INTERNATIONAL JOURNAL OF CURRENT RESEARCH IN CHEMISTRY AND PHARMACEUTICAL SCIENCES
(p-ISSN: 2348-5213: e-ISSN: 2348-5221)
www.ijcrcps.com
DOI: 10.22192/ijcrcps Coden: IJCROO(USA) Volume 5, Issue 6 - 2018

**Research Article**


**DOI:** http://dx.doi.org/10.22192/ijcrcps.2018.05.06.009

# Ovulation inducing and follicular genetic activity of Patchai Karpoora Melugu in virgin female wister rat

**Priya. M[1], Priya. S[2], Kingsly. A[3]**
[1]PG – Scholar, Department of Gunapadam, GSMC, Palayamkottai, Tirunelveli
[2]PG - Scholar, Department of Noinadal, GSMC, Palayamkottai, Tirunelveli.
[3]HOD-Department of Gunapadam, GSMC, Palayamkottai, Tirunelveli
Corresponding Author: **DR.M.PRIYA**, PG Scholar, Department of Noinadal.
E- mail: s.priya.dr@gmail.com

**Abstract**

PCOS is a major health issue in women at fertile age group all over the world. It is an endocrine disorder, and one of the major cause of infertility. PCOS accounts for about 75% of female infertility. In our Siddha system of medicine, many gynaecological disorders were quoted, among that the symptoms of Soothaga vaayu may be correlated with symptoms of PCOS.In Modern system, the treatment regimen of PCOS, includes oral hormonal pills and invasive procedures like Lap. ovarian drilling, ovarian cystectomy, that can trigger the ovulation. In this study, I establish the scientific validation of Patchai Karpoora Melugu(PKM) towards ovulation inducing and follicular genetic activity in virgin female wister rats.

**Keywords:** Patchai Karpoora Melugu (PKM), PCOS, Ovulation inducing and follicular genetic activity.

## Introduction

Polycystic Ovary Syndrome (PCOS) is a common endocrine system disorder among women of reproductive age. Women with PCOS may have enlarged ovaries that contains small collections of fluid-called follicles-located in each ovary.It has symotoms includes hirsutism and acne (Predominant androgenisation), obesity and infertility in case of bilateral PCOS.

High LH concentration is reported to be the second cause of female infertility since it interfere with normal ovarian cycle and is mainly associated with reduced rate of pregnancy and increased miscarriage rates.

The most frequently prescribed drug for anovulation is Clomiphene citrate. Eventhough this is the drug of choice, many adverse effects are reported. Among them hot flushes, ovarian enlargement, multiple cyst formation due to hyper stimulations of ovaries which may result in rupture and internal haemorrhage and multiple pregnancies, resulting from multiple ovulation are common.

In present days, there is a need of highly therapeutic remedy for PCOS with no or less adverse effect. Our traditional Siddha system has such enormous medicine with less adverse effect. From that Patchai Karpoora Melugu is taken from Anuboga Vaithiya Navaneetham for PCOS. In this study, the effect of the drug in Ovulation inducing and follicular genetic activity can be investigated.

## Materials and Methods

Patchai Karpoora Melugu is a herbo-mineral drug which is quoted in Anuboga Vaithiya Navaneetham indicated with ammenorrhoea, PCOS, Menorrhagia, Post eclampsia, Retention of placenta. The ingredients of PKM is Bornea camphor, Calomel, Red Sulphide of mercury, Cinnabar, Crocus sativus, Honey. The ingredients of drug was purified as per siddha regulations and the drug was prepared.

