How to Cite:
Bawane, V. (2022). Two arm open labelled randomized standard controlled prospective clinical study to assess the efficacy of Ashwagandha Kshirpaka for Folliculogenesis in management of Vandhyatva. *International Journal of Health Sciences*, *6*(S2), 12458–12466. https://doi.org/10.53730/ijhs.v6nS2.8788

Two arm open labelled randomized standard controlled prospective clinical study to assess the efficacy of Ashwagandha Kshirpaka for Folliculogenesis in management of Vandhyatva

Dr.Vishnu Bawane
Ph.D.Scholar, Department of *Prasutitantra-Striroga*, School of Ayurveda D.Y. Patil deemed to be University, Nerul, Navi Mumbai.
Corresponding author email: drvcbawane@gmail.com

***Abstract*---**Background: Anovulation or inability to produce fertile ovum is an important cause among the women for infertility. One of the formulations given in *Bhavaprakasha* under *Yonirogadhikara*, *Ashwagandha Kshirpaka,* can be effective in Folliculogenesis in females. Objectives: The present study was conducted to compare the efficacy of *Ashwagandha kshirpaka* & Clomiphene citrate on Folliculogenesis in the management of *Vandhyatva*. Methods: In present two arm, open labeled randomized standard controlled prospective clinical study, 60 patients suffering with *Vandhyatva* with anovulatory cycles fulfilling the inclusion and exclusion criteria were selected by purposive sampling technique from the study centre and randomized into two groups. 30 patients in Trial Group received *Ashwagandha kshirpaka* whereas 30 patients in Control Group 30 patients received Clomiphene citrate. All the patients were subjected to haematological and hormonal profile assay and follicular studies on three consecutive menstrual cycles. Assessment of follicle size & endometrial thickness were done with USG Follicular Study whereas interval between two menstrual cycles & menstrual regulations were assessed and documented in CRF. Results: The follicular size, endometrial thickness and interval between two MCs improved in patients who received *Ashwagandha Kshirpaka* and Clomiphene citrate treatment i. e. Trial group and Control group which was statistically significant too. Inter-Group comparison showed that the difference in objective as well as subjective parameters in trial and control groups were statistically insignificant, indicating that *Ashwagandha Kshirpaka* is comparable to Clomiphene citrate in folliculogenesis. *Ashwagandha Kshirpaka* was also found to be safe. Conclusion: *Ashwagandha Kshirapaka* and Clomiphene citrate were found to be equally efficacious in Folliculogenesis. *Ashwagandha*

kshirpaka was concluded as an effective remedy for *Samprapti Bhanga* of *Abijotsarga* i.e anovulation.

***Keywords*---**Ashwagandha Kshirapaka Clomiphene citrate, anovulation, Vandhyatva.

Introduction

Reproduction is the process that requires the interaction and the integrity of the female and male reproductive tracts. As successful pregnancy is a multi-step chain of events, even if one of the events or conditions is not met in right amount of time, pregnancy may not happen or reach successful completion until normal birth of a baby. According various research journals, ovarian factors contribute to 25-30% causes of the female infertility. So, it is the second common cause of infertility. Ovulatory cause is an important subset in infertility among women, accounting about 40% of cases. Anovulation or inability to produce fertile ovum is an important cause among the women for infertility. [1-3]

Infertility treatment has taken considerable lead in today's world of science and technology. Though physicians are able to give hope to many patients with ART, but still there is no surety of the success. Many therapies have been developed, but they have unsatisfactory results, are expensive and have lots of side effects like ovarian hyper-stimulation, frequent abortions, multiple gestations and possibility of ovarian cancer. [4] Hence, the infertility especially due to ovarian factor needs an immediate attention from complementary and alternative medicines. Ayurveda may give a promising hand to cure this disease even though there is no specific treatment according to factors. The ancient system of Ayurvedic medicine advocated variety of natural medicines, which may provide good results on this factor without any harmful effect.

