antioxidant activity

Hexane, chloroform, methanolic and aqueous crude extracts of Cyanthillium cinereum exhibited potent antioxidant property. Phytochemical constituents scavenge different free radicals and provide a protective action against oxidative damage to biological macromolecules. The anti oxidant activity exhibited by Cyanthillium cinereum could be due to presence of flavonoids, polyphenolic compounds and tannins (Leelavathi.L, 2023).

(c) Anticancer activity

Crude ethanolic extract of Cyanthillium cinereum were tested for anticancer activity in MCF-7 breast cancer cell lines. The leaf extract of Cyanthillium cinereum exhibited high anticancer activity (R. Ashok Kumar et al., 2011). Cyanthillium cinereum plant extract induced cell death by apoptosis in MCF-7 breast cancer cells (Suja et al., 2016).

(d) Antispasmodic activity

Ethanolic extract of leaves of Cyanthillium cinereum possess high degree of antispasmodic activity by blocking cholinergic receptors. Cyanthillium cinereum exhibits comparable antispasmodic activity of standard antispasmodic agent (Atropine). Cyanthillium cinereum being a herbal origin drug with high degree of safety and efficacy could be used as a suitable alternative for existing drugs (Suja et al., 2016).

(e) Anti-malarial activity

Due to the presence of Sesquiterpene lactones, Cyanthillium cinereum possess anti-malarial activity.

(f) Anti-inflammatory activity and Antipyretic activity

Thailand is well known for the traditional application of aerial parts extracts of C. cinereum for immune-related remedies and anti-inflammation. The alcoholic extract of the flowers of C. cinereum caused a reversal of the inflammatory processes in adjuvant arthritic rats. Further, leaf extracts of C. cinereum have been reported with significant analgesic, anti-inflammatory and antipyretic properties. C. cinereum is also widely used with quinine for its antipyretic benefit on malarial fever (Suresh et al., 2023).

1.2.7 Therapeutic uses of Cyanthillium cinereum (L) H. Rob

Cyanthillium cinereum has therapeutic potentials against;

- Fever
- Malaria
- Asthma
- Cancer
- Cholera, colic pain, cough, diarrhea
- Leprosy and chronic skin diseases
- Conjunctivitis
- Arthritis
- Rheumatism

3.DISCUSSION

In the developing countries increased cost of medicine as well as their side effects has become a great task when public health is concerned. From time to time, investigations have been carried out to develop different types of polyherbal formulations to enhance the overall therapeutic potential of the formulation. And so, nowadays the traditional medical system and their herbal / herbo-mineral preparations used for various ailments are becoming more popular.

The phenolics, particularly polyphenols exhibit a wide variety of beneficial activities in mammals including antiviral, antibacterial, immune stimulating, antiallergic, antihypertensive, ant ischemic, antiarrhythmic, antithrombotic, hypocholesteromic, hepatoprotective, anti-inflammatory, anticarcinogenic actions. Flavonoids are an important group of polyphenols and are reported to inhibit prostaglandin synthesis, which are known mediators of inflammation (Ishan et al., 2021) Fever may be due to infection or one of the sequele of tissue damage, inflammation, graft rejection, or other disease states (Jaiswal et al., 2017). Regulation of body temperature requires a delicate balance between production and loss of heat, and the hypothalamus regulates the set point at which body temperature is maintained. In fever this set point elevates (S. A. Kumar et al., 2015b). Yeast induced fever is called pathogenic fever. Its etiology includes production of prostaglandins, which set the thermoregulatory center at a lower temperature (Taher et al., 2015).

4. CONCLUSION

The present study supports that methanolic extract of Cyanthillium cinereum (MECC) have significant antipyretic activity. In this study Cyanthillium cinereum whole plant extracts obtained with methanol solvents were analysed for its potential antioxidant and antipyretic activities. The extracts screened for preliminary phytochemicals showed the presence of alkaloids, carbohydrates, saponin and phenolic compounds. The extracts obtained with methanol solvents had high free radical scavenging activities compared with their respective standard. The study concluded that the whole plant methanol extract had therapeutic potential with antipyretic efficiency against pathogenic microorganisms.

On the basis of the study the formulation showed significant in vitro anti-oxidant activity by terminating the actions of free radicals.. Antibacterial activity was evaluated via disk diffusion method. The potential activity of extract may be due to the presence of phenols, flavonoids and other phytochemical constituents present in it.

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