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Original Research Article

Berberine and Cinnamon-Fortified Probiotic Yogurt Mitigates Metabolic and Hormonal Disturbances in a DHT and Fructose-Induced PCOS Rat Model

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Abstract:
Introduction: Polycystic ovary syndrome (PCOS) is one of the most common endocrine and metabolic disorders affecting women of reproductive age, with global prevalence estimates ranging from 6% to 20%, depending on diagnostic criteria. It is characterized by a complex interplay of hormonal and metabolic disturbances, most notably hyperandrogenism, chronic anovulation, insulin resistance, and ovarian dysfunction. ***Material And Methods***: The estrous cycle phases were examined during the study by analyzing the relative percentages of epithelial, leukocytes, and cornified cells. All tests complied with the Institutional Animal Ethics Committee (IAEC) guidelines, as approved with approval number IAEC 2242/PO/Re/S/23/CCSEA. ***Results***: The primary endpoint of this study was at least 10% weight loss with <40% food and water intake. At the end of the experiments, the secondary endpoint was assessed by checking for at least a 20% weight loss without food or water intake, accompanied by repeated or prolonged convulsions or ataxia. At termination, rats were anesthetized, blood was collected via retro-orbital bleeding, vaginal smears were obtained, and parametrial fat pads were dissected and weighed. ***Conclusion***: The synergistic effects observed in combination therapy highlight its potential as a scalable intervention targeting multiple aspects of PCOS pathophysiology

Keywords: Polycystic ovary syndrome (PCOS), Insulin resistance, Estrous cycle regulation, Combination therapy, Metabolic dysfunction

INTRODUCTION

Polycystic ovary syndrome (PCOS) is one of the most common endocrine and metabolic disorders affecting women of reproductive age, with global prevalence estimates ranging from 6% to 20%, depending on diagnostic criteria. It is characterized by a complex interplay of hormonal and metabolic disturbances, most notably hyperandrogenism, chronic anovulation, insulin resistance, and ovarian dysfunction. These clinical hallmarks are often accompanied by menstrual irregularities, infertility, and hirsutism, significantly impacting quality of life. Beyond reproductive concerns, PCOS poses substantial long-term health risks, including metabolic syndrome, type 2 diabetes mellitus, obesity, dyslipidemia, and increased cardiovascular disease susceptibility. The etiology of PCOS is multifactorial, involving genetic predisposition, hormonal dysregulation, insulin signaling defects, chronic low-grade inflammation, and gut microbiota imbalances [1].

Current management strategies focus primarily on symptom reduction and risk mitigation through lifestyle modifications such as diet and exercise, pharmacological interventions aimed at restoring hormonal balance, and hormonal contraceptives to regulate menstrual cycles. Insulin-sensitizing agents

like metformin are also widely prescribed. However, these approaches often provide partial relief, may produce side effects, and do not address all underlying pathophysiological mechanisms of the condition. Consequently, there is growing interest in complementary and integrative therapies, particularly those utilizing functional foods and bioactive phytochemicals to enhance metabolic and hormonal regulation in a safe, non-invasive manner.

Among natural agents, cinnamon (*Cinnamomum zeylanicum*) has been recognized for its antioxidant, anti-inflammatory, and insulin-sensitizing properties. It has shown promise in lowering fasting blood glucose, improving lipid profiles, and ameliorating menstrual irregularities in women with PCOS. Similarly, berberine, an isoquinoline alkaloid extracted from plants such as *Berberis aquifolium*, exhibits multifaceted pharmacological actions—improving insulin sensitivity, modulating gut microbiota, reducing inflammation, and exerting lipid-lowering effects. Notably, berberine has demonstrated efficacy comparable to metformin in improving metabolic and reproductive outcomes in PCOS models [2, 3].

Probiotic-enriched yogurt offers another promising dietary intervention. Probiotics can restore gut microbial balance, reduce systemic inflammation, and influence metabolic signaling, which are all relevant to PCOS pathophysiology. The combination of probiotics with polyphenol-rich and alkaloid-based compounds such as cinnamon and berberine could potentially provide a synergistic effect, simultaneously targeting insulin resistance, hyperandrogenism, inflammation, oxidative stress, and gut dysbiosis [4].

Despite the individual benefits reported for these bioactive agents, scientific investigation into their combined use within a functional food matrix is scarce. Therefore, the present study aimed to develop a novel functional yogurt formulation fortified with berberine, cinnamon, and probiotics for PCOS management. The therapeutic potential was assessed using a DHT + Fructose-induced PCOS rat model (83 µg/day/kg/BW and fructose 20% in H_2O) [5]. In this study, we evaluated the effect of present formulations on hormonal regulation, insulin sensitivity, and ovarian morphology. The DHT + Fructose-induced PCOS model was selected for its reliability in replicating PCOS phenotypes, including ovarian cysts and endocrine disruptions. By combining phyto-therapeutic compounds with probiotics, this strategy targets multiple pathological pathways simultaneously, offering a non-pharmacological strategy to mitigate PCOS symptoms. This dual focus bridges functional food science with endocrine research, addressing a critical gap in holistic PCOS interventions while exploring the therapeutic potential of dietary synergism.

MATERIAL AND METHODS

2.1. Animal Care

8-10-week-old cyclic adult female Wistar rats weighing 200 ± 20 g were taken and acclimatized for one week under specific pathogen-free conditions with an automatic *12h*:*12h light*/*dark* cycle and *ad libitum* access to water and food. The estrous cycle phases were examined during the study by analyzing the relative percentages of epithelial, leukocytes, and cornified cells. All tests complied with the Institutional Animal Ethics Committee (IAEC) guidelines, as approved with approval number IAEC2242/PO/Re/S/23/CCSEA.

2.2. Experimental Design

Seventy-two animals were randomly assigned to 12 groups to determine the pharmacological effects of our formulation (**Table 1**). Each group contains 6 animals. All doses were administered orally daily for seven weeks, and vaginal smears were examined daily. Berberine [6], cinnamon [7] and metformin doses [8, 9] were selected based on earlier publications.

