Protective Effect of *Coscinium fenestratum* Bark Extract on Letrozole Induced Polycystic Ovary Syndrome (PCOS) in Sprague Dawley Rats

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Authors’ contributions

This work was carried out in collaboration between both authors. Both authors contributed equally in establishing the topic of the research and design experiment. Both authors read and approved the final manuscript.

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ABSTRACT

**Aim:** Polycystic ovary syndrome (PCOS) is a common endocrine disorder. The aim of the present study is to evaluate the effects of the oral administration of *Coscinium fenestratum* bark extract in letrozole-induced PCOS in rats.

**Methods:** Thirty female Sprague-Dawley (SD) rats were randomly divided into five groups of 6 animals each. Letrozole (1 mg/kg) was administered orally for a period of 21 days to induce PCOS. Control rats received 1 mL/day of distilled water, and the three groups of PCOS rats were given 1mg/kg of clomiphene citrate, 250 mg/kg of *Coscinium fenestratum* extract and 500 mg/kg of *Coscinium fenestratum* extract by oral gavage for 15 days. Blood samples were collected and the ovary of rats was removed, weighed, and fixed in 10% buffered formalin to determine the hormonal and antioxidant parameters.
Results: In rats treated with BECF, significant increase in Catalase, Superoxide Dismutase and Lipid peroxidation level was decreased when compared with letrozole group. Histopathology showed that the BECF treated rats had normal ovarian structure, which was seen distorted due to the PCOS condition in letrozole treated rats. The results show significant decrease in total testosterone level and LH and significant increase in FSH. Therefore, the extract of BECF show significant effect on PCOS rats. The significance p value is >0.001.
Conclusion: Based on these result, the BECF extract and traditional wisdom of this plant when used regularly can be a good treatment a suitable treatment option for PCOS which can be regularly used.

Keywords: Polycystic ovary syndrome; Coscinium fenestratum; clomiphene citrate; luteinizing hormone; follicle stimulating hormone.

ABBREVIATION

BECF: Bark Extract of Coscinium fenestratum

1. INTRODUCTION

Polycystic ovary syndrome (PCOS) is referred as hyper-androgenic and common endocrine disorder in women reproductive age [1]. The complex condition is characterized by elevated androgen levels [2]. The manifestation of PCOS affects women of child bearing age (from 18-30) because the ovaries develop into numerous small collection follicles and failed to regulate the release of dominant follicle (chronic anovulation). Prevalence of PCOS in India ranges from 3.7 to 22.5 % depending on the population studied and the criteria used for diagnosis [3]. In PCOS, high level of LH secretion contributes to high level of androgen and low level FSH and it may lead to poor egg development, inability to ovulate and also lead to deficiency of progesterone production and often leads to absence of menstrual cycle period(ref). The characteristics of PCOS is overweight [4], insulin resistance [5], anovulation, irregular periods, infertility, hypoandrogenemia and inflammation [6]. The symptoms of PCOS are irregular periods, acne, hirsutism, weight gain, alopecia and heavy bleeding [7]. The hormone is involved in PCOS is follicle stimulating hormone (FSH) and luteinizing hormone (LH) which is responsible for reproduction by testis in male and ovarian ovaries in female(ref). FSH is released by stimulation of GnRH secretion from the hypothalamic pituitary hormone from the anterior pituitary following the stimulation by GnRH from hypothalamus (hypothalamic-pituitary –ovarian axis), it regulates development, growth, pubertal maturation and reproduction. Pubertal maturation, and secondary sexual characteristics development. In female – FSH also initiates follicular growth, development of oocytes and, yolk deposition and increase the production of growth factors inside the ovary [8]. LH is a hormone produced by gonadotropic cells from the anterior pituitary gland. In female LH is responsible for ovulation and progesterone release after ovulation from the corpus luteum [9].

Obesity is the major key component for metabolic syndrome and associated with level of insulin which is increased in ovarian androgen production. The excessive adipose tissue is responsible for ovulatory dysfunction and menstrual abnormalities [10]. Obesity and insulin resistance increase the risk of type 2 diabetes and cardiovascular disease [11]. The decrease in HDL-C and increase in triglycerides and LDL can lead to PCOS[12].

The phenotype limits of PCOS is defined by specific phenotype which determine the long term morbidity [13]. The factors influencing the PCOS are genetic factors, environmental factors and endocrine factors.

The Rotterdam consensus include the diagnostic criteria national institute of Health (NIH) proposed set any two from the three must present to diagnosis PCOS: Hyperandrogenism, ovulatory dysfunction and polycystic ovaries by transvaginal ultrasound.[14].

Tree turmeric is a beneficial plant in the world were its root and stem serve as excellent antibiotic and antiseptic properties, which belongs to menispermaceae family and botanically known as Coscinium fenestratum. Berberin is the active ingredient that gives the medicinal properties. The major alkaloids are yellow crystalline berberine, protoberberine and jatrorrhizine [15]. The Coscinium fenestratum
mainly from South Asia and parts of South-East Asia [16].

