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Original Research Article

Effect of myoinositol plus metformin versus metformin alone on high serum AMH level in infertile PCOS women: clinical and biochemical response

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ABSTRACT

Background: Polycystic ovary syndrome (PCOS), a common endocrine disorder in reproductive-age women, is characterized by insulin resistance, hyperandrogenism, and elevated serum AMH levels. Both metformin and myoinositol improve insulin sensitivity through different mechanisms, potentially enhancing reproductive outcomes. This study aimed to compare the clinical and biochemical effects of combined myoinositol and metformin therapy versus metformin alone in reducing high serum AMH levels in infertile PCOS women.
Methods: This randomized controlled trial at BSMMU's department of reproductive endocrinology and infertility (July 2022-June 2023) randomized 80 PCOS women (18-35 years) to myoinositol+metformin (n=40) or metformin alone (n=40) for 3 months. We assessed AMH (primary outcome), BMI, waist-hip ratio, menstrual regularity, hormones (LH/testosterone), glucose metabolism, and pregnancy rates, analyzed via SPSS 23 (t-tests; $p<0.05$ significant).
Results: In 80 subfertile PCOS women (40/group), myoinositol+metformin (group A) outperformed metformin alone (group B): greater reductions in BMI (27.1 to 24.5 versus 27.4 to 25.3), waist-hip ratio (0.91 to 0.86 versus 0.90 to 0.88), AMH (12.2 to 9.5 versus 12.3 to 8.6 ng/ml), LH (10.1 to 6.4 versus 9.9 to 6.9 mIU/ml), and testosterone (2.1 to 1.4 versus 2.3 to 1.7 ng/ml). Group A showed higher menstrual regularity (95% versus 87.2%) and pregnancy rates (10% versus 5.1%).
Conclusions: Combination therapy of myoinositol and metformin is more effective than metformin alone in improving clinical and biochemical outcomes in infertile PCOS women.

Keywords: Anti-Müllerian hormone, Myoinositol and metformin, Polycystic ovary syndrome

INTRODUCTION

Polycystic ovary syndrome (PCOS) represents the most prevalent endocrine disorder among women of reproductive age.[1] It is a leading contributor to anovulatory infertility, affecting approximately 5-10% of women in this population group.[1-3] This complex and heterogeneous condition is believed to arise from a multifaceted interplay between genetic predisposition and environmental influences.[4,5] While its origin involves multiple factors, the core underlying mechanisms are hyperinsulinemia and hyperandrogenemia, which account for the characteristic reproductive disturbances and the associated long-term metabolic effects.

Hyperinsulinemia works in conjunction with luteinizing hormone (LH) to stimulate androgen synthesis by theca cells.[6] Elevated insulin levels in circulation contribute to hyperandrogenism, which disrupts normal folliculogenesis and leads to polycystic ovarian morphology, along with increased levels of anti-Müllerian hormone (AMH). AMH, a dimeric glycoprotein, is part of the transforming growth factor-beta (TGF-β) superfamily of regulatory proteins.[7,8] It is solely secreted by granulosa cells within developing pre-antral and small antral follicles.[9-11] The concentration of AMH in the blood serves as an indicator of the ovarian follicle pool. Research has demonstrated that AMH levels in women diagnosed with PCOS are two to three times higher than those in women without the condition.[12,13] Therefore, serum AMH measurement is considered a dependable marker for assessing ovarian reserve and function.[14]

Due to the strong link between PCOS and insulin resistance, insulin-sensitizing agents are often employed to improve endocrine and metabolic disturbances, support normal follicular development, and reduce long-term complications associated with the syndrome.[15] These agents enhance insulin sensitivity in peripheral tissues and have been shown to improve reproductive outcomes. Among them, metformin- a widely used oral biguanide- has been thoroughly investigated in the context of PCOS. It works by reducing glucose absorption in the intestines, inhibiting hepatic gluconeogenesis, and increasing insulin sensitivity in peripheral tissues.[6] Fleming et al observed a marked reduction in serum AMH levels following 8 months of metformin therapy in women with PCOS.[16] Similarly, Neagu et al reported the resumption of ovulatory cycles in most patients after two months of treatment with 850 mg metformin twice daily, along with a notable decrease in AMH levels.[17] These findings suggest that metformin can lower AMH concentrations in PCOS, likely through mechanisms linked to improved insulin sensitivity.[18]