RESULTS

Table 1: Experimental groups for the DHT + Fructose-induced PCOS model study

Group	Dose (mg/kg/day)	Route of Drug Administration
Control (CMC)	0.5ml/kg	P. O
Disease Control (DHT + Fructose)	83 µg/day/kg/BW, fructose 20% in H_2O	S.C P. O
Berberine	200	P. O
Low Dose (Berberine Fortified Yogurt)	200	
High Dose (Berberine Fortified Yogurt)	400	
Cinnamon	200	
Low Dose (Cinnamon Fortified Yogurt)	200	
High Dose (Cinnamon Fortified Yogurt)	400	

Standard Fortified Yogurt	200	
Metformin	500	
Low Dose (Berberine + Cinnamon Fortified Yogurt)	200	
High Dose (Berberine + Cinnamon Fortified Yogurt)	400	

DHT and Fructose-induced PCOS in Rat

Rats were subcutaneously injected daily with DHT (83 µg/day/kg BW; *S.C*) dissolved in 0.5 ml CMC (Sigma-Aldrich, USA) for up to 13 Weeks. The dose of DHT was selected based on a previous study [5]. Rats in the control group were given CMC per oral without DHT. The primary endpoint of this study was at least 10% weight loss with <40% food and water intake. At the end of the experiments, the secondary endpoint was assessed by checking for at least a 20% weight loss without food or water intake, accompanied by repeated or prolonged convulsions or ataxia. At termination, rats were anesthetized, blood was collected *via* retro-orbital bleeding, vaginal smears were obtained, and parametrial fat pads were dissected and weighed. The ovaries were dissected and fixed in 4% PFA at 4°C overnight before processing. ELISA measured serum insulin, LH, testosterone, and 17-β estradiol levels. Vaginal cytology was performed to determine the stage of the estrous cycle. Paraffin-embedded ovaries were sectioned at 10 µm and stained with hematoxylin and eosin [10].

2.3. Hormonal analysis

For hormonal analysis, blood samples were collected via cardiac puncture and centrifuged at low temperatures to separate serum, and then analyzed for insulin, LH (luteinizing hormone), estrogen, and testosterone, measured by ELISA kits according to the manufacturer's instructions [11].

2.4. Assay for lipid profile

The lipid profile, including total lipid, total cholesterol (TC), HDL (high-density lipoprotein), and triglycerides (TGs), was quantified using standard colorimetric kits (Accurex Pvt. Ltd.) [12].

2.5. Histological Examination of Ovary and Uterus

The histological examination was carried out by using hematoxylin-eosin (H&E) dye as described [10].

2.6. Statistical analysis

Results are expressed as a Mean±SD. For multiple comparisons, a one-way analysis of variance (ANOVA) followed by post hoc analysis was performed using Tukey's test. $p<0.05$ was considered statistically significant.

RESULT AND DISCUSSION

3.1. Effect on Body Weight Gain and Lee's Index

Induction of PCOS using the DHT and fructose model resulted in a marked and statistically significant increase in body weight (BW) over the experimental period. Specifically, animals in the DHT + fructose group exhibited an increase in BW from an initial mean of 200.5 ± 4.9 g to 240.7 ± 5.8 g, representing a gain substantially higher than that of the healthy control group (##$p < 0.01$). This pronounced weight gain is consistent with previous reports indicating that androgen excess combined with high-fructose intake promotes adiposity, metabolic dysfunction, and increased body mass in rodent models of PCOS.

Administration of both the standard treatment and the experimental probiotic yogurt formulations yielded a moderating effect on BW progression (**$p < 0.01$ versus PCOS group). In particular, the groups receiving berberine- and cinnamon-fortified yogurt demonstrated notably lower BW gain compared to the untreated PCOS model group. Over the intervention period, the low-dose formulation group recorded an average gain of 21.4 ± 2.2 g (10.7% ± 1.1), while the high-dose group showed a gain of 23.2 ± 2.3 g (11.6% ± 1.1). Both values were significantly lower than those observed in the DHT + fructose model group ($p < 0.05$), indicating that the functional yogurt interventions attenuated the excessive BW increase typically associated with this PCOS model.

These findings suggest that berberine and cinnamon, when delivered in a probiotic yogurt matrix, may help mitigate weight gain linked to androgen-induced and diet-induced metabolic imbalances. The effect could be attributed to improved insulin sensitivity, modulation of lipid metabolism, and potential anti-inflammatory actions of the bioactive compounds, supported by the probiotic's role in enhancing metabolic homeostasis. Full results are presented in **Table 2.**

Table 2: Average Body Weights and Percentage Weight Gain (DHT + Fructose-Induced PCOS Model)

Group	Initial Weight (g)	Final Weight (g)	Weight Gain (g)	Percentage Increase (%)
Control (CMC)	200.2 ± 5.1	218.8 ± 5.5	18.6 ± 2.0	9.3% ± 1.0
Disease Control (DHT + Fructose)	200.5 ± 4.9	240.7 ± 5.8	40.2 ± 2.5	20.1% ± 1.2##
Berberine	200.8 ± 5.2	223.6 ± 5.3	22.8 ± 2.1	11.4% ± 1.0**
Low Dose (Berberine Fortified Yogurt)	200.1 ± 5.0	220.3 ± 5.4	20.2 ± 2.0	10.1% ± 1.0**
High Dose (Berberine Fortified Yogurt)	200.3 ± 5.3	225.0 ± 5.5	24.7 ± 2.2	12.3% ± 1.1**
Cinnamon	200.6 ± 5.4	224.1 ± 5.6	23.5 ± 2.3	11.8% ± 1.1**
Low Dose (Cinnamon Fortified Yogurt)	200.7 ± 5.2	221.0 ± 5.5	20.3 ± 2.3	10.1% ± 1.0**
High Dose (Cinnamon Fortified Yogurt)	200.4 ± 5.3	224.7 ± 5.6	24.3 ± 2.4	12.1% ± 1.1**
Standard Fortified Yogurt	200.9 ± 5.4	222.0 ± 5.3	21.1 ± 2.1	10.6% ± 1.0**
Metformin	200.7 ± 5.5	224.0 ± 5.4	23.3 ± 2.3	11.6% ± 1.1**
Low Dose (Berberine + Cinnamon Yogurt)	200.5 ± 5.1	221.9 ± 5.4	21.4 ± 2.2	10.7% ± 1.1**
High Dose (Berberine + Cinnamon Yogurt)	200.3 ± 5.1	223.5 ± 5.6	23.2 ± 2.3	11.6% ± 1.1**

