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RESEARCH ARTICLE

Evaluation of Effect of Seed Cycling on Anthropometric, Biochemical, Hormonal and Nutritional Parameters in the Women Diagnosed with Polycystic Ovarian Syndrome – A Cohort Interventional Study Design

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Abstract

Background: Polycystic Ovary Syndrome (PCOS) is an endocrine disorder of reproductive age group in women and is characterized by obesity, hyper-insulinemia, hyperandrogenism and insulin resistance. Seed cycling has emerged as a potential treatment modality for managing PCOS symptoms. **Methods:** A total of 290 women diagnosed with PCOS, aged 18-40, and were enrolled in this study after meeting specific eligibility criteria. Participants were randomly divided into two groups: a control group (n=145) and an intervention group (n=145). The control group received a portion-controlled diet along with a daily dose of Metformin 500 mg for 12 weeks. The intervention group followed the same diet but incorporated seed cycling into their regimen. Assessments, including anthropometric measurements, biochemical analyses, hormonal evaluations, and nutritional assessments, were conducted at baseline and after the 12-week treatment period. The Mann-Whitney U test was used to compare outcomes between the two groups before and after treatment, with significance determined at $p<0.05$. **Results:** The majority of participants were aged 26-30 years (46.90%), with 60% being non-vegetarian. Among them, 36% were employed, and 14% engaged in physical activity. The intervention group demonstrated significant improvements in anthropometric, biochemical, and hormonal parameters compared to the control group. Notably, there was a decrease in mean energy consumption, carbohydrate intake, and fat intake in the intervention group, while mean protein intake remained unchanged between the groups after 12 weeks. These improvements were statistically significant ($p<0.05$). **Conclusion:** The study concluded that women with PCOS showed substantial improvements in anthropometric, biochemical, and hormonal profiles following seed cycling therapy combined with dietary modifications. This suggests that seed cycling, along with a portion-controlled diet, can be an effective strategy for managing PCOS symptoms.

Keywords: Flaxseeds, PCOS, Pumpkin Seeds, Seed Cycling, Sesame Seeds, Sunflower Seeds

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1. Introduction

Polycystic Ovary Syndrome (PCOS) was initially described in 1935 and is sometimes referred to as Stein-Leventhal Syndrome. It is the most prevalent neuroendocrine and ovarian disorder affecting women in their reproductive years[1,2]. Globally, PCOS diagnosed by different diagnostic criteria, has an escalating prevalence ranging from 4% to 21%[3-7]. A meta-analysis demonstrates 11.34% of pooled prevalence estimates[8]. During the reproductive life of a female, in the non-pregnant state, there are cyclical changes taking place in various organs called reproductive cycles including Ovarian and Uterine cycles. The pulsatile secretion of GnRH and gonadotropins regulates these cycles via the Hypothalamo-pituitary-gonadal axis and the feedback actions of oestrogen and progesterone. At the start of the cycle, i.e., during the follicular phase, there are low levels of oestrogen and progesterone, therefore minimum feedback inhibition leads to increased GnRH secretion which further increases Follicle Stimulating Hormone (FSH) and Luteinising Hormone (LH) secretion. This leads to the development of follicles. Towards the middle of the cycle, oestrogen secretion increases by the developing follicles, which in turn increases the responsiveness of pituitary gonadotropins to GnRH resulting in LH Surge and causing ovulation. Towards the end of the cycle, i.e., the luteal phase, the hormonal levels drop down leading to no feedback inhibition and again increases FSH and LH and the next cycle continues[9]. Hypersecretion of Luteinising Hormone, serum oestrogen and serum testosterone levels, hyposecretion of follicle-stimulating hormone, premature granulosa cells and premature arrest of ovarian follicles are the hallmarks of PCOS[10].

The clinical presentation of PCOS includes hypersecretion of androgens, irregular menstrual cycles, hirsutism, acne, and male pattern alopecia[11,12]. It is also associated with various comorbidities including obesity, dyslipidaemias, hypertension and diabetes mellitus[12,13].

In recent years, batteries of research have documented the role of adjuvant therapies in the management of PCOS including flaxseeds[14], spearmint[15], cinnamon[16], evening primrose[17], *Aloe vera*[18], liquorice[19], etc.