Besides metformin, myoinositol is available as a nutritional supplement and is known to improve insulin resistance. It is a stereoisomer of a C6 sugar alcohol that belongs to the inositol family. It reduces hyperinsulinemia by acting as a post-receptor mediator (second messenger) of insulin signalling via the membrane-associated sodium-dependent inositol co-transporter GLUT4. Myoinositol improves ovarian function, lowers the LH/FSH ratio, reduces serum androgen levels, raises sex hormone binding globulin (SHBG), and lowers serum total and free testosterone, thereby restoring the metabolic and hormonal profile in women with PCOS.[19] By improving insulin sensitivity, inositol might indirectly contribute to healthier AMH levels.[20]

Because the two insulin sensitizers work through different mechanisms, they may be used together to improve clinical, biochemical, and reproductive outcomes in infertile PCOS women. On these grounds, determining the change in serum AMH levels during treatment with myoinositol and metformin in comparison to metformin alone may offer significant hope for fertility experts and anovulatory infertile PCOS patients with high serum AMH. The purpose of this study was to assess and compare the clinical and biochemical outcomes of combined myoinositol and metformin therapy versus metformin monotherapy in reducing elevated serum anti-Müllerian hormone (AMH) levels among infertile women diagnosed with polycystic ovary syndrome (PCOS).

Objective

The aim of the study was to evaluate the clinical and biochemical effects of myoinositol plus metformin versus metformin alone on high serum AMH levels in infertile women with PCOS.

METHODS

This randomized controlled trial was conducted at the department of reproductive endocrinology and infertility, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh, from July 2022 to June 2023. The study included women diagnosed with polycystic ovary syndrome (PCOS) and subfertility, who had elevated serum anti-Müllerian hormone (AMH) levels (>6 ng/dl), attending the outpatient department of reproductive endocrinology and infertility at BSMMU. A total of 80 participants were enrolled, and they were randomly allocated to either group A (myoinositol plus metformin) or group B (metformin alone).

Inclusion criteria

Patients diagnosed with PCOS as per the Rotterdam criteria. Age between 18-35 years. Serum AMH level >6 ng/ml.

Exclusion criteria

Women with hyperprolactinemia, thyroid disorders, chronic illness, tuberculosis, or uncontrolled diabetes. PCOS patients with AMH levels <6 ng/ml. Contraindications to metformin. Use of oral contraceptives or insulin sensitizers in the past 3 months.

The study evaluated socio-demographic (age, education, income, type of sub-infertility), clinical (BMI, waist and hip circumference, waist-hip ratio), and laboratory variables (serum AMH, FSH, LH, testosterone, fasting and post-glucose blood sugar). Primary outcomes included clinical response (BMI, waist-hip ratio, cycle regularity) and biochemical changes (AMH, FSH, LH, testosterone, glucose levels), while the secondary outcome was pregnancy rate. Based on sample size estimation, 40 participants per group were enrolled to account for dropouts. Randomization was done using permuted blocks with concealed allocation. Group A received myoinositol (750 mg twice daily) plus metformin (titrated to 1500 mg/day over 3 weeks for 3 months); group B received only

metformin (500 mg thrice daily for 3 months). Participants underwent screening with history, examination, and baseline investigations. Compliance and side effects were monitored, with common adverse effects including gastrointestinal discomfort and dizziness. Data were analysed using SPSS 23. Paired and independent t-tests were used for within- and between-group comparisons, with significance set at $p<0.05$. Ethical approval was obtained from the BSMMU IRB, and informed consent was taken from all participants in accordance with the Helsinki Declaration.

RESULTS

Table 1 shows that the mean age was 26.3±4.6 years in group A (myoinositol plus metformin) and 26.6±5.1 years in group B (metformin alone), with most participants aged between 21-30 years. Primary sub-fertility was more common in both groups: 77.5% in group A and 70.0% in group B. Among secondary sub-fertile patients, 33.3% in group A and 41.7% in group B had live births. No statistically significant differences were observed between the groups ($p>0.05$).

Table 1: Socio-demographic characteristics of study participants (n=80).