Values are mean ± SD; ##p < 0.01 vs. Control group, **p < 0.05 vs. Disease Control (DHT + Fructose) group.

Lee's index values in the DHT + Fructose model further validated the anti-adiposity effects of the treatments [13]. Like BW, the DHT + Fructose model treatment groups exhibited significantly lower Lee's index values than the control group. The Berberine group showed Lee's index of 296.5 ± 2.5. In contrast, the Berberine + Cinnamon yogurt groups displayed indices of 296.4 ± 2.5 (low dose) and 297.1 ± 2.5 (high dose), significantly lower than the DHT + Fructose group ($p < 0.05$). These findings highlight the efficacy of these treatments in managing adiposity, with Lee's index values approaching those of the control group **(Figure 1)**.

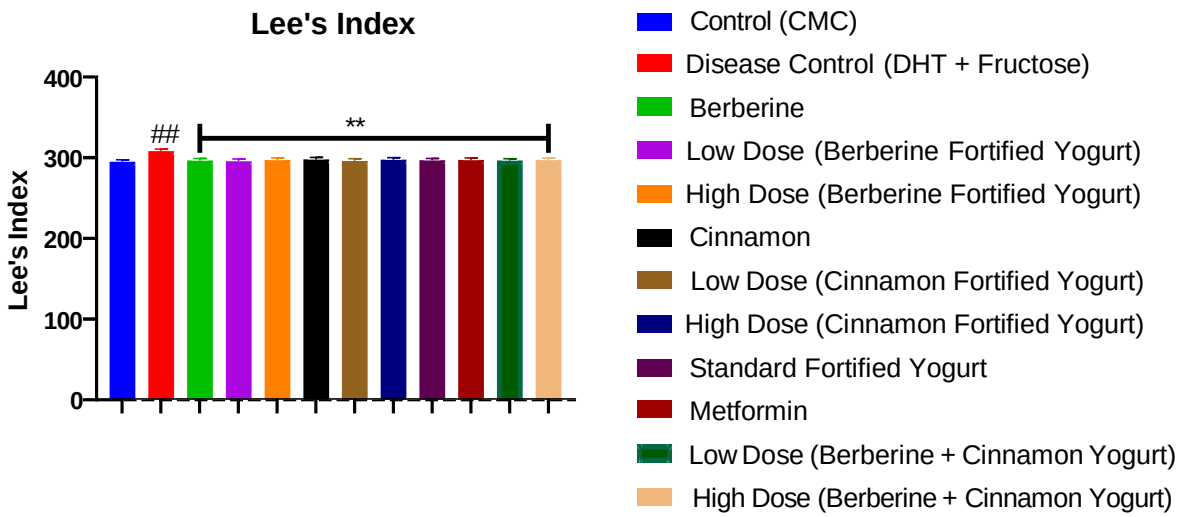

Figure 1: Lee's Index Values (DHT + Fructose-Induced PCOS Model). Values are mean ± SD; ##p < 0.01 vs. Control, **p < 0.05 vs. Disease Control (DHT + Fructose) group.

3.2. Effect on Hormonal Levels

Induction of PCOS using the DHT + fructose model produced a characteristic pattern of endocrine disruption consistent with the syndrome's pathophysiology. Animals in the model group exhibited a marked and statistically significant elevation in circulating insulin, luteinizing hormone (LH), and testosterone levels compared to the healthy control group, indicating the development of hyperinsulinemia, hyperandrogenism, and HPG-axis dysregulation ($p < 0.01$). Conversely, estrogen concentrations and the progesterone-to-estradiol (P/E) ratio were significantly reduced, further confirming impaired ovarian function and anovulatory status in the induced rats. This hormonal signature validates the successful establishment of the PCOS phenotype in the experimental model.

Therapeutic intervention with berberine alone resulted in a substantial improvement in the hormonal profile, reversing many of the endocrine alterations toward normal ranges. Importantly, treatment with berberine- and cinnamon-fortified probiotic yogurt produced an even greater restorative effect, with improvements observed in a dose-dependent manner. Both low- and high-dose formulations significantly reduced elevated insulin, LH, and testosterone levels while simultaneously enhancing estrogen concentrations and improving the P/E ratio ($p < 0.05$ vs. model group). The synergistic combination of berberine and cinnamon within the probiotic yogurt matrix exhibited superior efficacy compared with administration of either compound individually, suggesting enhanced bioactivity and systemic impact when combined. Among all intervention groups, the high-dose fortified yogurt formulation demonstrated the most pronounced normalization of hormonal parameters, reflecting its multi-targeted influence on insulin signaling, androgen excess, and ovarian steroidogenesis. These findings are comprehensively illustrated in **Figure 2**, highlighting the robustness of the hormonal restoration achieved with the combined functional food-based approach.