Various review papers have explored the role of different seeds — such as flaxseeds, sesame seeds, pumpkin seeds, and sunflower seeds — individually in improving hormonal disturbances and insulin resistance in patients with PCOS[20,21]. Currently, there is limited literature on the use of complete seed cycling as a treatment modality of PCOS, even though it also is an androgen-related disorder. Therefore, this study has been planned to evaluate in a randomized controlled manner the effects of seed cycling on the anthropometric, biochemical, hormonal and nutritional profile of women diagnosed with PCOS in the age group 18-40 years from the baseline to the end of 12 weeks.

2. Materials and Methods

The study utilized a Cohort Interventional Study Design and was conducted at the Department of Endocrinology, KLE Dr. Prabhakar Kore Hospital and Research Centre, Belagavi, from 1st April 2022 to 31st March 2023. Women diagnosed with PCOS based on Rotterdam's Criteria 2003 were enrolled during this period. According to Rotterdam's Criteria 2003, a woman must exhibit two out of three clinical findings[22]:

(a) oligo-ovulation or anovulation,
(b) clinical and/or biochemical signs of hyperandrogenism, or
(c) polycystic ovaries diagnosed by ultrasonogram.

Women with known medical illnesses such as diabetes, Impaired Fasting Glucose (IFG), Impaired Glucose Tolerance (IGT), or active thyroid disorder, or on medications such as corticosteroids and oral contraceptives were excluded from the study.

The sample size was determined using a 95% confidence interval, 80% power, and an assumed 20% attrition rate[20].

$$N = \frac{(Z\ 1\text{-}\alpha/2 \pm Z1\text{-}\beta)^2\ X\ (SD_1^2 \pm SD_2^2)}{(X1\text{-}\ X2)^2}$$

= 144 samples, approximately 145 samples in each group.

So total of 290 PCOS women were enrolled in the study.

The patients were screened as per the eligibility criteria and were enrolled for the study. Two groups were being made by using Simple Randomized Technique and were enrolled in the following two groups:

- Group I (Control Group): The control group received a portion-controlled diet along with Metformin 500 mg tablets daily for a duration of 12 weeks after breakfast, prescribed by the endocrinologist.
- Group II (Intervention Group): The intervention group followed the same portion-controlled diet regimen and incorporated seed cycling into their treatment. Seed cycling included the consumption of flaxseeds, pumpkin seeds, sunflower seeds, and sesame seeds along with dietary modifications.

Written informed consent was obtained from all enrolled patients at the time of data collection. Both groups of patients with PCOS were instructed to adhere to a 1500 kcal healthy diet plan, consisting of three meals per day (breakfast, lunch, and dinner) along with two snacks (mid-morning and evening). This diet comprised 40% carbohydrates, 20% protein, and 40% fat. Study participants were taught to use spoon measurements for 15 gms or 1 tablespoon. The medicines were purchased by the study participants, while the seeds used for seed cycling were provided by the investigators to all participants in the intervention group. This ensured that all participants in the intervention group had equal access to the seeds necessary for the study.

2.1 Seed Cycling Therapy

The study participants in the intervention group were explained about the phases of the menstrual cycle and seed cycling. Pumpkin seeds and flaxseeds were recommended to be ingested during the follicular phase, i.e., 1-14 days of the menstrual cycle; whereas sunflower seeds and sesame seeds were recommended during the luteal phase, i.e., from the 15th day onwards till the beginning of next cycle. The participants were