Socio-demographic characteristics		Group A (myoinositol plus metformin) (n=40)		Group B (metformin alone) (n=40)		P value
		N	%	N	%	
Age (years)	18-20	4	10	6	15	
	21-25	15	37.5	12	30	
	26-30	13	32.5	13	32.5	
	31-35	8	20	9	22.5	
	Mean±SD	26.3 ± 4.6		26.6±5.1		0.782^{ns}
	Range (min-max)	18.0-34.0		18.0-35.0		
Sub-fertility	Primary	31	77.5	28	70	0.446^{ns}
	Secondary	9	22.5	12	30	
Fertility outcome among secondary sub-fertile patients	Live birth	3	33.3	5	41.7	0.697^{ns}
	Miscarriage	6	66.7	7	58.3	

ns: non significance

Table 2: Pre- and post-treatment clinical parameters in myoinositol plus metformin group.

Variables	Pre-treatment (n=40)	Post-treatment (n=36)	Effect size	P value
BMI (kg/m²)	27.3±1.3	26.5±1.5	1.02	0.001^{s}
Waist circumference (cm)	91.5±6.0	87.9±7.2	0.91	0.001^{s}
Hip circumference (cm)	98.8±6.8	97.2±8.0	0.44	0.011^{s}
Waist-hip ratio	0.92±0.06	0.90±0.07	0.47	0.008^{s}

s: significance

Table 3: Pre- and post-treatment clinical parameters in metformin alone group.

Variables	Pre-treatment (n=40)	Post-treatment (n=37)	Effect size	P value
BMI (kg/m²)	27.5±1.6	26.7±1.6	0.79	0.001^{s}
Waist circumference (cm)	92.2±6.5	90.9±6.6	0.53	0.002^{s}
Hip circumference (cm)	98.9±6.1	98.1±6.4	0.43	0.012^{s}
Waist–hip ratio	0.93±0.05	0.92±0.05	0.50	0.013^{s}

s: significance

Table 4: Pre- and post-treatment biochemical parameters in myoinositol plus metformin group.

Parameters	Pre-treatment (n=40)	Post-treatment (n=37)	Effect size	P value
Serum AMH (ng/ml)	12.2±3.4	9.5±3.5	1.81	0.001^{s}
Serum FSH (µIU/ml)	6.1±1.4	6.1±1.6	0.01	0.978^{ns}
Serum LH (mIU/ml)	10.0±5.0	9.0±5.2	0.66	0.001^{s}
Serum testosterone (ng/ml)	59.0±18.4	43.5±14.7	1.55	0.001^{s}
FBS (mmol/l)	5.3±0.61	5.3±0.6	0.20	0.297^{ns}
2 hours after 75 gm glucose (mmol/l)	6.7±0.8	6.4±0.7	1.00	0.001^{s}

s: significance, ns: non significance

Table 5: Pre- and post-treatment biochemical parameters in metformin alone group.

Variables	Pre-treatment (n=40)	Post-treatment (n=36*)	Effect size	P value
Serum AMH (ng/ml)	12.3±4.1	8.6±3.6	2.28	0.001[s]
Serum FSH (µIU/ml)	6.0±1.7	6.2±1.9	0.17	0.297[ns]
Serum LH (mIU/ml)	9.6±7.4	6.9±4.5	0.91	0.001[s]
Serum Testosterone (ng/ml)	60.7±14.9	40.2±11.8	2.1	0.001[s]
Fasting blood sugar (mmol/l)	5.38±1.0	5.2±0.7	0.25	0.124[ns]
2-hour glucose after 75 gm glucose (mmol/l)	6.7±1.5	6.2±1.3	1.26	0.001[s]

s: significance, ns: non significance

Table 6: Comparison of menstrual cycle regularity before and after treatment.

Cycle regularity	Group A (myoinositol + metformin) N (%)	Group B (metformin alone) N (%)	RR (95% CI)	P value
Before treatment	**(n=40)**	**(n=40)**	–	–
Regular	0 (0.0)	0 (0.0)		
Irregular	40 (100.0)	40 (100.0)		
After treatment	**(n=40)**	**(n=39)**	1.09 (0.94 to 1.25)	0.205[ns]
Regular	38 (95.0)	34 (87.2)		
Irregular	2 (5.0)	5 (12.8)		

ns: non significance

Table 7: Comparison of pregnancy rate between two groups (n=79).

Pregnancy rate	Group A (myoinositol + metformin) (n=40)	Group B (metformin alone) (n=39)	RR (95% CI)	P value
Yes	4 (10.0%)	2 (5.1%)	1.95 (0.37 to 10.04)	0.350[ns]
No	36 (90.0%)	37 (94.9%)		

ns: non significance

Table 8: Side effects among study participants (n=73).

