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Evaluation of analgesic and antipyretic activity of ethanol extract of *Oxalis corniculata* Linn.

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ABSTRACT

To evaluate the analgesic and antipyretic activity of ethanol extract of *oxalis corniculata* Linn. Analgesic activity was carried out using acetic acid induced writhing method in swiss albino mice (25-30 gm),and antipyretic activity done by Brewer's yeast induced pyrexia in rast at the dose level of 200mg/kg p.o and400mg/kg p.o. Oral administration of ethanol extract of *oxalis corniculata*(EEOC) significantly reducing ($p<0.01$) the number of writhing and significantly ($p<0.01$)reduced pyrexia 1 to 4 hr compared to 0h of the same group animal at the dose level of 200mg/kg p.oand400mg/kg p.o.when compared to control . The result of pharmacological test was performed in the present study suggest that (EEOC) posses potent Analgesic and Anti pyretic activity. hence we can consider EEOC is better alternative for the treatment than the current treatment regimen NSAID and Analgesic drug.

Keywords: *Oxalis corniculata,* Brewer's yeast, Acetic acid, NSAID, Analgesic activity, pyrexia.

1. INTRODUCTION

Pain is an unpleasant sensory or emotional experience associated with actual or potential tissue damage, or described in terms of such damage. Pain is always subjective. It is unquestionably a sensation in a part of the body, but it is also unpleasant, and therefore also an emotional experience.[1]Typically, it is a direct response to an untoward event associated with tissue damage, such as injury, inflammation or cancer, but severe pain can arise independently of any obvious predisposing cause (e.g. trigeminal neuralgia), or persistent long after the precipitating injury has healed (e.g. phantom limb pain). It can also occur as a consequence of brain or nerve injury (e.g. following a stroke or herpes infection). With many pathological conditions, tissue injury is the immediate cause of the pain, and this result in the local release of a variety of chemical agents, which are assumed to act on the nerve terminals, either activating them directly or enhancing their sensitivity to other of stimulation[2].

Pyrexia is caused as a secondary impact of infection , malignancy or other diseas states.[3] It is the body natural defence to create an environment where the micro organisum enter in to the body and its damaged to the tissue. Normally the infected or damaged tissue initiate the enhanced formation of pro inflammatory mediators (cytokines like interlukin1β, α,β and TNF α) Which increase the synthesis of PGE2 near peptic hypothalamus area and there by triggering the hypothalamus to elevate the body temp. As the temp regulatory system is governed by a nervous feed back mechanisum, so when body temp become high , its dilate the blood vessels and increasing the sweating to reduce the temp ,but when body temp become very low hypothalamus protect the initial temp by vasoconstriction. most of the antipyretic drug inhibit the COX-2 expression to reduce the elevated body temp by inhibiting PGE2 biosynthesis more over the synthetic agent irreversibly inhibit COX 2 with high selectivity but more toxic to hepatic cell, brain and heart muscle, whereas natural COX-2 have low selectivity with fever side effect.[4] *oxalis corniculata* is small plant found in river side and agricultural land in India. the whole plants has been used traditionally such as Antimicrobial, astrigent, cardiotonic febrifuge, hypotensive, Stomachi, vasodilator and immunomodulatory, Analgesic, fever, hyperlipidemia, Anti oxidant.[5-7]A bibliographic survey showed that there are no reports on the analgesic and anti-pyretic activity of *oxalis corniculata*. This prompted us to investigate the effects of pharmacological activities of *Borassus flabellifer* in experimental models of analgesia and pyrexia.

2. MATERIALS AND METHODS

2.1. Collection of plant materials

The whole plant of *oxalis corniculata* Linn ,were collected from various parts of salem district, Tamilnadu during the month of july to September and were authenticated from ABS garden salem.

2.2. Extraction procedure

Whole plant *ofoxalis corniculata* were collectedand was dried for7 days . After drying they were again pulverized. The size is reduced. The dried whole plant of *oxalis corniculata* powder mixture was weighed about 500g Extracted by soxhlet apparatus using 90% of ethanol as a solvent for 72 hours. The yield of product was 6.825% w/w.

2.3. Experimental Animals

Swiss Albino Mice (25-30g) and Albino Rats (180-250g) of either sex were used in the study. They were procured from Venkateshwara Enterprises, Bangalore, Karnataka, India. They were randomly distributed into groups and housed in cages (6 per cage) and maintained under standard conditions at 26 ± 2°C and relative humidity 44–56% and 10h light: 14h dark cycles each day for one week before and during the experiments. All animals were fed the standard rodent pellet diet (Amrut, India) and water ad libitum. This project was cleared by Institutional Animal Ethical Committee.