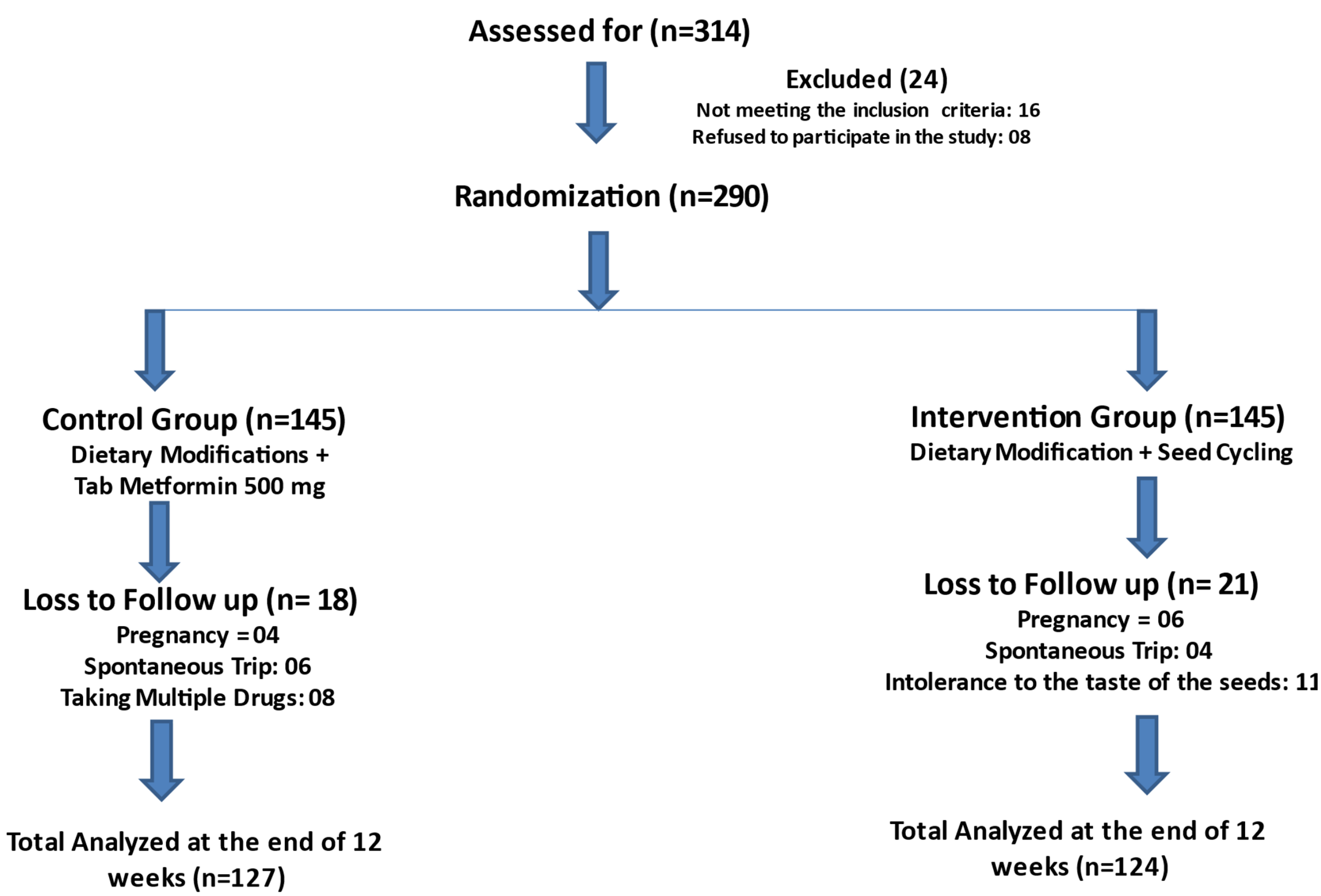

Figure 1. Consort flow diagram for enrolment.

asked to use the seeds to add to salads, grind with chutneys, consume it as a mouth freshener post-meal, mix with curd or buttermilk or sprinkle it over soups.

2.2 Study Parameters

A pre-tested, semi-structured questionnaire, validated through a pilot study using Cronbach's alpha test with a score of 0.85, was administered to collect information on age, diet preferences, working status, and physical activity practices.

2.3 Anthropometric Parameters

2.3.1 Weight in Kgs

Body weight was measured (to the nearest 0.01 kg) with the subject standing still on the electronic weighing scale, feet about 15 cm apart, and weight equally distributed on each leg. Subjects were instructed to wear minimum outerwear (as culturally appropriate) and no footwear while their weight was being measured.

2.3.2 Height in cms

Height was measured using a non-stretchable tape (to the nearest 0.1 cm) with the subject in an erect position against a vertical surface and the head positioned so that the top of the external auditory meatus was at level with the inferior margin of the bony orbit.

2.3.3 Body Mass Index (BMI)

BMI was calculated using the formula – weight (kg)/ height (m^2), i.e., Quetelet's equation. Subjects Body Mass Index (BMI) was categorized by the Asia-Pacific classification: underweight (<18.5 kg/m^2), normal weight (18.5-22.9 kg/m^2), overweight (23.0-24.9 kg/m^2), obesity Class I (25.0-29.9 kg/m^2), and obesity Class II (≥ 30.0 kg/m^2)[23].

2.3.4 Waist Circumference (cm)

Waist circumference was measured using a measuring tape, around the trunk at a marked level on the right side, with minimal respiration, and recorded to the nearest 0.1 cm[23].

2.3.5 Hip Circumference (cm)

Hip circumference was measured by using measuring tape placed at the maximum extension of the buttock[23,24].

2.3.6 Waist Hip Ratio (WHR)

The waist-hop ratio was calculated by dividing the waist circumference by the hip circumference[23,24].