Adverse effects	Group A (myoinositol + metformin) (n=36)	Group B (metformin alone) (n=37)	P value
GIT upset	4 (11.1%)	3 (8.1%)	0.484[ns]
Headache	1 (2.8%)	0 (0.0%)	0.493[ns]
Weight gain	0 (0.0%)	1 (2.7%)	0.507[ns]

ns: non significance

In the myoinositol plus metformin group, there was a significant reduction in clinical parameters following treatment. The mean BMI decreased from 27.3±1.3 to 26.5±1.5 kg/m^2, waist circumference from 91.5±6.0 to 87.9±7.2 cm, hip circumference from 98.8±6.8 to 97.2±8.0 cm, and waist-hip ratio from 0.92±0.06 to 0.90±0.07. All changes were statistically significant ($p<0.05$).

In the metformin alone group, significant reductions were observed in clinical parameters after treatment. The mean BMI decreased from 27.5±1.6 to 26.7±1.6 kg/m², waist circumference from 92.2±6.5 to 90.9±6.6 cm, hip circumference from 98.9±6.1 to 98.1±6.4 cm, and waist-hip ratio from 0.93±0.05 to 0.92±0.05. All reductions were statistically significant ($p<0.05$). In this study myoinositol plus metformin group is more effective than metformin alone in reducing the BMI and waist-hip ratio for over 3 months.

In the myoinositol plus metformin group, significant post-treatment reductions were observed in several biochemical parameters. Serum AMH decreased from 12.2±3.4 to 9.5±3.5 ng/ml, serum LH from 10.0±5.0 to 9.0±5.2 mIU/ml, serum testosterone from 59.0±18.4 to 43.5±14.7 ng/ml, and 2-hour post-glucose levels from 6.7±0.8 to 6.4±0.7 mmol/l. These changes were statistically significant ($p<0.05$). No significant changes were noted in serum FSH or fasting blood sugar (FBS).

In the metformin alone group, significant reductions were observed post-treatment in serum AMH (12.3±4.1 to 8.6±3.6 ng/ml), LH (9.6±7.4 to 6.9±4.5 mIU/ml), testosterone (60.7±14.9 to 40.2±11.8 ng/ml), and 2-hour post-glucose levels (6.7±1.5 to 6.2±1.3 mmol/l), all with p

values <0.05. No significant changes were observed in serum FSH or fasting blood sugar. Overall, the Myoinositol plus Metformin group showed greater efficacy than Metformin alone in reducing serum AMH, LH, and testosterone levels over the 3-month treatment period.

Before treatment, all patients in both groups had irregular menstrual cycles. After treatment, 38 patients (95.0%) in group A (myoinositol + metformin) and 34 patients (87.2%) in group B (metformin alone) achieved cycle regularity. Although group A showed a higher rate of improvement, the difference was not statistically significant (RR=1.09, 95% CI: 0.94-1.25; p=0.205).

The pregnancy rate was higher in group A (myoinositol + metformin) compared to group B (metformin alone) (10.0% versus 5.1%). However, this difference was not statistically significant (RR=1.95; 95% CI: 0.37-10.04; p=0.350).

Table 8 shows that gastrointestinal (GIT) upset occurred in 4 patients (11.1%) in group A (myoinositol + metformin) and 3 patients (8.1%) in group B (metformin alone). Headache was reported in 1 patient (2.8%) in group A but not in group B. Conversely, weight gain was observed in 1 patient (2.7%) in group B but not in group A. None of the differences between the groups were statistically significant (p>0.05).

DISCUSSION

Polycystic ovary syndrome (PCOS) is the most common endocrine disorder affecting reproductive-aged women, with anovulation and infertility being the primary clinical manifestations.[21] PCOS requires long-term management, and there is no universally accepted treatment protocol. The therapeutic course often exceeds three months, primarily aiming to regulate menstrual cycles, reduce androgen levels, and induce ovulation in patients seeking fertility.[22] Both metformin and myoinositol act as insulin sensitizers through different mechanisms. Myoinositol has emerged as a promising and safe alternative in PCOS treatment, demonstrating favorable outcomes in ovulation induction, hyperandrogenism, and metabolic parameters with minimal side effects and better patient compliance.[23] Their study also showed a significantly higher live birth rate and improved menstrual cyclicity in patients treated with a combination of metformin and myoinositol. Based on this context, our study compared the clinical and biochemical efficacy of myoinositol plus metformin versus metformin alone in infertile PCOS women with high serum AMH levels.