To achieve conception, *Acharya Sushruta* has described four essential factors for fertility, viz. *Rutu*, *Kshetra*, *Ambu* and *Beeja.* Among them, *Beeja* is the cornerstone of the female reproductive process and in its absence *Garbha* (foetus) cannot be formed in spite of all the other factors. [5] Vitiated *Vata Dosha* causes *Yoni Dushti* which leads to *Vandhyatva.* [6] *Vata Dosha* is the governing factor of whole reproductive system. So, ovulation is also governed by *Vata dosha.* One of the formulations given in *Bhavaprakasha* in *Yonirogadhikara*, *Ashwagandha Kshirpaka* has *Ashwagandha* as its main content which has *Vata-kaphaghna* property. *Godugdha* and *Goghruta* are also its contents effective on aggravated *Vata* and *Pitta dosha.* [7] Clomiphene citrate (clomiphene), is the most commonly used drug for ovulation induction, since it is inexpensive, highly effective and user-friendly. Keeping all these aspects in mind, the clinical study entitled "Two Arm Open Labelled Randomized Standard Controlled Prospective Clinical Study to Assess the Efficacy of *Ashwagandha Kshirpaka* for Folliculogenesis in Management of *Vandhyatva*" was undertaken.

Materials & Methods

Ethical considerations

The study was carried out in accordance with the principles of Good Clinical Practice (GCP), Declaration of Helsinki (Brazil update 2013), ICMR (Indian Council of Medical Research) and local regulatory authorities. The study protocol was approved by the Institutional Ethics Committee of study site and registered in the Clinical Trial Registry – India. The study was conducted at dept. of Prasutitantra and Streerog, D. Y. Patil deemed to be University School of Ayurveda-Navi Mumbai, Maharashtra.

Study design

It was a two arm, open labeled, randomized, standard controlled, prospective clinical study.

Enrolment of patients

Patients suffering with *Vandhyatva* with anovulatory cycles irrespective of occupation, religion & *Prakruti*, socio-economic status, type of infertility fulfilling the inclusion and exclusion criteria were selected from O.P.D and I.P.D. of D.Y. Patil School of Ayurveda Hospital, Navi Mumbai.

Inclusion criteria

Female patients in the Age group of 21 - 40 years having regular or irregular menstrual history or diagnosed with polycystic ovarian syndrome (PCOS), who were ready to provide written informed consent and who were ready to willingly participate and follow the protocol requirements of the clinical study were included in the study.

Exclusion criteria

Females with Congenital absence or deformities of Uterus & ovaries, ovarian tumour, malignancy of Uterus, Cervix or ovary, also patients with Thyroid disorders, DM, TB, HIV, HBsAg positive were excluded from the study. subjects participating in any other clinical study or having participated in any other study three months prior to screening in the present study, subjects having a past history of allergy to any drugs or other conditions, which in the opinion of the investigator made the patient unsuitable for enrolment or could interfere in adherence to the study protocol were excluded from the study.

Laboratory Investigations

All the patients were subjected to Complete Blood Count (CBC), ESR, Fasting and Post-Prandial Blood Sugar Levels, routine and microscopic urine examination, HIV, VDRL, HbA1C and follicular studies on three consecutive menstrual cycles.

Sample size

The primary population for this study was per protocol population. A minimum statistically relevant sample size of 60 completers (30 each in the two groups) was considered for the present study**.** Total 60 patients of *Vandhyatva* with anovulatory cycles have been selected by purposive sampling technique & screened after informed valid written consent for the research work. Those 60 patients were enrolled and divided in two groups.