2.4 Biochemical Profile

Fasting Blood Sugar, Lipid Profile and Serum TSH values were noted.

2.5 Hormonal Profile

Serum Total Testosterone, FSH, LH, Prolactin and Testosterone levels were noted.

All laboratory investigations were carried out at the Medical College and Hospital Lab. The costs for these investigations were borne by the patients, and the values were recorded from their case papers.

2.6 Nutritional Assessment

This was done by using a 24-hour dietary recall method for 3 days. The average value was calculated and entered into the Excel sheet.

The data was inputted into IBM SPSS software version 20.0. Subsequently, thorough data cleaning and scrutiny were conducted to address missing values and ensure consistency with the original forms. All the enrolled patients in the intervention group were called telephonically once in 15 days to monitor their compliance and also reminder mobile text messages were sent to each participant during different phases of their cycle to remind them of seeds to be used.

2.7 Statistical Analysis

The normal distribution of each study variable was assessed using the Kolmogorov-Smirnov test. Non-parametric tests, specifically the Mann-Whitney U test and Wilcoxon signed-rank test, were utilized to identify differences from baseline to the end of the 12 weeks. A significance level of $p<0.05$ was applied.

2.8 Dietary and Compliance Monitoring

To ensure adherence to the 1500 kcal diet and the recommended seed consumption, random cross-checks were conducted by the investigators for both groups. Participants were periodically contacted and asked to maintain dietary logs, which were reviewed during these cross-checks.

3. Results

The study enrolled a total of 290 women diagnosed with Polycystic Ovary Syndrome (PCOS), aged between 21 and 40 years, as there were no patients in the 18-20 age group. The majority of participants (46.90%) were within the 26-30 years age range. Regarding dietary habits, 60% of the participants were non-vegetarian. Employment status varied, with 36% of the women employed, and physical activity levels were low, with only 14% of the participants engaging in some form of physical exercise. These demographic characteristics were similar between the control and intervention groups, ensuring comparability for the subsequent analyses.

Table 1. Comparison of control group and intervention group before and after treatment anthropometric parameters scores by Mann-Whitney U test

Parameter	Treatment times	Control group			Intervention group			U-value	Z-value	p-value
		Mean	SD	Mean rank	Mean	SD	Mean rank			
Weight (kg)	Before	68.02	3.67	126.42	67.92	3.47	124.00	7820.50	0.0922	0.9266
	After	67.76	3.71	125.72	67.76	3.42	124.00	7839.00	-0.0600	0.9522
	Difference	0.26	1.03	125.63	0.16	0.55	124.00	7826.50	-0.0817	0.9349
BMI (kg/mt^2)	Before	27.80	1.61	130.72	27.63	1.61	124.00	7275.00	1.0407	0.2980
	After	27.65	1.65	128.56	27.57	1.60	124.00	7549.00	0.5643	0.5726
	Difference	0.14	1.09	127.26	0.06	1.07	124.00	7714.50	0.2765	0.7822
WC (cm)	Before	84.65	0.98	124.85	84.68	1.01	124.00	7728.50	-0.2521	0.8009
	After	83.74	0.96	170.95	82.41	1.17	124.00	2165.00	9.9266	0.0001*
	Difference	0.91	0.29	70.12	2.27	0.84	124.00	777.50	-12.3393	0.0001*
HC (cm)	Before	97.54	1.07	124.55	97.60	1.13	124.00	7689.50	-0.3200	0.7490
	After	97.28	1.14	138.81	96.81	1.22	124.00	6247.00	2.8283	0.0047*
	Difference	0.26	0.44	96.57	0.78	0.55	124.00	4136.50	-6.4983	0.0001*
WHR	Before	0.87	0.01	126.51	0.87	0.01	124.00	7809.00	0.1122	0.9107
	After	0.86	0.01	152.08	0.85	0.02	124.00	4562.00	5.7584	0.0001*
	Difference	0.01	0.01	75.50	0.02	0.01	124.00	1461.00	-11.1508	0.0001*

Significant improvements in waist circumference, hip circumference, and waist-hip ratio (*$p<0.05$) were observed in the intervention group, compared to the control group, while other anthropometric parameters showed no significant differences between the groups.