In the present study, the mean age of participants was comparable between groups: 26.3±4.6 years in the combination group (group A) and 26.6±5.1 years in the metformin-only group (group B), with no statistically significant difference. This finding aligns with other studies that reported similar age distributions without significant group differences.[24,25]

Most participants in both groups were cases of primary subfertility: 77.5% in group A and 70.0% in group B. Secondary infertility was present in 22.5% and 30.0% of patients in group A and B, respectively. Among those with secondary infertility, miscarriage was more common than live births in both groups, and the difference in outcomes was not statistically significant. These findings are consistent with prior research.[24]

The combination therapy showed significant reductions in body mass index (BMI), waist circumference, hip circumference, and waist-hip ratio after treatment. For example, the body mass index (BMI) showed a reduction from 27.3±1.3 to 26.5±1.5 kg/m^2, and the waist-to-hip ratio decreased from 0.92±0.06 to 0.90±0.07. These results indicate better metabolic outcomes with combination therapy. A similar trend was noted by Gade et al supporting the effectiveness of myoinositol with metformin in improving anthropometric parameters.[26]

In the metformin-alone group, there were also significant reductions in BMI and related measurements, although the degree of change was less pronounced than in the combination group. For example, the BMI declined from 27.5±1.6 to 26.7±1.6 kg/m^2. These findings suggest that while metformin alone is effective, the addition of myoinositol enhances the metabolic response. Ravn et al also reported significant BMI reduction with metformin alone.[27]

Regarding biochemical markers, Group A demonstrated significant reductions in serum AMH (12.3±4.1 to 8.6±3.6 ng/ml), LH (9.6±7.4 to 6.9±4.5 mIU/ml), testosterone (60.7±14.9 to 40.2±11.8 ng/ml), and 2-hour glucose levels (6.7±1.5 to 6.2±1.3 mmol/l) after treatment. These improvements were more notable than in the metformin-alone group. Gade et al observed similar hormonal improvements with combination therapy.[26]

In the metformin-only group, significant reductions were also observed in AMH (12.2±3.4 to 9.5±3.5 ng/ml), LH (10.0±5.0 to 9.0±5.2 mIU/ml), testosterone (59.0±18.4 to 43.5±14.7 ng/ml), and glucose (6.7±0.8 to 6.4±0.7 mmol/l). However, the extent of change was comparatively less. Studies by Chhabra et al and Foroozanfard et al confirm the effect of metformin in reducing AMH and androgen levels, although combination therapy appears to yield superior biochemical responses.[28,29]

Menstrual cycle regularity improved in both groups after treatment. All patients initially presented with irregular cycles. Following treatment, 95.0% of participants in group A and 87.2% in group B reported experiencing regular menstrual cycles. The difference was not statistically significant. However, studies have shown that myoinositol supplementation significantly enhances cycle

regularity, supporting the slight trend in favor of the combination therapy.[25]

With respect to pregnancy outcomes, the combination group showed a higher pregnancy rate (10.0%) compared to the metformin group (5.1%), though this was not statistically significant. Agrawal et al reported a markedly higher clinical pregnancy rate in the combination group (63.3%) than in the metformin-only group (33.3%), highlighting the potential reproductive benefits of combined therapy.[25]

The study had some limitations. The study was conducted with a relatively small sample size, which may limit the generalizability of the findings. Participants were recruited from a single selected center in Dhaka city, potentially affecting the external validity of the results. The study was carried out over a short duration, which may have restricted long-term outcome evaluation. Although selection bias was minimized through random allocation and allocation concealment, other potential sources of bias existed: performance bias due to the absence of blinding among participants and personnel administering the interventions. Detection bias owing to the lack of blinding during outcome assessment.

CONCLUSION

Over a three-month treatment period, the combination therapy of myoinositol and metformin yielded better results than metformin alone in reducing clinical parameters such as BMI and waist-hip ratio. Biochemical parameters, including serum AMH, serum LH, serum testosterone levels, and 2-hour post-glucose levels, were significantly decreased in the combination group after treatment. Menstrual cycle regularity significantly improved, and the pregnancy rate was higher in the combination group. Although post-treatment clinical and biochemical parameters between the two groups were not statistically significant, the effect size was greater in the combination group compared to the metformin-only group.

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