Trial Group - 30 patients received *Ashwagandha kshirpaka* (100ml *kshirpaka* was given orally from 5th day of menses for 12 days before breakfast)
Control Group - 30 patients received Clomiphene citrate (50mg Tab was given from 3rd day of menses for 5 days once in a day)
Before starting treatment, patients were observed for anovulatory cycle. Status of the patient was recorded as well as investigations were done. Treatment was given for 3 consecutive menstrual cycles. Follow-up was taken in each cycle and changes were recorded on CRF. Assessment of Follicle Size & Endometrial thickness done with USG Follicular Study (Objective Parameters). Interval between two menstrual cycles & menstrual regulations (Subjective Parameters) were assessed and documented in CRF.

Study duration & Visits

The total duration of the study treatment was 90 days. The study visits were planned & outcomes were assessed on Screening Visit, Baseline Visit (Day 0), Day Day 30, Day 60 and Day 90 i. e. end of the study.

Assessment of Efficacy Parameters

Objective parameters

Sr. No.	Parameter	Score/Grades
1	Follicle Size	0 = <10mm 1 = >10mm-14mm 2 = >14mm-18mm 3 = >18mm
2	Endometrial Thickness	0 = <5 mm 1 = > 5 to <10mm 2 = > 11 to <14 mm 3 = >14
3	Interval Between Two Menstrual Cycles	0 = 25-35 Days 1 = 35-40 Days 2 = 40-45 Days 3 = >45 Days

Table no. 1: Objective assessment parameters

Assessment of Safety

Safety of the study drugs was assessed by evaluating adverse events, overall safety and tolerability of the product by the physician and subject on global

assessment scale. Any clinically significant changes in laboratory parameters were also reported.

Plan for statistical analysis

The study data generated and collected was put to statistical analysis to reach to the final results and conclusions. The demographic data were presented in tables and graphs. The data obtained in the studies were subjected to tests of significance. GraphPad InStat (www.graphpad.com) software was used for statistical analysis of data: Kolmogrov-Smirnov test was applied to test the normality of data.

For within the groups' comparison (intra-group comparison): For objective parameters: Repeated measures ANOVA test (when data passed normality test), Friedman test (when data failed normality test) and Wilcoxon Matched – Pairs Signed – Ranks test were applied. For between the groups' comparison (inter – group comparison): For objective parameters: Unpaired 't' test (when data passed normality test), Mann - Whitney test (when data failed normality test) were applied. P value < 0.05 was considered as level of statistical significance.

Results

Demographic details

Total number of patients included in study were 60 and their age group was from 21 to 40 years, this shows that they are obvious from the reproductive age group as study is for Folliculogenesis. In Trial Group and Control Group, maximum patients were from graduate group i. e.16 (53.3%) and 18 (60%), respectively. Out of total 60 patients, 27 (45%) patients were housewives. Out of 60 patients enrolled for the study in both groups maximum patients were from Middle classs i. e. 34 (56.67%). Patients with minimum 1 year of marital status and maximum 9 years of marital status were enrolled in this study. All of them were having anovulatory cycles. About 49 (81.67%) patients were having primary infertility whereas only 11 (18.33%) patients were having secondary infertility.

Clinical assessment

a) Follicle size: In Trial Group, the mean Follicle Size before treatment, on 1st cycle, 2nd cycle and After Treatment were 0.6 ± 0.56 mm, 0.77 ± 0.576 mm, 1.23 ± 0.50 mm and 2.23 ± 0.90 mm respectively. The difference in Follicle Size was found to be statistically significant ($p < 0.0001$). In Control Group, the mean Follicle Size before treatment, on 1st cycle, 2nd cycle and After Treatment were 0.77 ± 0.63 mm, 2.00 ± 0.69 mm, 2.37 ± 0.72 mm and 2.57 ± 0.77 mm respectively. The difference in Follicle Size was found to be statistically significant ($p < 0.0001$). The difference in mean Follicle Size of Trial Group and Control Group on 1st and 2nd cycle were statistically significant ($p < 0.0001$, $p < 0.0001$) whereas it was statistically insignificant at the end of the treatment ($p = 0.1196$).
b) Endometrial Thickness (ET): In Trial Group, the mean Endometrial Thickness (ET) before treatment, on 1st cycle, 2nd cycle and After Treatment