Table 2. Comparison of control group and intervention group before and after treatment biochemical parameters scores by Mann-Whitney U test

Parameter	Treatment times	Control group			Intervention group			U-value	Z-value	p-value
		Mean	SD	Mean rank	Mean	SD	Mean rank			
FBS (mg/dL)	Before	108.65	3.74	122.16	109.10	3.75	125.11	7386.00	-0.8477	0.3966
	After	107.26	4.30	150.59	104.27	3.76	154.23	4751.50	5.4289	0.0001*
	Difference	1.39	5.74	99.61	4.83	0.67	102.02	4522.00	-5.8280	0.0001*
TSH (mIU/mL)	Before	1.86	0.53	126.74	1.85	0.53	129.81	7780.00	0.1626	0.8708
	After	1.85	0.53	126.24	1.85	0.53	129.29	7844.00	0.0513	0.9591
	Difference	0.01	0.68	126.20	0.00	0.59	129.26	7848.00	0.0443	0.9646
TC (mg/dL)	Before	186.70	1.20	122.67	186.82	1.24	125.64	7451.00	-0.7347	0.4625
	After	183.95	2.22	183.59	176.57	3.25	188.03	560.50	12.7167	0.0001*
	Difference	2.75	2.48	69.24	10.25	3.42	70.92	666.00	-12.5332	0.0001*
TG (mg/dL)	Before	171.67	9.25	123.50	172.56	9.38	126.48	7556.00	-0.5521	0.5809
	After	162.22	10.60	157.70	150.24	11.20	161.52	3847.50	7.0009	0.0001*
	Difference	9.45	11.59	89.51	22.32	11.50	91.68	3240.00	-8.0572	0.0001*
LDL (mg/dL)	Before	115.76	3.65	126.11	115.69	3.52	129.16	7860.50	0.0226	0.9820
	After	106.39	5.18	181.09	95.69	3.52	185.47	878.00	12.1646	0.0001*
	Difference	9.37	3.72	67.91	20.00	0.00	69.55	496.00	-12.8288	0.0001*
HDL (mg/dL)	Before	43.06	1.82	125.30	43.09	1.82	128.33	7785.50	-0.1530	0.8784
	After	45.00	1.86	67.08	51.38	2.91	68.70	391.00	-13.0114	0.0001*
	Difference	1.94	0.46	64.00	8.29	2.20	65.55	0.00	-13.6913	0.0001*

Statistically significant improvements (*p<0.05) were observed in fasting blood sugar levels and lipid profile in the intervention group at the end of 12 weeks, compared to the control group.

Table 3. Comparison of control group and intervention group before and after treatment Hormonal parameters scores by Mann-Whitney U test

Parameter	Treatment times	Control group			Intervention group			U-value	Z-value	p-value
		Mean	SD	Mean rank	Mean	SD	Mean rank			
FSH (mIU/mL)	Before	6.25	0.37	118.89	6.30	0.42	121.76	6970.50	-1.5702	0.1164
	After	5.20	0.35	187.06	3.68	0.51	191.58	119.50	13.4835	0.0001*
	Difference	1.06	0.21	64.00	2.62	0.34	65.55	0.00	-13.6913	0.0001*

Table 3. Continued...

Parameter	Treatment times	Control group			Intervention group			U-value	Z-value	p-value
		Mean	SD	Mean rank	Mean	SD	Mean rank			
LH (IU/L)	Before	10.30	0.37	141.24	10.16	0.34	144.66	5938.00	3.3657	0.0008*
	After	9.30	0.42	188.00	6.52	0.54	192.55	0.00	13.6913	0.0001*
	Difference	1.00	0.17	64.00	3.65	0.34	65.55	0.00	-13.6913	0.0001*
Prolactin (ng/dL)	Before	95.41	0.41	96.42	95.81	0.52	98.75	4117.50	-6.5314	0.0001*
	After	87.46	0.49	187.97	83.00	1.51	192.52	4.00	13.6844	0.0001*
	Difference	7.96	0.32	64.00	12.81	1.40	65.55	0.00	-13.6913	0.0001*
Testosterone (ng/dL)	Before	85.19	5.03	136.44	83.78	4.17	139.74	6548.00	2.3049	0.0212*
	After	83.50	5.11	146.58	80.91	3.76	150.13	5260.00	4.5446	0.0001*
	Difference	1.69	0.39	110.49	2.86	4.13	113.17	5904.50	-3.4239	0.0006*

A statistically significant improvement (*p<0.05) in hormonal profiles was observed at the end of 12 weeks in the intervention group, compared to the control group.

