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**EVALUATION OF ETHANOLIC EXTRACT OF  
PORTULACA TUBEROSA ROXB LEAVES FOR  
ANTIPYRETIC ACTIVITY**

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**Abstract**

*Portulaca tuberosa* Roxb belongs to family of Portulacaceae. Plant's infusion is given internally in dysuria; externally applied to erysipelas traditionally. The herb shows diuretic, calculolytic, analgesic and antipyretic properties. The aerial parts contain diterpenoids, pilosanone A and B. Previously studies showed that the *Portulaca tuberosa* has good effect against diuretic, analgesic, however no work was carried out for in vivo antipyretic activity. The present study was made to investigate the ethanolic extract of *Portulaca tuberosa* leaves for antipyretic activity, compared to that of Aspirin as a reference drug. Pyrexia was induced by s/c injection of aqueous suspension of yeast 20% w/v in normal saline at the rate of 10ml/kg body weight. The experiment was done at the doses of 200 mg/kg, 400mg/kg and 800 mg/kg B/w. Rectal temperatures were recorded for 4 hrs by digital thermometer (Holden medical B.V, Netherlands). The result showed that *Portulaca tuberosa* Roxb has strong  $p < 0.01$  antipyretic activity and this strongly support the ethanopharmacological uses of *Portulaca tuberosa* Roxb as antipyretic agent.

**Keywords:** - : *Portulaca tuberosa* Roxb, Yeast-induced pyrexia, Antipyretic, Aspirin.

**INTRODUCTION**

The genus *Portulaca tuberosa* Roxb" commonly known as Bichhuu-buuti, belongs to family of Portulacaceae. The plant is native to Peninsular India, Sind in West Pakistan; Srilanka (Ceylon) near sea-coasts. A number of active chemical compounds such as tannins, phenol, terpenoids and saponins have been found in genus and exhibits antidiabetic, antimicrobial and neuroprotective properties. The plant is a herb Perennial, prostrate, diffusely branched, fleshy, glabrous, herb. Branches straggling, 4-15 cm long with 2-3 mm long internodes. Root tuberous, somewhat fusiform, 5-8 cm long. Leaves alternate, sessile to subsessile, linear or linear-lanceolate, 8-14 mm long, 1-1.5 mm broad, fleshy, acute; stipular hairs dense, 4-8 mm long, somewhat brownish. Inflorescence a small, sessile cluster of 2-3 flowers subtended by 6-8 leaved involucre. Flowers c. 10 mm across, yellow, surrounded by ring of bracteate hairs akin to stipular hairs. The literature review shown that ethanolic extract of plant has significant in vivo antipyretic effect.

In Ayurvedic system of medicine various parts of plant are used, the local use include analgesic, diuretic and fever. No in vivo antipyretic activity on *Portulaca tuberosa* Roxb has been reported, hence, present study was undertaken to determine the in vivo antipyretic activity of the ethanolic extract of *Portulaca tuberosa* Roxb by yeast induced pyrexia in swiss albino mice.

**2.MATERIALS AND METHODS**

**2.1 Chemicals**

Acetyl salicylic acid (CDH Ltd, New Delhi), Yeast *Saccharomyces cerevisiae* (CDH Ltd New Delhi), Digital thermometer (Dr. Morepen Digital Hard Tip Thermometer (MT 101)), Digital weighing balance (Scientech Zeta Analytical Balances), Absolute ethanol.

## 2.2 Test Animals

The animals used in the study were swiss albino mice (20-30gm), within age 3-4 week of either sex. The animals were kept in cages at animal house, and maintained at room temperature of  $25^{\circ}\text{C}\pm 2^{\circ}\text{C}$  with relative humidity ( $60 \pm 10\%$ ) under 12 hrs night and light cycle. All the animals were kept at overnight fasting before to every experiment. The animals used were approved according to animal ethics committee Teerthanker Mahaveer College of Pharmacy, Teerthanker Mahaveer University, Moradabad, U.P

## 2.3 Collection And Extraction Of Plant

The whole plant of *Portulaca tuberosa* Roxb was collected from District Gaya Bihar, and was authenticated. After washing the leaves, covered with cloth and dried in shade for 20 days at room temperature. The leaves were grinded through mechanical grinder to coarse of powder. The powdered of plant material (250gm) was extracted through maceration technique using 80% ethanol (1:5) as a solvent for 72 hours at room temperature with occasional manual shaking. After maceration, the mixture was then filter through Whatman filter No.1 paper in a flask and tightly capped. The extract was then concentrated under reduce pressure through rotary evaporator and then air dried.