were 0.63 ± 0.76 mm, 0.87 ± 0.86 mm, 1.40 ± 0.72 mm and 2.17 ± 0.83 mm respectively. The difference in ET was found to be statistically significant ($p < 0.0001$). In Control Group, the mean Endometrial Thickness (ET) before treatment, on 1st cycle, 2nd cycle and After Treatment were 0.90 ± 0.76 mm, 1.90 ± 0.71 mm, 2.13 ± 0.73 mm and 2.37 ± 0.67 mm respectively. The difference in ET was found to be statistically significant ($p < 0.0001$). The difference in mean ET of Trial Group and Control Group on 1st and 2nd cycles were statistically significant ($p < 0.0001$, $p = 0.0005$). The difference in mean ET after treatment of Group A and Group B was statistically insignificant ($p = 0.4451$).

c) Interval between two MCs: In Trial Group, the mean interval between two MCs before treatment, on 1st cycle, 2nd cycle and After Treatment were 2.33 ± 0.76 days, 2.00 ± 0.83 days, 1.40 ± 0.81 days and 1.03 ± 1.01 days respectively. The difference in mean interval between two MCs was found to be statistically significant ($p < 0.0001$). In Control Group, the mean interval between two MCs before treatment, on 1st cycle, 2nd cycle and After Treatment were 2.13 ± 0.78 days, 1.90 ± 0.76 days, 1.20 ± 0.76 days and 0.80 ± 0.92 days respectively. The difference in mean interval between two MCs was found to be statistically significant ($p < 0.0001$). The difference in mean interval between two MCs of Trial Group and Control Group on 1st, 2nd cycle and after treatment were statistically insignificant ($p < 0.6599$, $p = 0.3503$ and $p = 0.1059$ respectively).

Regulation of menses

Out of 30 patients from trial group, 13 (43.33%) patients had regulation of menses and out of 30 patients from control group, 18 (60%) patients had regulation of menses.

Conception

Out of 30 patients from trial group 05 (16.67%) patients from trial group and 07 (23.33%) patients from control group got conceived during study period.