2.4. Acute oral toxicity studies

Acute oral toxicity study was performed as per Organization for Economic Cooperation and Development (OECD) guideline 423 method. The test substance was administered in a single dose by gavage using specially designed mice oral needle. Animals are fasted 3 h prior to dosing (food was withheld for 3 h but not water). Following the period of fasting animals was weighed and test substance was administered. After the test substance administration, food was with held 2 h in mice. Animals are observed individually after dosing at least once during the first 30 minutes, periodically during the first 24 hrs, with special attention given during the first 4 hrs, and daily thereafter, for a total of 14 days. Animals are removed, if any humanely killed for animals welfare reasons or are found dead. All the observations are systemically recorded. [8-9]

2.5. Analgesic activity [10]

Acetic acid induced writhing method five groups of mice (n = 6) were randomly formed groups were treated as control group which received 1% v/w CMC as vehicle and standard drug is Aspirin (100mg/mL). 4&5 received (200 and 400 mg/kg). Respectively, Acetic acid solution 0.3% v/v (10 ml/kg) was injected by intraperitoneal route one hour after treatment and number of writhes (i.e. index of pain reaction against chemical stimuli characterized by abdominal muscle contraction together with turning of trunk and extension of hind limbs) was counted over a period of 20 min. Analgesic activity was expressed as percentage of inhibition of writhes with respect to the control group. [9]

Group-1: control group which received CMC 1% v/w p.o

Group-2: disease control group which received only Acetic acid(0.3%v/v)i.p

Group-3: Served as standard drug treatment group which received only Aspirin (100mg/mlp.o)

Group- 4: group 4 has received the EEOC 200mg/kg. p.o.

Group-5: group 5 has received EEOC 400mg/kg. p.o.

2.6. Anti Pyretic Activity [11-12]

Animals were selected for four groups the experiment after confirmation of approximate constant rectal temperature for 7 days. The antipyretic activity of the extracts was evaluated based on Brewer's yeast-induced pyrexia in rats. Pyrexia was induced by subcutaneous injection of 10 ml/kg of 15% w/v Brewer's yeast suspension in 0.09% of saline below the nape of the neck. The rectal temperature of each rat was measured at time, 0 h, using a thermometer and before injection of the yeast. At 18 h following yeast injection, the different groups were treated with the vehicle, extracts (200 and 400 mg/kg) and standard drug, paracetamol (150 mg/kg). The rectal temperature was then recorded over a period of 4 hrs**.**

Group-1: disease control group which received only brewar'syeast(15% v/w)

Group-2: Served as standard drug treatment group which received only Paracetamol (150mg/mlp.o)

Group-3: has received the EEOC 200mg/kg.p.o.

Group-4: has received EEOC 400mg/kg.p.o.

2.7. Statistical analysis

The values are expressed in mean ± SEM. One way ANOVA followed by Dunnet's multiple comparison Test was used to analyse the effect of different doses of drugs when compared to control, with the help of Graph Pad Instat software, version 3.01. $P<0.01$ is considered as significant. All data are expressed in mean ± SEM.

** P<0.01, *P<0.05ns One way ANOVA followed by Dunnet'smultiple comparison test with control.

Table - 1: Preliminary Phytochemical Screening Of *oxalis corniculata*

Name of the Phytoconstituents	EEOC extract
Carbohydrates	+
Total Phenolic substances	+
Glycosides	+
Alkaloids	-
Proteins and Amino acids	+
Flavanoids	+
Tannins	+
Phytosterols	+
Gums and Mucilage	-
Phenols	+
Terpenoids	-
Saponins	-
Fixed oil and Fats	-
Volatile oil	+

Table - 2: Analgesic Activity Of EEOC by acetic acid Induced writhing methods

Group	Dose level(mg/kg p.o)	No.of writhing in 20 min
Normal control	1% v/w CMC	30.66±2.58
Disease control	10 ml/kg	12.66± 1.751
Aspirin	100 mg/kg	24.2±3.64***
EOC	200 mg/kg	17.16±1.42**
EOC	400 mg/kg	19.7±1.862**

All data are expressed in mean ± SEM. ** P<0.01, *P<0.05ns One way ANOVA followed by Dunnet'smultiple comparison Test with control.

