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Research Article

Elucidating the *Vrushya* (Anti-Infertile) Role of *yashtimadhu* (*Glycyrrhiza glabra*) w.s.r.t. PMOS With Computational Pharmacology and Molecular Docking

Gunjan Agrawal*, Manjiri Palnitkar Bhende, Supriya Singh, Bharti Kapgate

Department of Swasthavritta, Government Ayurveda College, Nagpur, Maharashtra, INDIA.

ABSTRACT

Background: Previously known as Polycystic Ovarian Syndrome (PCOS), Polyendocrine Metabolic Ovarian Syndrome (PMOS) is a multifactorial illness that involves endocrine, metabolic, inflammatory, and reproductive abnormalities. Yashtimadhu (*Glycyrrhiza glabra*) is an Ayurvedic Rasayana medication with qualities related to systemic and reproductive health that is commonly referred to as vrushya. The current study used network pharmacology and molecular docking techniques to clarify its possible anti-infertile function in PMOS. **Materials and Methods:** *Glycyrrhiza glabra* phytochemicals were obtained from Dr. Duke's and IMPPAT databases, and ADMETlab 3.0 was used for screening. Swiss Target Prediction was used to predict the targets of the chosen phytoconstituents, and GeneCards and the Human Protein Atlas provided PMOS-associated targets. Venny 2.1.0 was used to identify overlapping targets, and then STRING, Cytoscape, CytoHubba, and DAVID were used for topological analysis, hub-gene discovery, protein-protein interaction analysis, and KEGG pathway enrichment. For a few phytoconstituents, molecular docking against important hub targets was carried out. **Results and Discussion:** 21 of the 432 distinct phytochemicals that were found passed the ADMET screening. Fifteen phytochemicals produced 467 predicted targets. 93 of the 733 relevant targets found by disease-target analysis overlapped with *G. glabra* and PMOS. While CASP3, IL6, TNF, STAT3, ESR1, PTGS2, MMP9, MTOR, JUN, and HIF1A were shown to be the top hub targets, ESR1, PPARG, and TNF had notable interactions. Glabrolide's docking affinity against IL6 was the highest, at −7.9 kcal/mol. **Conclusion:** According to the results, Yashtimadhu may have a potential multi-target, multi-pathway effect on PMOS, especially through metabolic, hormonal, inflammatory, and apoptotic pathways. Additional *in vitro, in vivo*, and clinical research is needed to confirm these computational results.

Keywords: *In silico* study, Phytochemicals, PMOS, yashtimadhu.

Correspondence:
Gunjan Agrawal
Department of Swasthavritta, Government Ayurveda College, Nagpur, Maharashtra, INDIA.
Email: agrawalgunjan1999@gmail.com

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INTRODUCTION

Polycystic ovarian syndrome is now renamed as Polyendocrine Metabolic Ovarian Syndrome (PMOS) due to the multisystem features of the condition (Arif *et al*., 2020). PMOS highlights the multiple interrelating endocrine irregularities rather than a single system involvement (Basu *et al*., 2018). The recent global prevalence of condition is reported as 12.1% of the women (Batiha *et al*., 2020). As the name of disease is changed and new guidelines and norms are under process the study used same data as of the previous disease. Studies confirmed that PMOS has genetically complex origins among metabolic, reproductive and neuroendocrine pathways (Burley *et al*., 2023). Neuroendocrine abnormalities and Hyperandrogenism caused due to ovarian/adrenal androgen excess are the defining features (Carlson, 2025). Metabolic dysfunction involves conditions such as impaired glucose tolerance, type 2 diabetes, obesity, hypertension, dyslipidaemia and increased cardiovascular risk (Chen *et al*., 2026). These conditions affect ovarian dysfunction due to disturbed steroidogenesis and follicular maturation, leads to menstrual disturbance, ovulatory dysfunction and infertility (Chin *et al*., 2014). The previously used term PCOS was considered inappropriate as the depth of pathology and multi systemic involvement was not focused and the change adds to multisystemic involvement of disease. The treatment modalities of PCOS consists of drugs for ovulation induction such as Clomiphene citrate. The antidiabetic agent metformin is the drug of choice. Other treatment modalities include aromatase inhibitors, gonadotropins, oral contraceptives, antiandrogens, and statins. All these treatment modalities have adverse effects such as hot flushes, skin irritation and redness, Ovarian

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hyperstimulation syndrome (Cui *et al.*, 2022). The treatment modalities try to correct the symptoms but often fail to cure the condition as the root cause is not focused. Stress can be a leading cause for conditions like PMOS as it triggers and worsen the hormonal disruption and insulin resistance (Dallakyan and Olson, 2015).

Yashtimadhu (*Glycyrrhiza glabra*) is a well-known medicinal herb from Fabaceae family. The root and rhizome are main useful part of the plant. The drug is widely used in traditional medicines in the form of root powder, decoctions and extracts (Dassault Systèmes, 2021). The documented phytoconstituents and pharmacological activity of the drug particularly antioxidants, anti-inflammatory and metabolic (Dunaif and Peña, 2012). Effects provide a basis for the relevance of the drug in multi systemic disorder involving endocrine and metabolic dysregulation. As the direct evidences for the relevance of the drug in PMOS is not available and considering systemic involvement of the disease, involving reproductive, psychological metabolic and endocrine component, a network pharmacology approach may help elucidate the potential interactions between the phytoconstituents of *Yashtimadhu* and the molecular targets and signalling pathways associated with these processes. Such an approach may provide a mechanistic bridge between the traditional *Rasayana* (rejuvenation) concept of *Yashtimadhu* and its potential therapeutic relevance in the multifactorial pathophysiology of PMOS.