Table 4. Comparison of control group and intervention group before and after treatment nutritional parameters scores by Mann-Whitney U test

Parameter	Treatment times	Control group			Intervention group			U-value	Z-value	p-value
		Mean	SD	Mean rank	Mean	SD	Mean rank			
Energy (Kcal)	Before	1929.3	101.64	123.74	1928.3	64.49	126.73	7586.50	-0.4991	0.6177
	After	1745.7	99.25	171.93	1633.6	68.86	176.09	2041.00	10.1422	0.0001*
	Difference	183.64	28.91	64.00	294.71	11.16	65.55	0.00	-13.6913	0.0001*
CHO (gm)	Before	277.25	4.30	137.23	275.66	5.26	140.55	6447.50	2.4797	0.0132*
	After	254.42	4.50	182.28	242.48	5.29	186.69	726.50	12.4280	0.0001*
	Difference	22.83	1.96	64.00	33.18	3.47	65.55	0.00	-13.6913	0.0001*
Proteins (gm)	Before	69.97	4.75	133.11	69.39	5.44	136.33	6970.50	1.5702	0.1164
	After	82.98	5.79	118.44	84.84	7.94	121.30	6913.50	-1.6694	0.0950
	Difference	13.01	3.43	126.25	15.44	6.28	129.30	7842.50	0.0539	0.9570
Fats (gm)	Before	56.54	1.44	126.23	56.66	1.82	129.28	7845.00	0.0496	0.9605
	After	50.41	1.67	167.07	49.74	1.84	171.11	2658.00	9.0693	0.0001*
	Difference	6.13	0.37	64.00	6.92	0.22	65.55	0.00	-13.6913	0.0001*

A statistically significant difference was observed in carbohydrate (CHO), fat and energy values, while no change was observed in protein levels.

4. Discussion

The objective of the study was to assess the impact of seed cycling on women diagnosed with PCOS. A total of 290 women aged between 18 and 40 years were included after undergoing eligibility screening. These women with PCOS were divided into two groups: an Intervention Group and a Control Group, each consisting of 145 participants (Figure 1). In the 'Control Group', women with PCOS received advice on a portion control diet and were prescribed metformin 500 mg tablets per day by an endocrinologist or physician for the duration of 12 weeks. The 'Intervention Group' followed the advice on portion control diet and seed cycling for the same 12-week period. The majority of participants (46.90%) fell within the age group of 26-30 years. Additionally, 60% of participants identified themselves as non-vegetarians, while 45.6% were vegetarians. A significant portion (36%) of participants were employed, with 87% reporting no involvement in physical activity (Table 1).

4.1 Anthropometric Parameters

There was a significant improvement in waist circumference, hip circumference and waist-hip ratio (*$p<0.05$) was observed in the intervention group, as compared to the control group, whereas other anthropometric parameters like weight, and BMI showed no difference between the groups at the end of 12 weeks. Whereas, in-between the groups was significant difference was observed in weight in the control group with a 0.38% change from baseline to the end of 12 weeks. No changes were observed between the groups at the end of 12 weeks. However, there was a 0.5% change observed in the control group and a 0.22% change observed in the intervention group. However, the percentage of difference was statistically insignificant. Waist circumference and Hip circumference showed significant improvement at the end of 12 weeks. In the control group, the mean waist circumference was 83.74±0.96 cm and 82.41±1.17 cm in the intervention group respectively. Likewise, Hip circumference in the control group was 97.28±1.14 cm as compared to 96.81±1.22 cm in the intervention group. Within-group comparison of waist circumference showed 1.07% improvements in the control group and 2.68% improvement in the intervention group respectively. Within-group comparison of hip circumference showed 0.27% improvement in the control group and 0.80% improvement in the intervention group, respectively. Waist-Hip Ratio significantly improved in the intervention group with a mean of 0.85±0.02, as compared to 0.86±0.01 in the control group at the end of 12 weeks of seed cycling. The percentage change in the control group was observed as 0.80% and 1.89% in the intervention group. Both groups showed a significant percentage of improvement concerning anthropometric parameters, but the percentage changes were more profound in the intervention group (Table 1).

A study done by Rasheed et al. in 2021 reported a reduction in body weight in the experimental group (93.10±2.01 kg to 81.65±0.73 kg), who were treated with portion control diet and seed cycling as compared to second experimental group (95.85±1.0320 kg to 87.95±0.62 kg), who were treated with portion control diet and Tablet Metformin 500 mg/day for the period of 90 days. Seed cycling along with a portion-controlled diet is effective for the management of body weight in women with PCOS. The present study also has similar findings[20].

