



## Effect of phenolic compounds released during degradation of Coir pith by *Oscillatoria annae* on Albino rat (*Rattus norvegicus*)

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**ABSTRACT:** This study was carried out to investigate the effect of degraded coir pith based cyanobacterial culture filtrate on *Rattus norvegicus*. The culture filtrate containing phenolic compound was administered at a rate of 42 mg/Kg for 15 days and its effect on serum glucose, protein, creatinine, urea, uric acid, alkaline phosphate (ALP), serum glutamic oxaloacetic transaminase (SGOT), serum glutamic pyruvic transaminase (SGPT) and sperm analysis was studied. The results showed a significant reduction in the levels of serum glucose and liver marker enzymes while serum protein and renal function test showed normal level in both experimental and control rats. Also, a significant reduction in semen count was reported. The results suggested that the phenolic compound of the culture filtrate appears to be non toxic in the tested animals @ JASEM

Coir pith is a highly lignocellulosic waste dumped in huge piles on roadside, because of its high lignin content and slow degradation in natural environment it creates environmental pollution problems (Bhat and Narayan, 2003). The use of coir pith in agriculture has brought in favourable changes in soil drainage, mulching, rooting, soil conditioning and seed germinating aspects because of its high water holding capacity of 5-6 times its weight. Enhanced technique shows the use of coir pith based cyanobacterial biofertilizer in sustainable integrated agro ecosystem. Thus, they are used to promote plant growth and increase the quality and quantity of crop yield (Hume, 2007). Besides, various research on degradation of coir pith by fungi and bacteria were already reported, recent research shows that coir pith can be partially decomposed through the action of cyanobacteria and can be used as biofertilizer for all varieties of food crops (Anandharaj, 2007). The presence of phenolic compound in the coir pith based cyanobacterial culture filtrate was reported by Viswajith (2008). The enzyme profiles like laccase, polyphenol oxidase, peroxidase and esterase was also reported in *Oscillatoria annae* on exposure to lignocellulosic material *Lantana camara* (Viswajith and Malliga, 2008). Mohmoud et al (2000) proved that plant derived phenolic compound manifest many beneficial effect and can potentially inhibit several stages of APC (Adenomatous Polyposis Carcinogenesis) associated intestinal carcinogenesis in vivo. Also, it is investigated that the efficiency of several plant derived phenolics, including caffeic acid phenethyl ether (CAPE), curcumin, quercetin and rutin is proved for the prevention of tumor in CS7BL/6J-Min/+(Min/+) mice, which bear a germline mutation in the APC gene and spontaneously

develop numerous intestinal adenomas by 15 weeks of age. They also found that dietary level of 0.15% CAPE decreased tumor formation in Min/+ mice by 63%, curcumin induced a similar inhibition but quercetin and rutin failed to alter tumor formation at dietary level of 2% carcin. Lin and Chin (2007) reported phenolic compounds are widely present in the plant kingdom. Many epidemiological studies have indicated that consumption of some plant derived food stuffs with high phenolic content is associated with the prevention of some diseases and that these compounds may have similar properties like antioxidants, anti-mutagenic, anti-thrombotic, anti-inflammatory, anti-HIV-1 and anti-cancer agents. Since, the degraded coir pith with cyanobacteria was used as biofertilizer to improve crop production this study was carried out to analyze the effect of the coir pith based cyanobacterial culture filtrate on *Rattus norvegicus* to assess the safety limits of the intermediate compound on the animal model.

### MATERIALS AND METHODS

**Animal model:** Wistar rats (*Rattus norvegicus*) weighing 150-200 gram of either sex were employed in this study. The cages were kept in a well ventilated room with temperature of 28°C, 50-60% humidity. The animals were fed with standard pellet diets and water was provided with ad libitum. The animals were used for this experiment with the permission of University animal ethical committee, Bharathidasan University, Tiruchirapalli, Tamilnadu, India.

**Preparation of Extract:** Coir pith was allowed to degrade upto 30 days by *Oscillatoria annae* by inoculating in fresh BGII medium (Rippka et al.,

1979) of 1:10 (wet:dry) ratio. The experimental set up was incubated at 25° C under white fluorescent light of 1500 lux with 10/12hr light/dark cycle. After incubation period the supernatant was collected, dried in a polyvinyl tray under room temperature. 0.3% concentration of the dried compound was prepared, filtered using whatman no.1 filter paper and used as feed for the experimental animal. Preliminary qualitative analysis of the extract showed the presence of phenolic compounds.

**Experimental groups:** Twelve male and twelve female rats were tested for this study up to a period of 15 days. Six from each group were kept as control and the remaining set was used for experiment. The experimental rats were orally fed with 42mg/Kg of the test compound and the control rats received distilled water.

**Biochemical investigation:** After oral treatment, the animals were anaesthetised and the blood samples were collected by ocular puncture technique. The serum was separated and collected in a labelled eppendorf vial and stored at -20°C with sodium azide as preservative until use. General biochemical test like serum glucose (Carroll et al., 1971), protein (Weichselbaum, 1946) were quantified. Estimation of serum creatinine (Spencer, 1986), urea (Fawcett and Scott, 1960) and

uric acid (Barham and Trinder, 1972) was carried out to check the effect of phenolic compound on kidney function. The biochemical tests Alkaline phosphatase (ALP) (Bergmeyer and Brent, 1974), serum glutamic oxaloacetic transaminase (SGOT) and serum glutamic pyruvic transaminase (SGPT) (Reitman and Frankel, 1957) were done to examine the function of liver. Sperm count was made by using a hemocytometer counting chamber according to Zaneveld and Polakoski (1977).