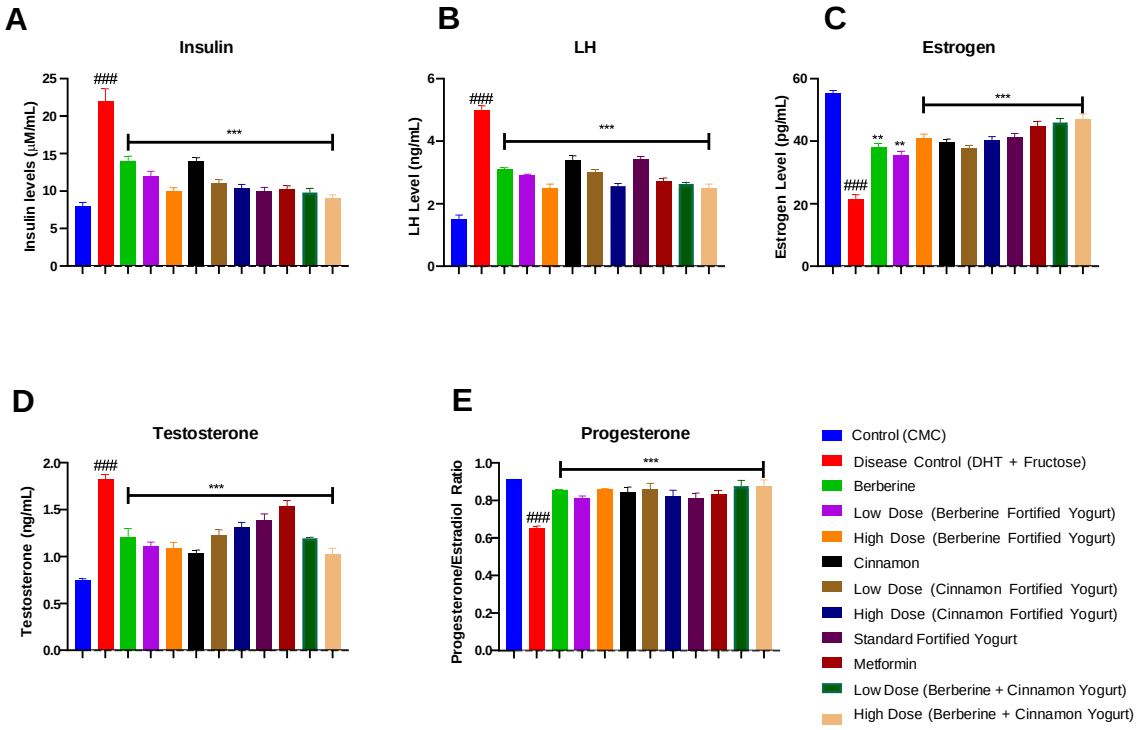

Figure 2: Effect of yogurt formulations on hormonal Levels in DHT + fructose-Induced PCOS Model. (A) Insulin, (B) LH, (C) Estrogen, (D) Testosterone, and (E) Progesterone/Estradiol ratio. Data are expressed in Mean ± SD (n=6). ###$p < 0.001$ vs. Control, **$p < 0.05$, ***$p < 0.001$ vs. Disease Control (DHT + Fructose) group.

Effect on lipid profiles

In the disease control group receiving DHT + fructose, there was a clear disruption in lipid metabolism, as evidenced by a significant elevation in total cholesterol (TC) and triglyceride (TG) concentrations (###$p < 0.001$ vs. healthy control) along with a concomitant reduction in high-density lipoprotein (HDL) levels (###$p < 0.001$). This dyslipidemic pattern is consistent with metabolic disturbances commonly reported in PCOS, where androgen excess and insulin resistance contribute to altered lipid handling and increased cardiovascular risk.

Intervention with the berberine- and cinnamon-fortified probiotic yogurt formulations resulted in a marked and dose-dependent normalization of these lipid parameters. Both low- and high-dose fortified yogurt groups exhibited

significant reductions in TC and TG, coupled with substantial increases in HDL levels when compared to the disease control group (***p < 0.001). The extent of lipid correction achieved with the fortified yogurt surpassed that observed in animals receiving berberine or cinnamon alone, indicating synergistic lipid-modulating effects when these phytoactive agents were combined within a probiotic matrix.

Notably, the high-dose berberine + cinnamon yogurt formulation showed the most pronounced improvements across all measured lipid parameters, bringing values closer to those of the healthy control group. Such superior efficacy may be attributed to multiple, complementary mechanisms — berberine's known ability to modulate lipid metabolism via AMPK activation, cinnamon's role in improving insulin sensitivity and lipid clearance, and probiotics' beneficial influence on gut microbiota and bile acid metabolism.

Collectively, these findings suggest that the combined functional yogurt intervention not only ameliorates hyperlipidemia associated with DHT + fructose-induced PCOS but also offers a multi-targeted dietary strategy for improving metabolic health. Detailed comparative values for each group are presented in **Figure 3.**