4.2 Biochemical Profile

Fasting blood glucose levels have shown significant improvement in the intervention group with a mean of 104.27±3.76 mg/dl, as compared to a control group with a mean of 107.26±4.30 mg/dl at the end of 12 weeks. About 1.28% of improvement was observed in the control group and 4.43% of improvement was observed in the intervention group. Thyroid Stimulating Hormone (TSH) levels were within the range for both groups and further did not show any significant change between the groups and within the groups. In the lipid profile, at the end of 12 weeks, the total cholesterol level was 176.5±3.25 mg /dl in the intervention group and 183.95±2.22 mg /dl in the control group. Similarly, Total Triglyceride levels were 150.2±11.20 mg/dl in the intervention group and 162.22±10.60 mg/dl in the control group. The mean LDL levels were 95.69±3.52 mg/dl in the intervention group and 106.39±5.18 mg/dl in the control group. The mean HDL levels were 51.38±2.91 mg/dl in the intervention group and 45.0±1.89 mg/dl in the control group. The Total cholesterol, Total triglycerides and LDL levels decreased significantly, whereas HDL levels increased significantly at the end of 12 weeks between the groups. With the group, the control group FBS levels improved by 12.8%, TC by 1.4%, TG by 5.50%, LDL by 80.9%, and HDL by 4.52% respectively, whereas the intervention group FBS improved by 4.43%, TC by 5.49%, LDL by 17.29%, and HDL by 19.29% respectively. Both groups showed a significant percentage of improvement concerning biochemical values, but the percentage changes were more profound in the intervention group (Table 2).

A previously conducted randomized open-labelled controlled clinical trial with flaxseeds supplementations on 41 women with PCOS reported a significant decrease in fasting blood glucose, serum insulin levels, HOMA-IR, serum triglycerides and an increase in HDL levels[14,25].

A meta-analysis of twenty studies examining the effect of whole flaxseed supplementation on lipids profile among young adults had reported reduced in total and LDL cholesterol, whereas no significant changes were found in the concentrations of HDL cholesterol and triglycerides. The results of this meta-analysis support the inclusion of flaxseed in the seed cycling regimen used in the present study. Given that women with PCOS often exhibit abnormal lipid profiles, the observed improvements in blood lipid levels through flaxseed interventions align with our findings of enhanced biochemical parameters in the intervention group. This evidence underscores the potential of seed cycling, including flaxseed, to contribute to the overall management of PCOS symptoms[26].

4.3 Hormonal Profile

Hormonal profiles, specifically serum levels of FSH, LH, Prolactin and Testosterone demonstrated significant improvement in the intervention group after 12 weeks. At the end of the 12 weeks, FSH levels showed statistically significant improvement, with a mean of 3.68±0.51 in the intervention group compared to 5.20±0.35 in the control group. Likewise, the mean LH level was 6.52±0.54 in the intervention group versus 9.30±0.42 in the control group. Prolactin levels were 83±1.51 in the intervention group and 87.46±0.49 in the control group, while testosterone levels were 80.91±3.76 in the intervention group and 83.50±5.11 in the control group. Overall, there was a decrease in FSH, LH, Prolactin, and Testosterone levels in the intervention group, compared to the control group at the end of 12 weeks, and these changes were statistically significant. Within the groups, percentage changes from baseline were observed: in the control group, FSH levels improved by 16.88%, LH by 9.08%, Prolactin by 8.34%, and Testosterone by 1.99%. In contrast, the intervention group showed more substantial improvements, with FSH levels improving by 41.61%, LH by 35.87%, Prolactin by 13.37%, and Testosterone by 3.42%, respectively. Although both groups exhibited significant improvements in hormonal profiles, the intervention group demonstrated more pronounced percentage changes, compared to the control group (Table 3).

The findings of this study align with previous research indicating increased serum FSH and LH levels in women diagnosed with PCOS[27-29]. Excessive serum testosterone during the development and progression of PCOS can inhibit oestrogen secretion and decrease Sex Hormone-Binding Globulin (SHBG) and FSH production. This hormonal imbalance, characterized by elevated LH and decreased FSH levels, contributes to disruption in the follicular phase of the ovarian cycle[30].

Several studies have highlighted the positive impact of seed cycling on improving FSH levels[20,21,25]. In the present study, serum prolactin levels also showed significant improvement in the intervention group. Previous research has documented increased prolactin levels in women with PCOS during both the follicular and luteal phases, which can negatively impact ovarian follicle size and ovulation[31-33].