Table - 3: Antipyretic Activity Of EEOC By yeast induced pyrexia Methods

Group	-18h	0h	1h	2h	3h	4h
CONTROL	37.4±0.172	38.28±0.04	38.41±0.09	38.84±0.07	38.93±0.06	39.07±0.05
STANDARD	37.36±0.68	38.66±0.62	37.51±0.17	37.21±0.292	37.18±0.61	37.08±0.32
EOC	37.08±0.0.257	38.56±0.12	37.68±0.10	37.58±0.14	37.51±0.03	37.01±0.160
EOC	37.16±0.07	38.21±0.16	38.01±0.23	37.63±0.15	37.26±0.11	37.18±0.613

All data are expressed in mean ± SEM. ** P<0.01, *P<0.05ns One way ANOVA followed by Dunnet'smultiple comparison Test with control.

3. RESULTS

3.1. Preliminary Phytochemical Screening

In this study ethanol extract of EOC (ethanol extact of oxalis corniculata)showed positive to following phytochemical constituent's alkaloids, carbohydrates, tannins, flavonoides, total phenolic substances, glycosides, amino acid and proteins and terpenoids. The results are tabulated in table 1

3.2. Analgesic activity

The effect of extract on acetic acid induced writhies in mice have been shown in table 2.aspirin 100 mg/kg and EEOC at the dose level of (200mg/kg and 400 mg/kg) significantly (P<0.01) reduced the pain when compared to the disease control group.

3.3. Anti pyretic activity

The antipyretic effect of *oxalis corniculata* have been shown in table 2. EEOC 200mg/kg and paracetamol 150 mg/kg after administration significantly (p<0.01) reduced pyrexia 1 to 4 hr compared to 0h of the same group animal. EEOC 400 mg/kg it will start to decrease the fever on 2h to 4h compared to the 0h of the same group animal.

5. DISCUSSION

Increased body temperature and pain are two major sign of the body against inflammation. A drug with anti-inflammatory activity usually also exhibit antipyretic and analgesic activity. the best example would be the NSAID drug which posses all three activity. [13-15]

Acetic acid induced writhing method of central and pheripheral analgesic activity.table showed aspirin 100 mg/kg and EEOC at the dose level of (200mg/kg and 400 mg/kg) significantly (P<0.01) reduced the pain when compared to the disease control group. Pain is a complex mediator (eg) PG, bradykinins, sub-p etc. acetic acid induced writhing model in animal the abdominal construction will produced by acetic acid because its releasing the PGE2 and PGF2 in the peritoneal fluid. So that the EEOC suppress the formation of

inflammatory mediator. So that the EEOC suppress the pain.

Subcutaneous injection of Brewer's yeast induces pyrexia by increasing the synthesis of prostaglandin. It is considered as a useful test for the screening of plant materials as well as synthetic drugs for their antipyretic effect. [16,17] Yeast-induced pyrexia is called pathogenic fever and its etiology could be the production of prostaglandins. The inhibition of prostaglandin synthesis could be the possible mechanism of antipyretic action as that of paracetamol and the inhibition of prostaglandin canbe achieved by blocking the cyclooxygenase enzyme activity. There are several mediators for pyrexia and the inhibition of these mediators is responsible for the antipyretic effect [17].

Normally the infected or damaged tissue initiate the enhanced formation of pro inflammatory mediator;s (cytokines like interlukin 1β ,α,β and TNF α) which increase the synthesis of PGE2 near peptic hypothalamus area and there by triggering the hypothalamus to elevate the body temp Most of antipyretic drug inhibit COX -2 expression tyo body temperature by inhibition PGE2 synthesis.[35]EOC 200mg/kg and paracetamol 150 mg/kg after administration significantly ($p<0.01$)reduced pyrexia 1 to 4 hr compared to 0h of the same group animal. EOC 400 mg/kg it will start to decrease the fever on 2h to 4h compared to the 0h of the same group animal.

From the Preliminary phytochemical screening result ethanol extracts of oxalis corniculata showed the following phytochemical constituents such as total phenolic substances, glycosides, alkaloids, flavanoids, tannins, terpenoids and saponins gave positive ,due to the presence of phytochemical constituents ,which might be in part responsible for the analgesic and anti pyretic activity. [18]

The result was conclude that EEOC will be safer drug in the long term treatment. hence we can consider EEOC is better alternative for the treatment than the current treatment regimen NSAID and Analgesic drug.

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