### Ovulation inducing and follicular genetic activity:

To carry out ovulation inducing activity, twenty four Virgin female wister rats weighing about 95-130 gm of 2 month old were randomly selected from the animal house at Vel's University, Chennai. Before entering into the ovulation inducing study, synchronization of the reproductive cycles was done in the experimental rats by the following method . 100µg estradiol was taken and dissolved in 2 ml olive oil which was administered subcutaneously. After the period of 24 hours, all the animals are administered intramuscular injections of 50 µg progesterone dissolved in olive oil. Few hours later, vaginal smears of all the rats were collected by vaginal lavage for the analyzation of ovulation and oestrous cycle . Vaginal smears were prepared by washing vaginal opening with 0.9% w/v of sodium chloride with a glass dropper and placed in a clean glass slide and viewed under light microscope at 40X magnification. Assessment of vaginal smears confirmed that all the animals were in the estrous stage.

Then the rats were grouped into four of six each.

Group I – Considered as normal Control given 2ml/kg of CMC solution only for 10 days.

Group II – Considered as *PKM* – 100 drug treated group which were administered 100 mg/kg of *Patchai Karpoora Melugu* for 10days,

Group III – Considered as *PKM*- 200 drug treated group and were received 200mg/kg of *Patchai Karpoora Melugu* for 10 days

Group IV- Served as a standard group received Clomiphene citrate 10mg/kg for 10 days. All the drugs were given orally. Body weight of all the animals were weighed daily after drug administration for 10 days.

At the end of experiment, 2ml of blood was collected from all the animals by retro orbital puncture. Blood samples were centrifuged at 4000 rpm for 15 minutes and the serum was separated. This samples were frozen at -20°C and LH, FSH, estradiol, progesterone and testosterone level were estimated by ELISA method.

### Histological analysis:

The ovary was separated from the uterus and placed in formalin fixative for 20-24 hours. Then these fixed tissue samples were placed in ascending concentrations of alcohol and embedded in paraffin. Tissues were sliced with 5-7 µm thickness and stained with haematoxylin and eosin, and then monitored and analyzed with a light microscope. For the evaluation of folliculogenesis activity of trial drug, all tissue blocks were serially sliced. Follicle identification was based on the detection of a nucleus. The numbers of follicles (primordial, primary, etc.) were counted. Follicle recognition criterion on the slides was based on the type of epithelial cells surrounding them. For example, primordial follicles have squamous cells whereas primary follicles are surrounded by cuboidal cells. The numbers of follicles per slide were randomly counted.

## Results

The effect of the administration of trial drug *Patchai Karpoora Melugu* and standard drug Clomiphene citrate on serum concentration of reproductive hormones are presented in the given table below and Graph No.1, Graph No.2, LH, FSH, Estrogen, Progesterone and Testosterone were analyzed. The result of Table No.26 showed that the administration of *Patchai Karpoora Melugu* in the dose of 100 mg and *200* mg caused no significant effect on LH which was nearly similar to the normal and standard group. There was significant increase in FSH level ($p<0.05$) in standard drug Clomiphene citrate. Animals are pre treated with *Patchai Karpoora Melugu* 50 mg showed the significantincreased the level of FSH ($p<0.05$) in dose - dependent manner. *Patchai Karpoora Melugu* in the dose of 100 mg was also an increase in the level of FSH though not statistically significant.