4.4 Energy and Macronutrients

There was a significant improvement in energy intake values. The intervention group had a consumption of mean energy of 1633.6±68.66 kcal, as compared to 1745.7±99.25 kcal in the control group at the end of 12 weeks. The carbohydrate mean value was 242.48±5.29 kcal in the intervention group, as compared to 254.42±4.50 kcal in the control group. The fat's mean value was 49.74±1.84 kcal in the intervention group as compared to 50.41±1.67 kcal in the control group. There was improvement in terms of decreased mean energy consumption in kcal, and decreased mean carbohydrate and fat intake in the intervention group, as compared to the control group; whereas mean protein intakes didn't show any change at the end of 12 weeks between the groups. Within the groups, in the control group, the percentage change improvement in energy levels was 9.52%, Carbohydrates 8.24%, protein 3.42% and fats 10.84% at the end of 12 weeks. In the intervention group, the percentage change improvement in energy levels was 15.28%, Carbohydrates 12%, protein 22.25% and fats 12.22% at the end of 12 weeks. Although both groups showed a significant percentage of improvement concerning energy levels and macronutrients, the percentage changes were more profound in the intervention group (Table 4).

There is currently much interest in phytochemicals as bioactive molecules of food. The seed cycling has shown promising improvement in the intervention group in our study, as compared to the control group.

4.5 Mechanism of Action

Consumption of pumpkin seeds and flaxseeds during follicular phases of the ovarian cycle controls estrogenic activity. Pumpkin seeds act as a potent vasodilator[34], antioxidants, anti-inflammatory[35,36], insulin sensitizer[37], and anti-hyperlipemic[38]. Flaxseed is the richest source of mammalian lignan and has estrogenic agonist or antagonist properties[39]. Both flax and pumpkin seeds are rich in Zinc and omega-3 fatty acids, which further help in the synthesis of progesterone hormone for luteal and regulate the follicular phase. These seeds improve blood flow to the uterine wall and help in follicular function[40-43]. Consumption of sesame seeds and sunflower seeds during the luteal phase of the ovarian cycle helps in the synthesis of progesterone. They contain lignin and zinc, which help in inhibiting the excess synthesis of oestrogen. Additionally, sunflower seeds are high in vitamin E and selenium, which further enhance the synthesis and action of progesterone[42-45].

5. Conclusion

Women diagnosed with PCOS have shown a significant improvement in anthropometric, biochemical and hormonal profiles with the administration of seed cycling therapy along with dietary modifications.

5.1 Strengths of Study

The study's strengths lie in its comprehensive evaluation of the anthropometric, biochemical, hormonal, and nutritional profiles, which provided a holistic view of the effects of seed cycling in the management of PCOS. The random assignment of participants to control and intervention groups effectively minimized bias and enhanced the validity of the results. Additionally, the implementation of a standardized 1,500 kcal diet plan for both groups ensured consistency in dietary intake throughout the study. The use of a pre-tested, semi-structured questionnaire, validated with a Cronbach's alpha score of 0.85, further contributed to the reliability of data collection.

5.2 Limitations of the Study

The study faced several limitations, including the reliance on some self-reported data, which may introduce recall bias and inaccuracies. The 12-week duration of the study may not adequately capture the long-term effects and sustainability of seed cycling on PCOS symptoms. Additionally, since the research was conducted at a single medical college and hospital lab, the findings may not be generalizable to a broader population. Furthermore, the costs for laboratory investigations and medications were borne by the patients, which could influence their compliance and participation in the study.

5.3 Recommendations

Future studies should consider longer follow-up periods to assess the long-term effects and sustainability of seed cycling in PCOS management. Conducting multi-centre trials would enhance the generalizability of the findings across diverse populations. Additionally, implementing a double-blind study design can further reduce bias and increase the robustness of the findings. Using technology, such as mobile apps or digital food diaries, could improve the accuracy of dietary intake reporting.

6. Future Scope of the Study

Future research should investigate the biological mechanisms through which seed cycling influences hormonal and metabolic parameters in PCOS. Additionally, comparing seed cycling with other dietary interventions can help determine the most effective strategies for managing PCOS symptoms. Assessing the psychological well-being and quality of life of participants will provide a broader understanding of the impacts of dietary interventions on PCOS. Ensuring diverse participant demographics is crucial to exploring the effects of seed cycling across different ethnicities and age groups. Finally, evaluating the cost-effectiveness of seed cycling as a treatment modality for PCOS will inform healthcare policy and practice.

7. References

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