## 2.4 Acute Toxicity Study

Acute toxicity study of the extract was determined on swiss albino mice of either sex. The dose of extract was increased one to three folds to determine the safety level of the extract. The mice were divided into three groups each contained two mice. The first group received only normal saline and the second and third groups received i.p injection of tested drug at doses of 1000 mg/kg and 3000 mg/kg. After administration of doses mice were observed for 72 hrs for any toxic effect.

## 3. ANTIPYRETIC ACTIVITY

The antipyretic effect of the ethanolic extract of *Portulaca tuberosa* Roxb was determined on swiss albino mice (20-30gm). The animals were divided into five groups contained six mice in each. The normal body temperature of each mouse was recorded by using digital thermometer, inserted in rectum at predetermined interval. Fever was induced by subcutaneous injection of yeast 20% w/v in normal

saline at the rate of 10 ml/kg body weight. 15 hrs after the injection of yeast, the rectal temperature of each animal was again recorded by digital thermometer. Only those animals that show a minimum increased of  $0.7^{\circ}\text{C}$  in temperature after injection of yeast were taken for experiments. Aspirin (100 mg/kg, i.p) was used as reference drug. Group 1st received only (10 ml/kg) normal saline i.p, group 2nd received Aspirin (100 mg/kg) as a reference drug. While group 3rd, 4th and 5th group received 200, 400 and 800 mg/kg B/w of HEOM respectively. After drug administration rectal temperature of each animal were then recorded following at 0, 1, 2, 3 and 4 hrs by digital thermometer. Significant decrease in fever in tested animals was compared to control group (Adams, S.S., Hebborn, P. and Nicholson 1998).

## 4. RESULTS

The present study was performed to find out the antipyretic effect of the ethanolic extract of *Portulaca tuberosa* Roxb. The result of the present study showed that the ethanolic extract of *Portulaca tuberosa* Roxb has significant antipyretic effect with a reasonable safe profile.

### 4.1 Statistical analysis

Results were expressed as mean  $\pm$  SEM. The statistical significance between control and treated groups were performed using analysis of variance ANOVA test. For multiple comparisons among the groups Bonferroni test was performed. A probability level of  $p < 0.05$ ,  $p < 0.01$  and  $p < 0.001$  was accepted statistically significant.

### 4.2 Acute toxicity Study

The ethanolic extract of *Portulaca tuberosa* Roxb was tested at two doses, 1000 mg/kg and 3000 mg/kg for toxicity and then compared with control (normal saline group). No major behavioral changes or mortality were noted post administration of the extract at dose of 1000 mg/kg for 72 hrs. While mortality was observed at the dose of 3000 mg/kg of extract for 72 hrs.

### 4.3 Antipyretic effect against yeast induced pyrexia

The antipyretic activity of the ethanolic extract of *Portulaca tuberosa* Roxb was determined by yeast induced pyrexia in mice. The result showed that tested drug at different doses caused lowering of the body temperature upto 4 hrs following its administration

**Table 1: Effect of *Portulaca tuberosa* (200, 400 and 800mg/kg) i.p on Yeast induced pyrexia in mice**

Treatment	Dose mg/kg	Rectal temperatur(C°)						
		After administration of drug						
		Normal (X)	After 15 hrs (Y)	0 hrs	1 hrs	2 hrs	3 hrs	4 hrs
Normal Saline	10ml/kg	35.73 ±0.178	37.88 ±0.111	37.63 ±0.148	37.46 ±0.133	37.36 ±0.136	37.30 ±0.132	37.46 ±0.141
Aspirin (STD)	100mg/kg	35.70 ±0.225	37.96 ±0.133	37.85 ±0.148	37.35 ±0.135	36.65 ±0.177	36.03 ±0.136	35.40 ±0.121
Extract	200mg/kg	35.66 ±0.254	37.73 ±0.199	37.66 ±0.150	36.23 ±0.176	36.96 ±0.131	36.37 ±0.145	36.53 ±0.145****
	400mg/kg	35.31 ±0.248	37.78 ±0.158	37.70 ±0.211	37.33 ±0.165	36.71 ±0.083 *	36.35 ±0.131 ***	36.15 ±0.145 ***
	800mg/kg	35.80 ±0.188	38.10 ±0.086	37.93 ±0.067	37.31 ±0.101	36.70 ±0.093 *	35.93 ±0.117 ***	35.63 ±0.0136****

Value are expressed as mean ± SEM, (number of mice N = 6), significant \*P < 0.05, \*\* P < 0.01 and \*\*\* P < 0.001, when compared to control.