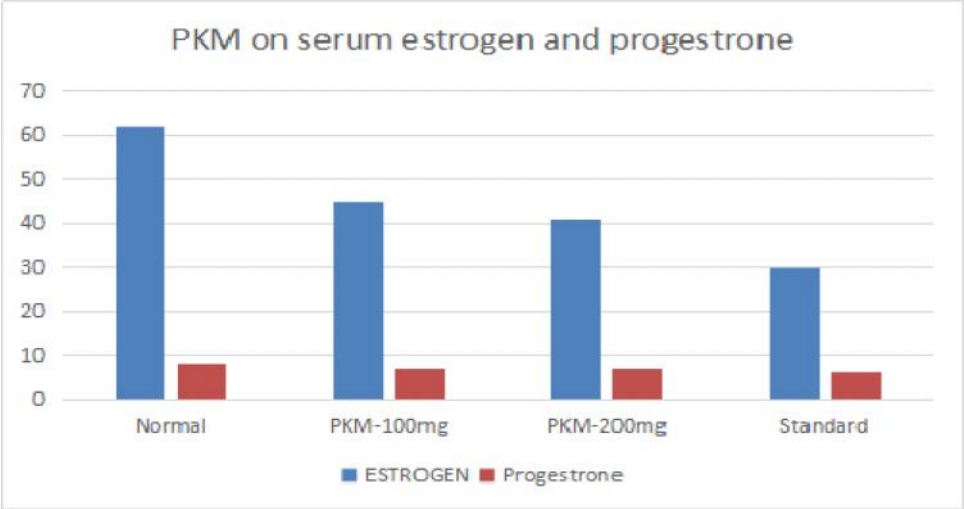
In accordance with the results related to estrogen and testosterone also showed a significant decrease after the administration of *Patchai Karpoora Melugu* when compared withstandard and normal control group. Both trial drug treated groups and standard group produce little decreasing effect on progesterone level which was insignificant.

### Histopathological study of ovary tissue:

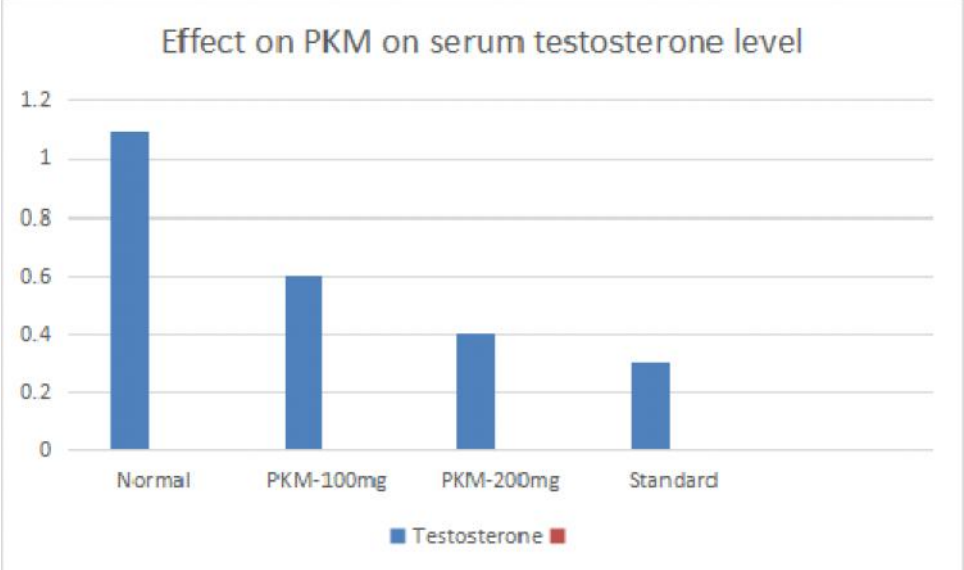
Histological studies of ovarian tissues of normal group, standard group, PKM – 100 and PKM -200 trial drug treated groups were presented in respectively. Theovarian tissue of the normal group showed the normal histological features with presence of few primordial follicles, matured graffian follicle. . Standard group and both doses of *Patchai Karpoora Melugu ( PKM –100* mg and *PKM* -200 mg)showed some well defined histological features with increased number of primary follicles, matured graffian follicles and also corpus luteum when compared with normal rat. These were more pronounced in rat ovary that received *PKM* – 100 mg and Clomiphene citrate.