The plant is used for treating inflammation, stimulate wounds and ulcers healing, jaundice, burns, skin disease, abdominal disorders and diabetes. The bark is used in gynaecological troubles [17]. The Coscinium fenestratum have activities like anti-diabetic [18], anti-gonococcal [19], anti-oxidant and anti-microbial [20], anti-hepatotoxic [21], immunomodulatory [22], anti-proliferative [23], anti-acne [24], hypotensive [25], CNS depressant and analgesic activity [26].

2. MATERIALS AND METHODS

The bark of Coscinium fenestratum were collected from local source, Tamil Nadu. The bark material was identified and authenticated by Dr. P Jayaraman, Plant Anatomy Research Centre, Tamil Nadu. Collected bark was ground into coarse powder. The powered bark was extracted with ethanol: water (3:1) in Soxhlet’s apparatus. 630g powder of Coscinium fenestratum is extracted with 1500ml at 80°C for 72h. The extract is filtered and vacuum dried. The dried residue of extract was cooled in desiccator for 30 min and then accurately weighed the % yield was 3.23 [27].

The female SD rats 3 weeks old was chosen which was procured form mass biotech. The animals were acclimated to laboratory conditions one week and provided with standard pellet chow and water ad libitum and maintained in animal house of C.L. Baid Metha College of Pharmacy. Ethical committee clearance was obtained from IAEC with IAEC REFERENCE NO: 02/321/PO/Re/S/01/CPSCEA – 17/11/2021 The animals were divided into 6 groups (n=6), Group I - control group, Group II – letrozole (1 mg/kg, P.O), Group III – Clomiphene citrate (1mg/kg, P.O.), Group IV – Low dose (250 mg/kg, P.O), Group V – High dose (500 mg/kg, P.O). The duration of study is for 36 days, letrozole was administrated orally through oral gavage for 21 days induction PCOS, followed by oral administration of BECF for 15 days. For confirmation of PCOS formation the estrus cycle was done. On the 36 days, the animals are anaesthetized and decapitated, ovaries and serum were collected. Histopathology, antioxidant and hormonal assay was performed.

3. RESULTS AND DISCUSSION

![FOLLICLE STIMULATING HORMONE](image1)

**Fig. 1.**

**GROUPING**

**FSH (ng/ml)**

**Group I**

**Group II**

**Group III**

**Group IV**

**Group V**

![LUTENIZING HORMONE](image2)

**Fig. 2.**

**GROUPING**

**LH (ng/ml)**

**Group I**

**Group II**

**Group III**

**Group IV**

**Group V**

![TOTAL TESTOSTERONE](image3)

**Fig. 3.**

**GROUPING**

**TT (ng/ml)**

**Group I**

**Group II**

**Group III**

**Group IV**

**Group V**

![SUPEROXIDE DISMUTASE](image4)

**Fig. 4.**

**GROUPING**

**SOD (U/L)**

**Group I**

**Group II**

**Group III**

**Group IV**

**Group V**
3.1 Hormonal Analysis

FSH, LH and Testosterone level was estimated using chemiluminescent method, the result shows the FSH level increased and Testosterone and LH level decreased when compared with group II as shown Figs. 1, 2, 3.

3.2 Anti-oxidant

Superoxide dismutase, Catalase and Lipid peroxidation was estimated using specific kits, the result shows Superoxide dismutase and Catalase level increased and Lipid peroxidation level decreased when compared with Group 4,5,6.

3.3 Histopathology

Group I – Healthy growing follicles and a corpus luteum is seen
Group II- Several cyst-like follicles were formed
Group III – Decrease in cyst size and generation of healthy follicles
Group IV – BECF (250mg/kg) show slight decreases in cysts size
Group V – BECF (500 mg/kg) shows decrease cysts size and regeneration of healthy follicles
3.4 Statistical Analysis

The data was analysed in terms of mean ± standard error of mean (SEM). For statistical analysis, multiple comparisons of data were made using Analysis of variance (ANOVA) followed by Dunnett's multiple comparison test. Software program GraphPad prism 9.3.1 was used for all data analysis.

4. CONCLUSION

Animals who were treated by BECF extract exhibited a regular pattern estrus cycle. The BECF treated rats were showed a marked increase in FSH level, while both testosterone and LH levels were markedly decreased. BECF extract also had some anti-androgenic effects represented by degeneration of immature follicles and decrease in cysts; size. Thus, this plant can help in restoration of normal estrogenic state, inhibiting hyper androgenic state and treating PCOS.

The Estrus cycle has been restored to regular pattern in the animals treated with BECF. Hence is concluded that this extract has produced potential healing effect on PCOS rat model, restoring the reproductive cycle and other complications to normal. The BECF treated group causes an increase in FSH level and decrease in LH and Testosterone level and BECF shows a remarkable antiandrogenic effect by degenerating the immature follicular cysts and decreasing the cysts size which can help in regulating normal ovulation. So this plant helps in restoring the estrogenic conditions and inhibit aspects of hyperandrogenism.

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CONSENT

It is not applicable.

ETHICAL APPROVAL

Animal Ethic committee approval has been collected and preserved by the author(s).

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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