**Figure 1: Antipyretic effect of ethanolic extract of *Portulaca tuberosa* on yeast induced pyrexia in mice**

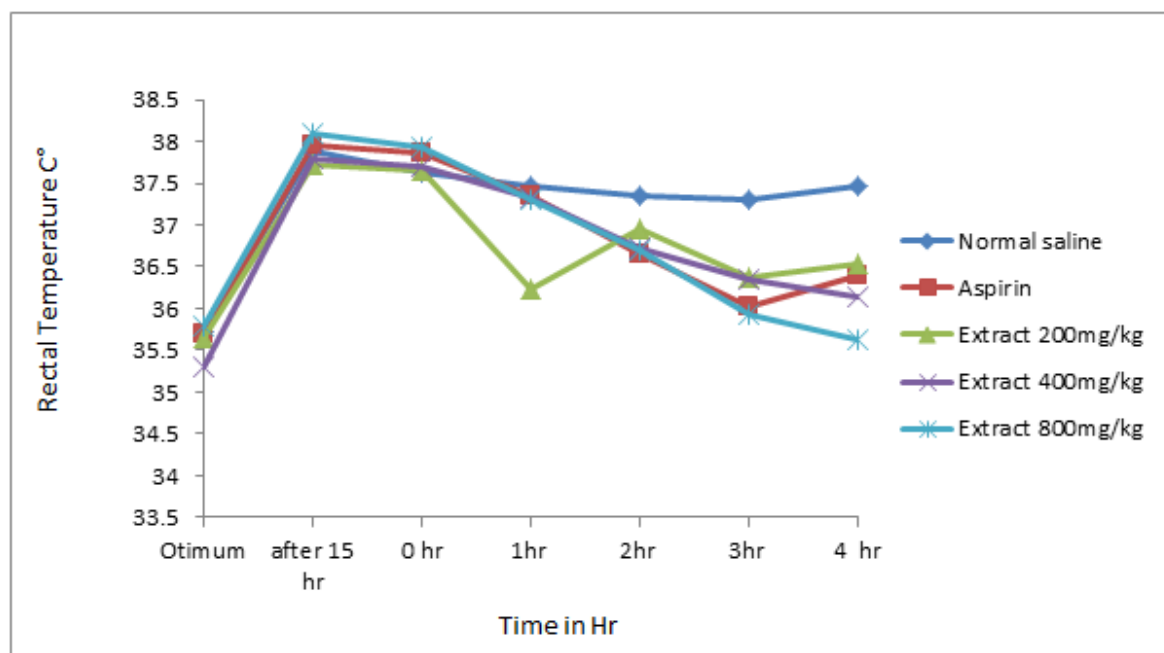


Figure: Effect of HEOM on yeast induced pyrexia. The data are expressed as mean ±SEM (N = 6), significant \*p< 0.05, \*\*p< 0.01 and \*\*\*p<0.001 when compared to normal control.



Seventeen hours after s/c injection of 20% of yeast; a significant increase in rectal temperature was observed in all animals. The effect of ethanolic extract on yeast induced pyrexia shows that rectal temperature was 38.10 C°, 15 hrs after the s/c injection of yeast suspension, further decrease to 37.31 C° within 1hr by the treatment of extract (800mg/kg), and subside after 4 hrs showing a sizeable reduction in rectal temperature and was comparable to reference drug Aspirin. The extract at a dose of 200mg/kg body weight showed reduction of pyrexia induced by yeast but it was not significant statistically  $p > 0.05$  upto 3 hrs and showed significant  $p < 0.001$  at 4hrs when compared to control. Treatments with ethanolic extract of *Portulaca tuberosa* at a dose of 400mg/kg and 800mg/kg body weight decrease rectal temperature significantly ( $p < 0.05$ ,  $p < 0.001$  respectively) at 2, 3 and 4 hrs after administration compared to normal control. Treatment with aspirin at a dose of 100mg/kg significantly  $p < 0.001$  reduced pyrexia induced by yeast at 3 hrs after administration. Treatment with ethanolic extract of *Portulaca tuberosa* at dose of 400 and 800mg/kg was nearly equally potent to reference compound Aspirin.