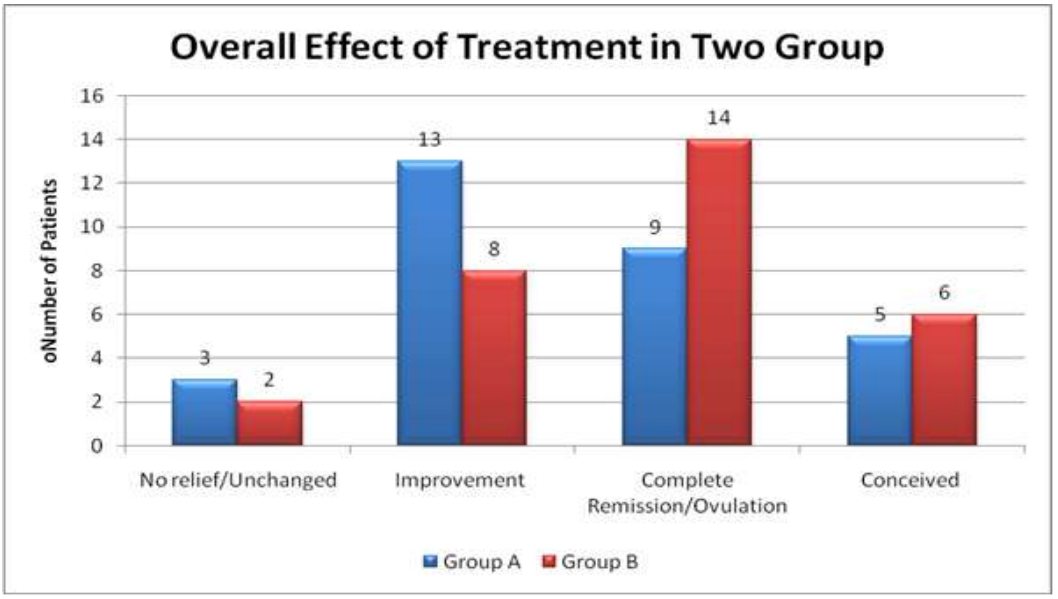
Laboratory investigations

Hormonal profile and haematological profile were carried out on the second day of menses before treatment and after treatment but there was no difference seen on Sr. LH, Sr. FSH, Sr. PRL, Sr. AMH, Hb gm%, RBC and WBC levels in inter group comparison and intra group comparison. *Ashwagandha kshirpaka* has significant effect on Platelet count and ESR. This suggests that it has immunomodulatory effect of it.

Overall assessment of treatment

In Group A, 05 patients conceived, 09 patients got complete remission/ovulation, 13 patients showed improvement whereas 03 did not get relief. In Group B, 06 patients conceived, 14 patients got complete remission/ovulation, 08 patients showed improvement whereas 02 did not get relief. On Overall Assessment of

treatment, the difference between two groups was found to be statistically insignificant (p = 0.1424)



Graph No. 1: Overall assessment of treatment in two groups

Discussion

For the clinical study 'Comparative study to assess the efficacy of *Ashwagandha Kshirpaka* for Folliculogenesis in management of *Vandhyatva* with Clomiphene Citrate' 60 patients were selected and divided in two groups. 30 patients received *Ashwagandha kshirpaka* (100ml *kshirpaka* was given orally from 5th day of menses for 12 days before breakfast) and 30 patients received Clomiphene citrate (50mg Tab was given from 3rd day of menses for 5 days once in a day) for consecutive three cycles. The patients were from age group of 21 to 40 years which showed that they are obvious from the reproductive age group as study is for Folliculogenesis. There may be maximum intake of *Mithya Ahar-Vihar*, change in life-style, excessive stress, and consumption of fast food which leads to disturbed hormonal levels and affects Folliculogenesis. The clinical assessment of patients before and after the treatment showed that-

Follicular size

It was seen that the follicular size increased gradually in patients who received *Ashwagandha Kshirpaka* and Clomiphene citrate treatment i. e. Trial group and Control group which was statistically significant too. Inter-Group comparison showed that the difference in mean follicular growth in trial and control groups was statistically insignificant. It can be inferred that *Ashwagandha Kshirpaka* is as potent drug as Clomiphene citrate which is considered to be the gold standard drug for the Folliculogenesis.

Endometrial thickness

It was seen that endometrial thickness increased with the consumption of *Ashwagandha kshirpaka* and clomiphene citrate too which was statistically significant. Inter-Group comparison showed that the difference in mean endometrial thickness in trial and control groups was statistically insignificant. Hence, it can be said that there was effect of *Ashwagandha kshirpaka* on endometrial thickness was comparable to that of Clomiphene citrate.

Interval between two MCs

It was seen that the mean interval between two MCs improved with the consumption of *Ashwagandha kshirpaka* and clomiphene citrate both which was statistically significant too. Inter-Group comparison showed that the difference in mean interval between two MCs in trial and control groups was statistically insignificant. Hence, it can be said that there was effect of *Ashwagandha kshirpaka* on mean interval between two MCs was comparable to that of Clomiphene citrate.

Regulation of menses

13 (43.3%) patients from trial group and 18 (60%) patients from control group had regulation of menses with no statistically significant difference between two groups.

Conception

Out of 30 patients from trial group 05 (16.67%) patients got conceived, whereas 07 (23.33%) patients got conceived from control group. So, it can be said that the conception rate is satisfactory in trial group compared to that of the control group because *Ashwagandha kshirpaka* is having anti-inflammatory effect, immunomodulation activity, antibiotic activity, anti-depressant activity. On Overall Assessment of treatment, the difference between two groups was found to be statistically insignificant.