## RESULTS AND DISCUSSION

Experimental results revealed that serum glucose level was decreased in 0.3% of coir pith based cyanobacterial culture filtrate when compared with that of untreated control rats. This may be due to hypoglycemic activity of the test compound. Supporting evidence showed the hypoglycemic activity of a polyphenolic oligomer-rich extract from the barks of *Cinnamomum parthenoxylon* (Jack) in normal, transiently hyperglycemic, and streptozotocin (STZ) induced diabetic rats. Oral administration of the extract at doses of 100, 200 and 300 mg/kg body wt. caused significant changes in body weight loss and fasting blood glucose levels of normal rats (Jia, 2009). No significant variation of serum protein between the control and test rats was found (Table-1) which indicates that the treated animals were normal.

**Table 1:** Effect of coir pith based cyanobacterial culture filtrate on biochemical test of white rat (*Rattus norvegicus*) (n=6)

Biochemical Test	Control Male	Experi-mental Male	Control Female	Experi-mental Female
Glucose (mgs %)	134.3 ±0.96	89.3* ±0.86	117.6 ±0.83	73.1* ±0.30
Protein (gms %)	5.8 ±0.07	6.1* ±0.07	6.5 ±0.06	6.4* ±0.15

Values are mean ± SE from six rats from each group; \* $p < 0.001$

**Table 2:** Effect of coir pith based cyanobacterial culture filtrate on renal function test of white rat (*Rattus norvegicus*) (n=6)

Renal Function Test	Control Male	Experi-mental Male	Control Female	Experi-mental Female
Creatinine (mgs %)	0.5 ±0.05	0.4** ±0.03	0.7 ±0.07	0.7** ±0.03
Urea (mgs %)	35.4 ±0.79	36.5* ±0.4	51 ±0.75	53.2* ±0.79
Uric acid (mgs %)	7.2 ±0.45	5.8* ±0.08	6.9 ±0.10	5.7* ±0.13

Values are mean ± SE from six rats from each group; \* $p < 0.001$ ; \*\* $p < 0.05$

The renal function test (Table-2) of the experimental rats does not showed any significant decrease or increase in the total creatinine content to that of normal rats. The urea content between the control and the test was found to be similar. However, significant reduction in serum uric acid was found in test rats when compared with control. This indicates that the animal's kidney was not affected. Similar report shows that milk extracts of

*Semecarpus anacardium* nuts on the effect of acute (72 hr) and sub acute (30 days) treatment of the drug with different dosage on liver and kidney functions and hematological parameters showed decrease in urinary urea, uric acid and creatinine levels during the sub acute toxicity studies (50, 100, 250 and 500 mg/kg body weight (Vijayalakshmi, 2000).

The level of ALP, SGOT, SGPT enzymes were found to be decreased in rats treated with coir pith based cyanobacterial culture filtrate when compared to control (Table-3). This result proves the protective effect of extracts on liver cells and this can be due to the presence of flavonoids compounds and their antioxidant effects (Sallie et al., 1991). It is observed that administration of phenolic extract on experimental rats showed decrease in sperm count (Table-4). This may be due to the direct effect of phenolic compound on pituitary gland secretory function causing to decrease in androgen. It has been demonstrated that

the process of spermiogenesis and the accessory reproductive organ function is androgen dependent (Khouri and El-Akawi, 2005). Thus lower dose administration of 42mg/Kg was found to be effective in interfering the sperm count. Orisakwe et al. (2003) reported the sub-chronic effect of *Hibiscus sabdariffa* calyx aqueous extracts on the rat testes and showed that significant decrease in epididymal sperm count in 4.6g/kg group compared to 1.15, 2.30 g/Kg of extract and control. From this it is concluded that our results are consistent with the above report in regard to decrease in sperm count.

**Table 3:** Effect of coir pith based cyanobacterial culture filtrate on liver function test of white rat (*Rattus norvegicus*) (n=6)

Enzymes	Control Male	Experi-mental Male	Control Female	Experi-mental Female
Alkaline phosphatase (KA/U)	36 ±0.56	23.6* ±0.60	34 ±0.61	24* ±0.87
Serum glutamic oxaloacetic transaminase SGOT(u/l)	99 ±0.38	56.6* ±0.60	80 ±2.7	61.5* ±0.64
Serum glutamic pyruvic transaminase SGPT(u/l)	75 ±0.84	54* ±0.90	68 ±0.73	50* ±0.79

Values are mean ± SE from six rats from each group; \*<sub>p < 0.001</sub>

**Table 4:** Effect of coir pith based cyanobacterial culture filtrate on sperm count of *Rattus norvegicus* (n=6)

Sperm count	Control Male	Experi-mental Male
Sperm count (million/cu.mm)	53 ±0.38	40* ±0.65

Values are mean ± SE from six rats from each group; \*<sub>p < 0.001</sub>

Thus the present study suggest that the phenolic compounds released during degradation of coir pith by *Oscillatoria annae* has no deliberative effect on tested animals when compared to control. However, the sperm count was found to be decreased but, toxicity was not reported in any cases. Therefore, further study is required to optimize the dosage level for a better utilization of the phenolic compound.

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