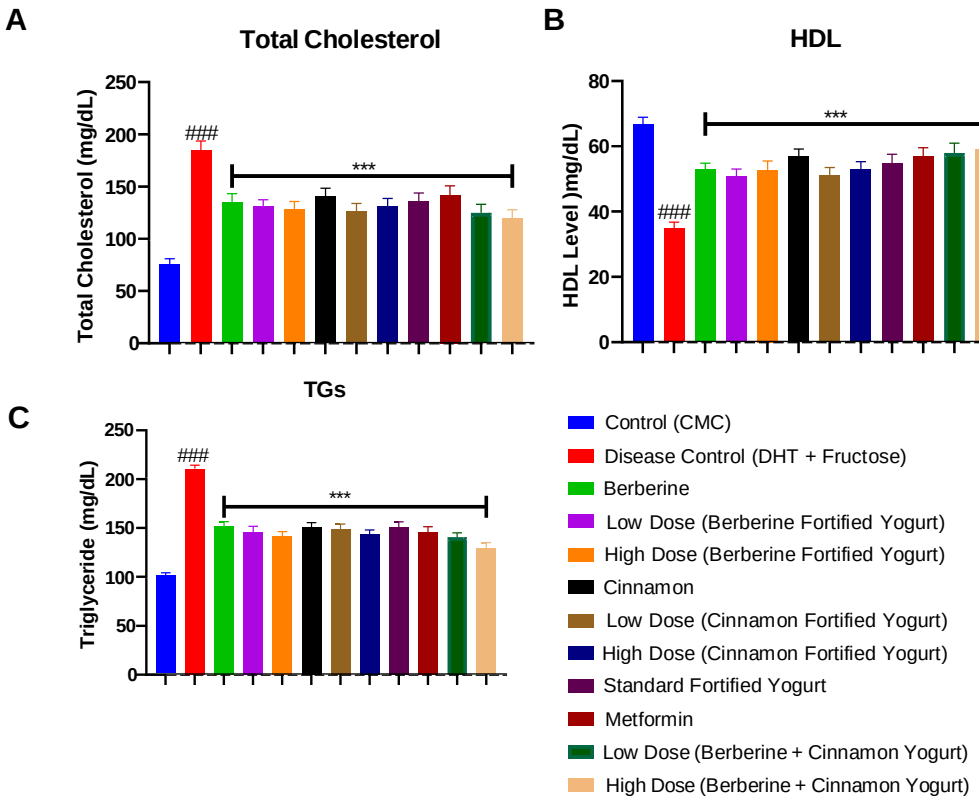

Figure 3: Effect of yogurt formulations on lipid profile in DHT + fructose-induced PCOS Model. (A) Total cholesterol, (B) HDL, and (C) Triglyceride. Data are expressed in Mean ± SD (n=6). ###p < 0.001 vs. Control, ***p < 0.001 vs. Disease Control (DHT + Fructose) group.

3.4. Effect on Inflammation

Our study demonstrated a significant increase in inflammatory cytokines TNF-α and IL-6 in the Disease Control group (DHT + Fructose) compared to the Control group (CMC). Both TNF-α and IL-6 levels are substantially higher in the Disease Control group (###p<0.001), highlighting the inflammatory response induced by DHT and fructose administration. However, all treated groups—including those given standard berberine, cinnamon, and various forms of fortified yogurt (berberine, cinnamon, or both, at low or high doses), as well as metformin—exhibited a marked reduction in both TNF-α and IL-6 levels relative to the Disease Control group (***p<0.001). These reductions brought the cytokine levels in treated groups much closer to the Control group, showcasing the effectiveness of these interventions in mitigating the inflammatory response. Collectively, these findings suggest that berberine, cinnamon, their combinations, and metformin all have strong potential to counteract inflammation induced by DHT and fructose, as evidenced by normalized TNF-α and IL-6 levels **(Figure 4).**

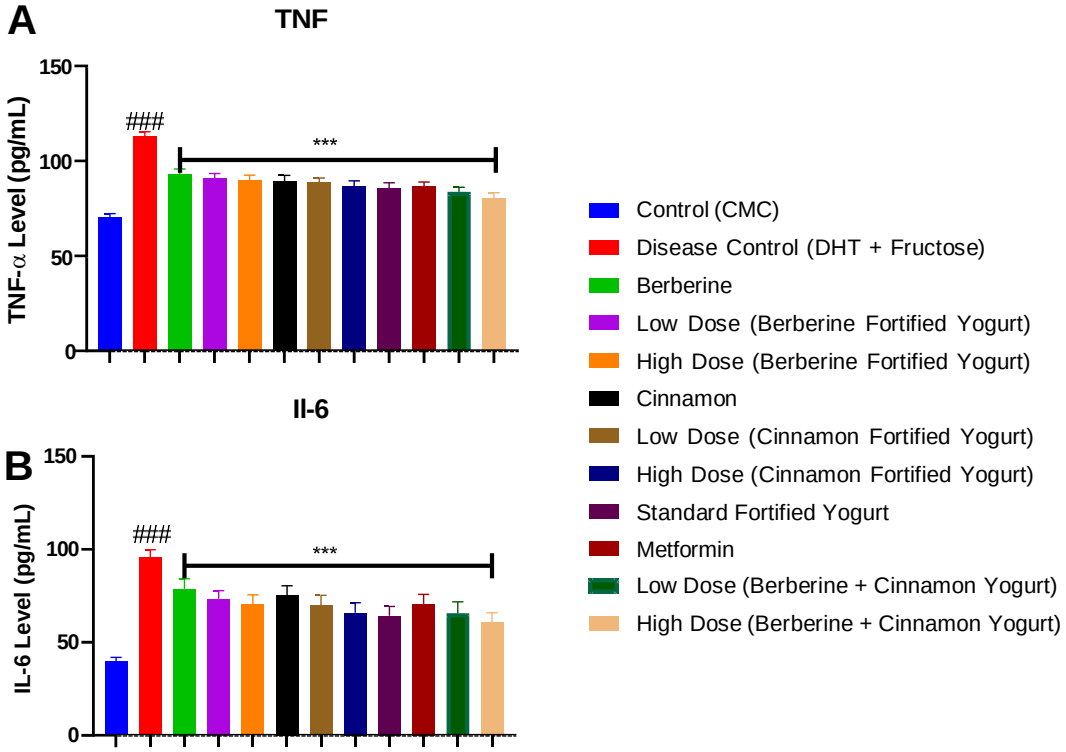

Figure 4: Effect of yogurt formulations on cytokine profile in DHT + fructose-induced PCOS Model. Level of (A) TNF-α and (B) IL-6. Data are expressed in Mean ± SD (n=6). ###p < 0.001 vs. Control, ***p < 0.001 vs. Disease Control (DHT + Fructose) group.

3.5. Effect on Oxidative Stress

In the disease control group (DHT + Fructose, red bars), both MDA and nitrite levels are substantially elevated, while SOD activity is markedly reduced compared to the Control (###p<0.001). All treatment groups significantly reduced MDA and nitrite levels and increased SOD activity (***p<0.001). Interestingly, as observed in earlier results, the high-dose berberine + cinnamon yogurt formulation showed the most pronounced improvements across all oxidative stress parameters **(Figure 5)**.