Consumption of fast food, bakery items, sedentary lifestyle, and stress are some of the causative factors that were noted during this study. All these causative factors disturb the normal functioning of H-P-O axis which leads to the impairment of the normal functioning of Folliculogenesis. *Ashwagandha Kshirpaka* has insignificant effect on conception. This was the additional observation that was observed.

As mentioned by *Bhavaprakasha* in *Yonirogadhikar*, main content of *Ashwagandha Kshirpaka* is *Ashwagandha.* Along with it, *Godugdha* and *Goghrita* are also added in its preparation. With the help of their individual properties, it helps to correct Folliculogenesis as well as ovulation. [7,8] Regular intake of *Vata prakopak Ahar-vihar* has its effect on *dosha* i. e. *Vatavruddhi* and *Kaphakshaya* takes place. Due to *Dosha-Dushti,* there is abnormality in the normal functioning of the *Beeja* and no further *Beeja Vriddhi* is seen leading to defect in *Beejotsarga*. *Ashwagandha Kshirpaka* with its properties like *Laghu, Snigdha Guna* and *Madhur Vipaka*, alleviates the aggravated *Vata dosha* ultimately increases *Kapha*

dosha. Madhura Rasa with its *Brimhana* and *Pushtikara* properties nourishes *Rasavaha Srotas* which in turn nourishes *Artavavaha Srotas* and *Beeja Vriddhi* takes place resulting in *beejotsarga* i. e. ovulation. The small sample size and limited geographical coverage as well as less advanced efficacy parameters were the limitations of the present study.

Conclusion

Folliculogenesis is one of the major factors responsible for the fertilization. *Ashwagandha Kshirapaka* and Clomiphene citrate were found to be equally efficacious in Folliculogenesis. No side-effects were observed with *Ashwagandha kshirpaka*. *Ashwagandha kshirpaka* was concluded as an effective remedy for *Samprapti Bhanga* of *Abijotsarga* i.e anovulation.

References

1. Deshpande, P. S., & Gupta, A. S. (2019). Causes and Prevalence of Factors Causing Infertility in a Public Health Facility. *Journal of human reproductive sciences*, *12*(4), 287–293. https://doi.org/10.4103/jhrs.JHRS_140_18
2. Walker MH, Tobler KJ. Female Infertility. [Updated 2021 Dec 28]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 Jan-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK556033/
3. Katsikis, I., Kita, M., Karkanaki, A., Prapas, N., & Panidis, D. (2006). Anovulation and ovulation induction. *Hippokratia*, *10*(3), 120–127
4. Zech NH, Zech M, Baldauf S, Comploj G, Murtinger M, Spitzer D, Hradecký L, Ajayi R, Schuff M, Zech H. Ovarian stimulation in ART - Unwinding pressing issues. Minerva Ginecol. 2015 Apr;67(2):127-47.
5. Acharya Sushruta. Sushrut Samhita with commentary Ayurveda Tatvasandipika by Kaviraj Ambikadatta Shastri, Chaukhamba Sanskitti Sansthan, Varanasi 2012, Sharirsthana Ch:2/33, p. 348.
6. Acharya Agnivesha: Charaka Samhitha with Dipika commentary of Chakrapanidatta, edited by Vaidya Yadavji Trikamji Acharya, Published by-Chaukhambha Surbharathi Prakashan, Varanasi, Reprint Edition 2000; Chikitsa Sthana Ch:30/48, p. 637.
7. Pandit Shree Brahma Shankara Mishra, Bhavaprakasha of Shri Bhavmishra, Volume 2, Chapter No.70, Chaukhambha Sanskrit Bhawan, Varanasi 2015, Page no. 771
8. Prof. Premvati tewari, ayurvediya prasutitsntran evum striroga, Part1, prasutitantra, Chapter No.2, Chaukhambha Orientalia, Varanasi, 2nd edition 1999, Reprint 2017, Page no. 40-44