**Effect of *Patchai Karpoora Melugu* on serum concentration of reproductive hormones of female rats after 10 days treatment**

| Group | Treatment and dose | LH (IU/ml) | FSH (IU/ml) | Estrogen (pg/ml) | Progesterone (pg/ml) | Testosterone (ng/ml) |
|---|---|---|---|---|---|---|
| Normal | 2ml/kg 2% CMC | 0.31±0.06 | 0.36±0.04 | 62.24±3.2 | 8.2±1.12 | 1.1±0.10 |
| Test-I | *PKM*-100mg/kg | 0.39±0.08 | 0.50±0.06 | 45.27±2.2 | 7.1±1.00 | 0.6±0.05 |
| Test-II | *PKM*- 200mg/kg | 0.48±0.08 | 0.61±0.08 | 41.55±1.4 | 6.8±0.82 | 0.4±0.03 |
| Standard | Clomiphene 10mg/kg | 0.56±0.14 | 0.67±0.10 | 30.62±1.0 | 6.2±0.61 | 0.3±0.02 |




**Graph no. 1**




**Graph no. 2**

## Discussion

Test drug *Patchai Karpoora Melugu* 100mg and *Patchai Karpoora Melugu* 200mg decrease serum testosterone level significantly ($p<0.01$), which is one of the requirement to manage PCOS as the serum concentration of testosterone is the primary reason for hyperandrogenism (hirsutism, hair loss appearing as thinning hair on the top of the head, acne, oily skin, obesity, depression and deepening of voice.) in PCOS.

*Patchai Karpoora Melugu* 100 mg significantly increases the serum FSH in animal model ($p< 0.05$) which is primary step for inducing ovulation. The early stages of follicular growth are primarily driven by intraovarian factors, whereas maturation to the state required for ovulation, including the resumption of meiosis in the oocyte, requires the combined stimulus of FSH and Luteinizing hormone (LH). Gonadotropic hormones especially the FSH causes accelerated growth of 6 to 12 primary follicle each month. The early growth of the primary follicle up to the antral stage is stimulated mainly by FSH alone. After a week or more of growth before ovulation occurs, one of the follicles begins to outgrow all the others; The reminder begin to involute (process called atresia) and these follicle are said to become atretic. This process of atresia is important one, because it allows only one of the follicle to grow large enough to ovulate. Different dosages of the *Patchai Karpoora Melugu* slightly increased the number of atretic follicles; a greater increase was observed at 200 mg/kg.

Treatment with doses of 100 and 200 mg/kg of *Patchai Karpoora Melugu* significantly increased thenumber of primary follicles, however it was statistically significant only in the 50mg/kg group.

The treatment with *Patchai Karpoora Melugu* caused an alteration in the amount of FSH, which was statistically significant. *Patchai Karpoora Melugu* in the first stage of ovarian folliculogenesis strongly stimulates the maturation of primordial follicles. This effect was more pronounced at the 200mg/kg dose of the *Patchai Karpoora Melugu*, in which it acted as a stimulant, causing progres-sion of ovarian folliculogenesis to the stage of primary follicle formation. However, at the next stage, *Patchai Karpoora Melugu* caused an increase in the number of growing follicles. The *Patchai Karpoora Melugu* also caused an increase inthe number of atretic follicles, which confirmed the repressing effect of the *Patchai Karpoora Melugu* on the natural growth of follicles

## Conclusion

The results of ovulation effect of *Patchai Karpoora Melugu* revealed the significant influence at the dose level of 200mg/kg and this marked effect was ensured with the histological evaluation of ovary of experimental rats also. Hence it may be concluded that the *Patchai Karpoora Melugu* is an excellent traditional medicine in the treatment for anovulatory conditions like PCOS and the effect may be attributed to the elevation of the ovulation stimulatory hormones in animal models.

## Acknowledgments

The authors wish to thank The Vice Chancellor, The Dr.MGR Medical University, Guindy, Chennai and to Indian Medicine and Homoeopathy Department, Arumbakkam, Chennai and special thanks to the Principal, Govt.Siddha Medical College, Palayamkottai. Dr. G. Esakki Pandian, $MD_{(S)}$, Lecturer, GSMC, Dr. Antony Duraichi, $MD_{(S)}$, Assistant Lecturer, GSMC, Palayamkottai. Dr. P. Madhusamy, PG Scholar, GSMC, Palayamkottai.