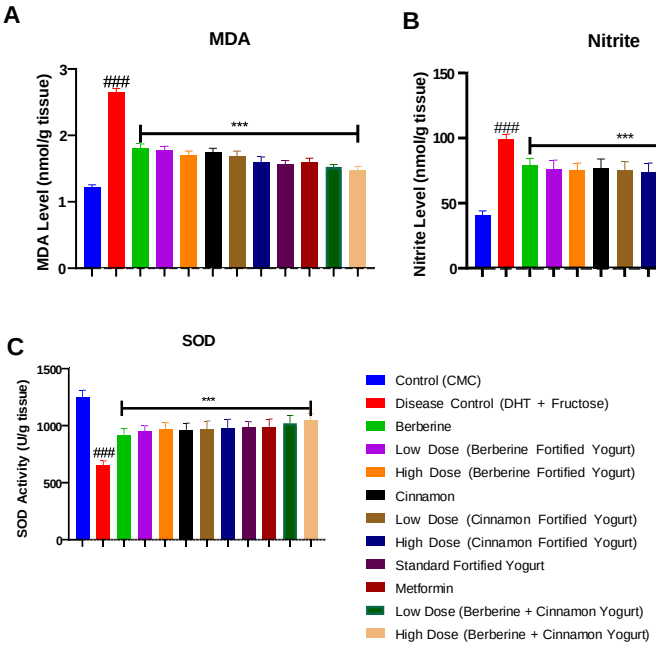

Figure 5: Effect of yogurt formulations on oxidative stress in DHT + fructose-induced PCOS Model. Level of (A) MDA, (B) Nitrite, and (C) SOD. Data are expressed in Mean ± SD (n=6). ###p < 0.001 vs. Control, ***p < 0.001 vs. Disease Control (DHT + Fructose) group.

3.6. Histology

The Control group (CMC) displayed a normal uterine structure, featuring a healthy endometrium, well-formed glands, and proper vascularization. In contrast, the Disease Control group exhibited severe pathological changes that are typical of PCOS-related abnormalities. Treatment with standard berberine led to a moderate reduction in hyperplasia, partial restoration of vascular structures, and mild fibrosis. When berberine was delivered through fortified yogurt at a low dose, there was moderate efficacy, while a higher dose achieved near-complete restoration of uterine histology.

Standard cinnamon treatment resulted in moderate improvements in vascular and glandular structures, though some residual fibrosis persisted. Low-dose cinnamon-fortified yogurt produced moderate recovery with mild remaining fibrosis, whereas the high-dose group demonstrated improvements, including normalized endometrium, glands, and vascularization. The standard fortified yogurt group showed limited effectiveness and only minimal structural improvement in the uterus. Metformin treatment significantly reduced hyperplasia, with just mild residual fibrosis. Combining berberine and cinnamon in fortified yogurt at a low dose produced synergistic effects, while a higher dose completely restored uterine histology, making this combination the most effective treatment among all groups. Representative images are shown in **Figure 6.**

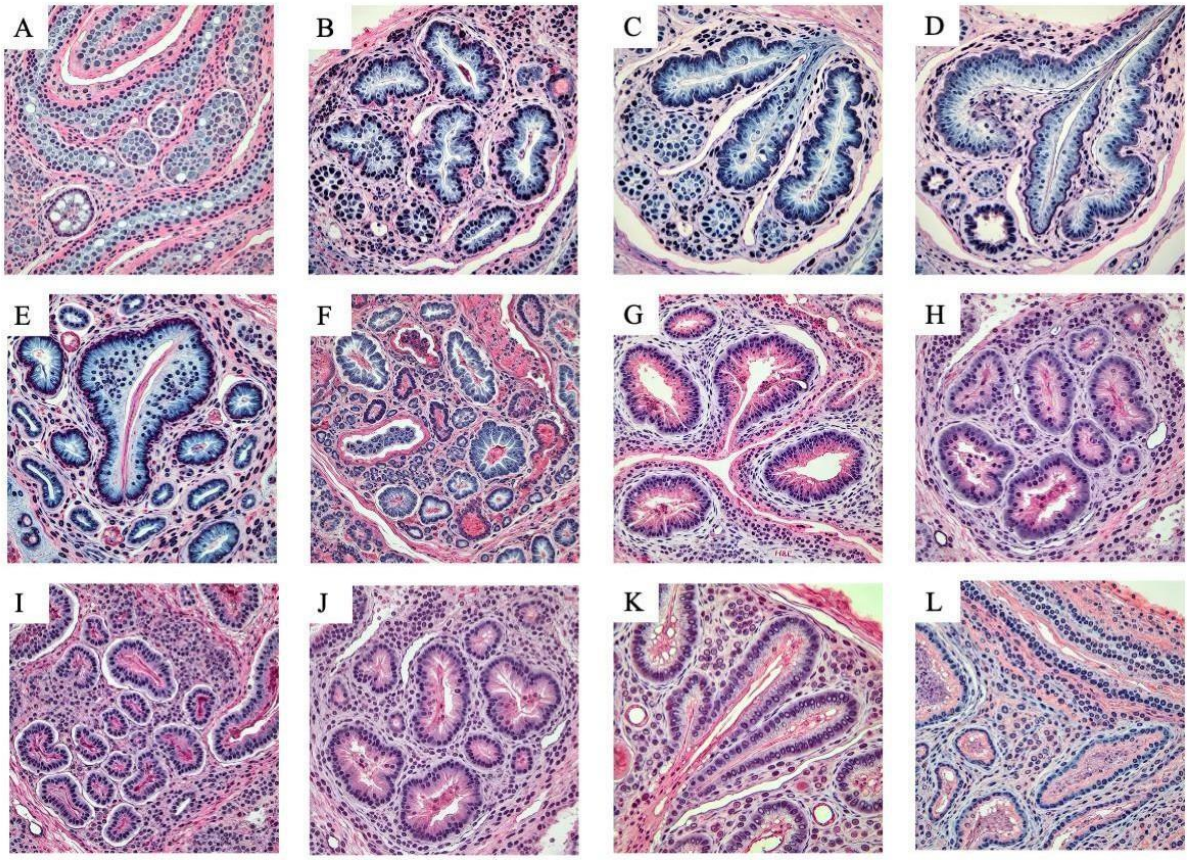

Figure 6: Histological Assessment of Uterine Tissue Across Treatment Groups DHT + Fructose-Induced PCOS Model. Histological sections of uterine tissues stained with hematoxylin and eosin (H&E) illustrate the morphological changes across experimental groups.