## 5. DISCUSSION

The aim of the present research was to validate the traditional uses of the extract of *Portulaca tuberosa* as antipyretic agent. The result of the present study showed that ethanolic extract exhibited significant effect against fever produced by s/c injection of yeast. The antipyretic activity produced by the ethanolic extract of *Portulaca tuberosa* is of considerable importance and justified its significance in fever as suggested in traditional medicines. Fever or pyrexia is a common medical sign associated primarily with abrupt increase in body temperature above normal and caused by certain illness related behavioral features like fatigue, depression, lethargy, anorexia, hyperalgesia (Hossain, E., S.C. Mandal, and J. Gupta, 2011). The elevation of body temperature occurs as a result of the release of certain chemical substances by immune system (Indumathy.S et. al. 2011). Infected or injured tissue enhances the formation of pro-inflammatory mediator i.e. cytokines like interleukin-1beta, alpha, beta and TNF- alpha which increase the synthesis of prostaglandin E2 (PGE2) and there by stimulating hypothalamus to raise body temperature (Flier, J.S, et. Al.1986).

Fever or pyrexia is a normal response against invading microorganisms to provide defense against infections (Tonks Fawcett and R. Watson 2003). Antipyretic

drugs inhibit COX-2 enzyme expression and thus inhibiting biosynthesis of PGE2 to reduce high body temperature (Cao, Y. and S.M. Prescott, 2002). Cyclooxygenase (COX) also known as prostaglandin (PG) H synthetase catalyzes the conversion of Arachidonic acid into prostaglandin H2 (Seibert, K Masferrer J, Zhang Y., 1995) The common therapy for management and control of fever NSAIDs. They are used for relief of inflammation, headache, anti arthritis pain, heart attacks and stroke. (Buffum, M. and Buffum J.C.2000). NSAIDs drugs inhibit prostaglandin and its derivatives produced through cyclooxygenase enzyme that cause inflammation, fever, pain and related diseases (Fung, H.B. and H.L. Kirschenbaum, 1999, Fung, H.B. and H.L. Kirschenbaum, 2012). However, NSAIDS produces a number of side effects like gastrointestinal bleeding, mucosal erosion, hepatotoxicity, renal toxicity and nephropathy (Greisman, L.A. and Mackowiak P.A., 2012). Meanwhile, in order to avoid from side effect, there are development and introduction of new antipyretic agents that compete with NSAIDs. The use of natural remedies for the treatment of inflammatory and painful condition has long history starting with Ayurvedic treatment, extends to the Europe. Plant drugs are known to play a vital role in the management of inflammatory diseases. Intraperitoneal administrations of the yeast to mice significantly increase rectal temperature and the tested drug significantly reduced rectal temperature. Thus it can be hypothesized that the extract contained pharmacologically compounds that interfere with the release of prostaglandin. The present results shows that extract possess significant as well as dose dependent antipyretic effect in yeast induced pyrexia which is comparable to that of standard drug. The effect of ethanolic extract on yeast induced pyrexia shows decrease in rectal temperature within 1 hr of the extract (800mg/kg) treatment, and at 4 hrs a sizeable reduction in rectal temperature which was comparable to reference drug aspirin. The tested drug at a dose of 400mg/kg and 800mg/kg body weight decreased rectal temperature significantly ( $p < 0.001$ ) respectively after administration as compared to negative control. Treatment of the tested drug at a dose of 400 and 800mg/kg produced a decrease in rectal temperature that was comparable to reference compound aspirin. The evaluated body temperature intensified the process of lipid per oxidation, which indicates that increase of oxidative stress causes pyrexia. The supplementation of antioxidant decreased the lipid per oxidation processes (Sehgal, Momekova D, Pencheva V, 2011).The flavonoids have antioxidant activity. Thus,

antioxidant activity of *Portulaca tuberosa* may be one of the possible mechanisms to reduce the elevated body temperature. The lowering of body temperature observed can be recognized to the presence of flavonoids in *Portulaca tuberosa* that might be responsible to lowered body temperature (Nwafor, P.A., et.al.2012).The extract may reduce PGE2 by its action on cyclooxygenase (Cox2) or by increasing the production of the body's own antipyretic substances like vasopressin and arginine (Saranya, R,et.al.2012).Various studies show that formulation containing tannins, alkaloids, flavonoids and carbohydrates has been reported for their antipyretic potential.

## 6. CONCLUSION

The results of the present study showed that plant extract significantly reduced elevated body temperature in dose dependent manners. These results validate the basis for the traditional use of *Portulaca tuberosa* against fever. The preliminarily analysis shows the presence of phenols, flavanoids, alkaloids, saponins, tannins and terpenoids. The pharmacological activity of the plant extract may be due to presence of these phytochemicals. However, further detail studies are necessary for isolation of pure secondary metabolites from the plant to understand the exact mechanism responsible for pharmacological activities of the plant.

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