## References

1. Anuboga vaidhya navaneetham 7th part,page no-56,57
2. Anna Glasier, Metin Gülmezoglu, George P Schmid, Claudia Garcia Moreno, Paul FA Van Look. Sexual and reproductive health: a matter of life and death. The Lancet Sexual and Reproductive Health Series, October 2006
3. World Health Organization, Sexual and Reproductive Health 1, lancet_1.Geneva, (2006).
4. Douglas T.Carrell, Catherine Racowsky, Peter N.Schlegel, Bart C.Fauser; Biennial Review ofinfertility, Volume 1, Springer Dordrecht Heidelberg London New York; pp. 23, (2009)
5. Gautam N Allahbadia, Rina Agarwal, Polycystic ovary syndrome, Anshan Ltd, pp.15, (2007)
6. Gabor Kovacs and Robert Norman, Polycystic ovary syndrome, 2nd edition, Cambridge University Press; pp.68, (2007)
7. Zaidi J, Campbell S, Pittrof R et al, Ovarian Stromal blood flow in women with polycystic ovaries; a possible marker for diagnosis. Human Reproduction, 10; 199-6, (1995)
8. R.S.Satoskar, Nirmala N. Rege, S.D.Bhandarkar Pharmacology and Pharmacotherapeutics, 22nd edition, Popular prakasham private Ltd; pp. 964,(2011)
9. Hakkeem Mohamed Abdullah Sahib “Anubhoga Vaidya Navaneetham, Part III Edition II, Tamarai press, pp.66, (1975)
10. Thiyagarajan R. Gunapadam Thathu – Jeeva Vaguppu Part (2 & 3) Edn 2, Indian Medicine and Homeopathy Dept. Chennai, pp.408, (2006)

11. Organization for Economic Cooperation Development (OECD) Guideline, 425, Guideline Document on Acute Oral Toxicity. Environmental Health and Safety Monograph Series on Testing and Assessment No. 24, (2000)
12. Aarthi V et al, Effect of Ehretia microphylla lamk on stimulation of reproductive function and ovarian folliculogenesis in rats, Int J Pharm Bio Sci 2012 July; 3(3): (P) 273 – 280.
13. Richards JS, Perspective: the ovarian follicle-a perspective in 2001. Endocrinology, 142(6): 2184-2193, (2001)
14. Hirshfield AN, Development of follicles in the mammalian ovary. Int Rev Cytol, 124: 43-101, (1991)
15. Marcondes FK, Bianchi FJ and Tanno AP, Determination of estrous cycle phases of rats: some helpful considerations. Braz J Biol, 62 (4), 609-614, (2002)
16. Catherine A. Picut et al, Ovarian Follicle Counts Using Proliferating Cell Nuclear Antigen (PCNA) and SemiAutomated Image Analysis in Rats. Toxicologic Pathology, 36: 674-679, (2008).
17. J. Jameson, Stephen Hauser, Anthony Fauci, Dennis Kasper, Joseph Loscalzo, Dan Longo. Harrison's Principles of Internal Medicine Ed.18, Tata Mcgraw-Hill Education; pp.3029, (2012)
18. C.Guyton, John E. Hall, Textbook of Medical Physiology; 12th edition, Elsevier Health Science pp.1037, (2011)

**Access this Article in Online**

Website: www.ijcrcps.com

Subject: Siddha Medicine

Quick Response Code

DOI: 10.22192/ijcrcps.2018.05.06.009

How to cite this article:
Priya.M, Priya.S, Kingsly.A. (2018). Ovulation inducing and follicular genetic activity of Patchai Karpoora Melugu in virgin female wister rat. Int. J. Curr. Res. Chem. Pharm. Sci. 5(6): 43-47.
DOI: http://dx.doi.org/10.22192/ijcrcps.2018.05.06.009