DISCUSSION

This study presents a novel approach to managing polycystic ovary syndrome (PCOS) by investigating the therapeutic potential of probiotic-enriched yogurt fortified with berberine and cinnamon. The research addresses a critical gap in the current understanding of functional foods tailored for PCOS management, merging phytotherapy with food science to offer a non-pharmacological intervention that targets the multifaceted pathophysiology of the disorder. Using a

DHT + fructose-induced PCOS model in Wistar rats, the study provides compelling evidence of the therapeutic potential of this innovative formulation.

The results reveal that the dose-dependent efficacy of berberine- and cinnamon-fortified yogurt improves uterine histology, hormonal balance, and metabolic parameters. Higher doses of berberine-fortified yogurt and cinnamon-fortified yogurt demonstrated significant reductions in pathological changes, including endometrial hyperplasia, stromal fibrosis, and inflammation. These findings are consistent with previous studies highlighting berberine's anti-inflammatory and insulin-sensitizing properties, which are mediated through AMPK activation and androgen suppression [14]. Cinnamon's polyphenolic compounds, known for enhancing insulin sensitivity and modulating lipid profiles, further improved vascularization and glandular health. Notably, the combination therapy exhibited synergistic effects, with the higher dose achieving complete restoration of uterine histology [15]. This underscores the therapeutic advantage of combining bioactive compounds to target multiple pathways involved in PCOS.

Chronic administration of DHT and fructose led to significant increases in the pro-inflammatory cytokines TNF-α and IL-6, as well as heightened oxidative stress, reflected by increased MDA and nitrite levels and reduced SOD activity. These findings are in line with current evidence that PCOS induces low-grade chronic inflammation, contributing to impaired insulin sensitivity and metabolic dysfunction through elevated TNF-α and IL-6, particularly in adipose tissue [16]. Treatment with berberine, cinnamon, their combinations in fortified yogurt, and metformin effectively restored the cytokine and oxidative stress parameters, significantly reducing TNF-α, IL-6, MDA, and nitrite, while increasing SOD activity to near-normal levels. These therapeutic effects suggest that the interventions possess anti-inflammatory and antioxidant properties, helping to counteract the systemic inflammation and oxidative imbalance characteristic of PCOS models

Using yogurt as a delivery matrix for these bioactive compounds adds an innovative dimension to this study. Yogurt serves not only as a palatable food product but also as a functional vehicle that enhances the bioavailability and efficacy of berberine and cinnamon. Probiotics such as ABY-1 and P. jensenii 702 likely played a crucial role in modulating gut microbiota, reducing systemic inflammation, and supporting hormonal balance [17]. Histological analysis further confirmed the efficacy of the fortified yogurt formulations in reversing PCOS-associated uterine abnormalities. The combination therapy resolved hyperplasia, fibrosis, and inflammation, highlighting its superiority over monotherapies such as metformin. These findings suggest that functional foods enriched with natural bioactives can be effective alternatives or adjuncts to conventional pharmacological treatments for PCOS. By addressing underlying pathophysiological mechanisms rather than merely managing symptoms, this approach offers a holistic solution to PCOS management.

The study also highlights the potential mechanisms underlying the observed therapeutic effects. Berberine's ability to regulate hormonal levels and improve insulin sensitivity may have reduced androgen levels and restored ovarian cyclicity. Cinnamon's role in enhancing antioxidant capacity and modulating lipid metabolism likely complements berberine's actions, improving reproductive tissue health. The probiotics in the yogurt matrix may have further amplified these effects by promoting gut health and reducing systemic inflammation through short-chain fatty acid production.

Despite its promising findings, this study has limitations that warrant further investigation. The preclinical nature of the research necessitates validation through human trials to establish translational relevance. Long-term studies are also required to assess the sustainability of therapeutic effects and potential impacts on gut microbiota composition. Additionally, personalized nutrition frameworks should be explored to optimize dose-dependent responses based on individual metabolic profiles.

CONCLUSION

In conclusion, this study introduces a groundbreaking functional food approach for PCOS management by combining berberine, cinnamon, and probiotics in a yogurt matrix. The synergistic effects observed in combination therapy highlight its potential as a scalable intervention targeting multiple aspects of PCOS pathophysiology. These findings pave the way for future investigations into personalized nutrition strategies that leverage functional foods for reproductive health. By bridging phytotherapy with food science, this research addresses an unmet need for holistic PCOS treatments that are both effective and accessible. This innovative approach holds significant promise not only for improving reproductive health outcomes but also for enhancing overall metabolic well-being in women with PCOS. As interest in non-pharmacological interventions continues to grow, functional foods such as fortified yogurt represent a sustainable solution that aligns with modern dietary preferences while addressing complex health challenges like PCOS.

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Conflict of Interest

Authors declare no conflict of interest

CRediT Statement

Priyanka Kashyap contributed to the data collection, formal analysis, draft writing, review, and editing conceptualization and design of the study. Vir Vikram did data validation, supervision, and editing. All authors read and approved the final manuscript. The authors further declare that all data were generated in-house.

Ethical approval

The Institutional Animal Ethics Committee (IAEC) approved the experiment with the protocol numbers (2242/PO/Re/S/23/CCSEA). It complies with the ARRIVE (Animal Research: Reporting of In Vivo Experiments) guidelines. All studies were carried out by the U.K. Animals (Scientific Procedures) Act, 1986, and associated guidelines, EU Directive 2010/63/EU for animal experiments, or the National Institutes of Health guide for the care and use of Laboratory animals (NIH Publications No. 8023, revised 1978).

REFERENCES

1. S. Singh, N. Pal, S. Shubham, D.K. Sarma, V. Verma, F. Marotta, M. Kumar, Polycystic Ovary Syndrome: Etiology, Current Management, and Future Therapeutics, Journal of clinical medicine 12(4) (2023).
2. L. Dou, Y. Zheng, L. Li, X. Gui, Y. Chen, M. Yu, Y. Guo, The effect of cinnamon on polycystic ovary syndrome in a mouse model, Reproductive biology and endocrinology : RB&E 16(1) (2018) 99.
3. O.M. Ionescu, F. Frincu, A. Mehedintu, M. Plotogea, M. Cirstoiu, A. Petca, V. Varlas, C. Mehedintu, Berberine-A Promising Therapeutic Approach to Polycystic Ovary Syndrome in Infertile/Pregnant Women, Life (Basel, Switzerland) 13(1) (2023).
4. G.N. Saleem, R. Gu, H. Qu, G. Bahar Khaskheli, I. Rashid Rajput, M. Qasim, X. Chen, Therapeutic potential of popular fermented dairy products and its benefits on human health, Frontiers in nutrition 11 (2024) 1328620.
5. G.S. Kumar, P. Tirgar, M. Dalal, Development and evaluation of novel rodent model of PCOS mimicking clinical phenotype in human disease, Middle East Fertility Society Journal 27(1) (2022) 25.
6. J. Yu, C. Ding, Z. Hua, X. Jiang, C. Wang, Protective effects of berberine in a rat model of polycystic ovary syndrome mediated via the PI3K/AKT pathway, The journal of obstetrics and gynaecology research 47(5) (2021) 1789-1803.
7. Maleki, A.H. Faghfouri, F.P.F. Tabrizi, J. Moludi, S. Saleh-Ghadimi, H. Jafari-Vayghan, S.A. Qaisar, Mechanistic and therapeutic insight into the effects of cinnamon in polycystic ovary syndrome: a systematic review, Journal of ovarian research 14(1) (2021) 130.
8. Y. Zhang, M. Hu, F. Meng, X. Sun, H. Xu, J. Zhang, P. Cui, N. Morina, X. Li, W. Li, X.K. Wu, M. Brännström, R. Shao, H. Billig, Metformin Ameliorates Uterine Defects in a Rat Model of Polycystic Ovary Syndrome, EBioMedicine 18 (2017) 157-170.
9. B. Xu, W. Dai, L. Liu, H. Han, J. Zhang, X. Du, X. Pei, X. Fu, Metformin ameliorates polycystic ovary syndrome in a rat model by decreasing excessive autophagy in ovarian granulosa cells via the PI3K/AKT/mTOR pathway, Endocrine Journal 69(7) (2022) 863-875.
10. P. Chansela, N. Goto-Inoue, N. Zaima, T. Hayasaka, M. Sroyraya, N. Kornthong, A. Engsusophon, M. Tamtin, C. Chaisri, P. Sobhon, M. Setou, Composition and Localization of Lipids in Penaeus merguiensis Ovaries during the Ovarian Maturation Cycle as Revealed by Imaging Mass Spectrometry, PloS one 7 (2012) e33154.
11. F. Fazel Torshizi, M. Chamani, H.R. Khodaei, A.A. Sadeghi, S.H. Hejazi, R. Majidzadeh Heravi, Therapeutic effects of organic zinc on reproductive hormones, insulin resistance and mTOR expression, as a novel component, in a rat model of Polycystic ovary syndrome, Iranian journal of basic medical sciences 23(1) (2020) 36-45.
12. C. Hubscher, D. Brooks, J. Johnson, A quantitative method for assessing stages of rat estrous cycle, Biotechnic & histochemistry: official publication of the Biological Stain Commission 80 (2005) 79-87.
13. M.A.J. van Oeteren, N. Simons, P. Simons, M.P.H. van de Waarenburg, M.E. Kooi, E.J.M. Feskens, E. van der Ploeg, M.D.G. Van den Eynde, A. Houben, C.G. Schalkwijk, M. Brouwers, Fructose restriction has beneficial effects on adipose tissue distribution but not on serum adipokine levels: Post-hoc analysis of a double-blind randomized controlled trial, Clinical obesity 15(1) (2025) e12714.
14. R. Mariangela, A. Riva, G. Petrangolini, P. Allegrini, A. Giacosa, T. Fazia, L. Bernardinelli, C. Gasparri, G. Peroni, and S.Perna, Berberine phospholipid is an effective insulin sensitizer and improves

metabolic and hormonal disorders in women with polycystic ovary syndrome: A one-group pretest–post-test explanatory study. Nutrients 13, 10 (2021): 3665.

15. Q. Bolin, K. S. Panickar, and R. A. Anderson, Cinnamon: potential role in the prevention of insulin resistance, metabolic syndrome, and type 2 diabetes. Journal of diabetes science and technology 4, (2010): 685-693.
16. H. Deng, Y. Chen, J. Xing, N. Zhang, and L. Xu, Systematic low-grade chronic inflammation and intrinsic mechanisms in polycystic ovary syndrome. Front Immunol 15 (2024) 1470283.
17. T. Sepide, S.Z. Motlagh, Y. Jalilpiran, N. Payandeh, S. Ansari, H. Mohammadi, K. Djafarian et al. The effects of pro-, pre-, and synbiotics supplementation on polycystic ovary syndrome: an umbrella review of meta-analyses of randomized controlled trials. Frontiers in Nutrition 10 (2023): 1178842.