MATERIALS AND METHODS

Ethical statement

The study does not need clearance from the Institutional Ethical Committee or the Institutional Animal Ethical Committee because it does not involve any human or animal subjects.

Collection of phytochemicals and its related targets

The phytochemicals associated with the plant that are part-specific were gathered using two databases, Dr. Dukes and IMPPAT (Erchen Decoction Study, 2025; Fico *et al.*, 2015). Additionally, the listed phytochemical's SMILES (Simplified Molecular Input Line Entry System) were gathered and subsequently uploaded into ADMET Lab 3.0 for the evaluation of Absorption, Distribution, Metabolism, Excretion, and Toxicity (Fu *et al.*, 2024). With a cutoff score greater than 0.67 for QED, Lipinski as 0 or 1, and a medium cutoff score of 0.7 for Human Intestinal Absorption, Blood-Brain Barrier, Bioavailability and Carcinogenicity, the ADMET evaluation was performed. Next, each SMILES is uploaded into Swiss Target Prediction for the procurement of targets associated with each phytochemical depending on the evaluation (Gfeller *et al.*, 2014).

Disease target and Overlapping targets

After selecting the disease for which the plant must be assessed, the disease-related targets are gathered from GeneCards and HPA (Human Protein Atlas) (He *et al.*, 2025; Hiam *et al.*, 2019). The targets in the GeneCards database are filtered based on the relevance score, and duplicates in both databases are eliminated. Next, to obtain the overlapping targets, the targets associated with disease and plant phytochemicals are uploaded into Venn 2.1.0 (Joham and Teede, 2022).

Protein interaction and Top 10 genes

The overlapping targets are entered into STRING 12.0 in order to evaluate how each target interacts with the others. Additionally, Cytoscape 3.10.4 was used to upload the targets in order to anticipate the top 10 common targets (Ju *et al.*, 1989). CytoHubba was utilized to predict the top ten gene targets (Kim and Chae, 2014).

KEGG pathway prediction and Topological analysis

The Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway prediction was performed in DAVID Bioinformatics using the overlapped targets (Kocaoğlu *et al.*, 2025). After uploading the targets and obtaining the pathways associated with them, the top 10 pathways were taken into consideration based on the number of genes. After separating the targets, chemicals, and pathways and forcing them to combine to create a network, topological analysis is carried out. In order to determine which phytochemicals and targets are suitable for molecular docking, edge betweenness and degree layout are evaluated.

Molecular docking

The 3D structure of the ligand is obtained from PubChem, and the target related structure is obtained from RCSB PDB (Li *et al.*, 2022; Mohanraj *et al.*, 2018). The hetero atoms and the water molecules were removed from the target further polar hydrogen bond, and Kollman charges were added to the target. Using PyRx software, the ligands were uploaded with the force field of UFF and mmff94 after energy reduction (Mukherjee *et al.*, 2020). Followed by that targets are uploaded into PyRx for docking and checking binding affinity score. With Discovery studio 2025 the 2D and 3D visualization of the docking is done (Nanda *et al.*, 2019).

RESULTS

Phytochemical collection and Targets related

The plant's name "*Glycyrrhiza glabra*" was entered into IMPPAT and Dr. Dukes and a total of 347 phytochemical was acquired in Dr. Dukes while a total of 157 phytochemicals were acquired in IMPPAT. Followed by that after merging and removing the duplicates, a total of 432 phytochemicals were present at the last. These phytochemicals were entered into PubChem one by one and later a total of 257 SMILES were obtained, related to the phytochemicals. Later, when ADMET analysis was done a total of 21 phytochemicals were obtained at the last. While uploading

each SMILES into Swiss Target Prediction, a total of 1292 targets related to 15 phytochemicals were present. After removing the duplicates, a total of 467 targets were obtained at the last related to the plant's phytochemicals.

Disease related target and Common targets prediction

The disease "Poly Cystic Ovarian Syndrome" was entered into Gene cards and HPA, followed by that 2027 targets were present in Gene cards while HPA had 7 targets followed by merging and removing 2027 targets were present related to protein coding. With a relevance score of 0.50, 733 targets were found. By merging the plant related targets and disease related targets, the overlapping targets were predicted and the number was 93 in total.

Protein Interaction, Hub Genes, KEGG pathway and Topological analysis

The overlapping targets were uploaded to visualise the interaction between the proteins, followed by that visualisation the most interacted targets were ESR1, PPARG and TNF. While assessing the hub genes prediction on Cytohubba the top 10 targets among the 93 were, CASP3, IL6, TNF, STAT3, ESR1, PTGS2, MMP9, MTOR, JUN and HIF1A. The common targets also gave the pathways related to them with DAVID bioinformatics with a total of 159 pathways, among these the top 10 pathways were segregated on the basis of gene count in them. The Pathways in cancer had a gene count of 32 while the Proteoglycans in cancer had a count of 22. The 10 pathways, 467 targets and 15 compounds were uploaded into Cytoscape 3.10.4, liquiritin, glabrolide, beta-carotene and p-cymene were the compounds with highest degree layout. The Figure 1 shows the PPI, Hub genes, common targets and Figure 2 depicts the topological analysis of *Glycyrrhiza glabra*.

Molecular docking

The molecular docking was done with the 10 phytochemicals towards the top 3 hub gene targets, CASP3, STAT3 and IL6. Among this Glabrolide had the highest binding affinity of -7.9 kcal/mol, while the chemical 22,23-Dihydrostigmasterol lowest binding affinity of -4.3 kcal/mol. Meanwhile few of the

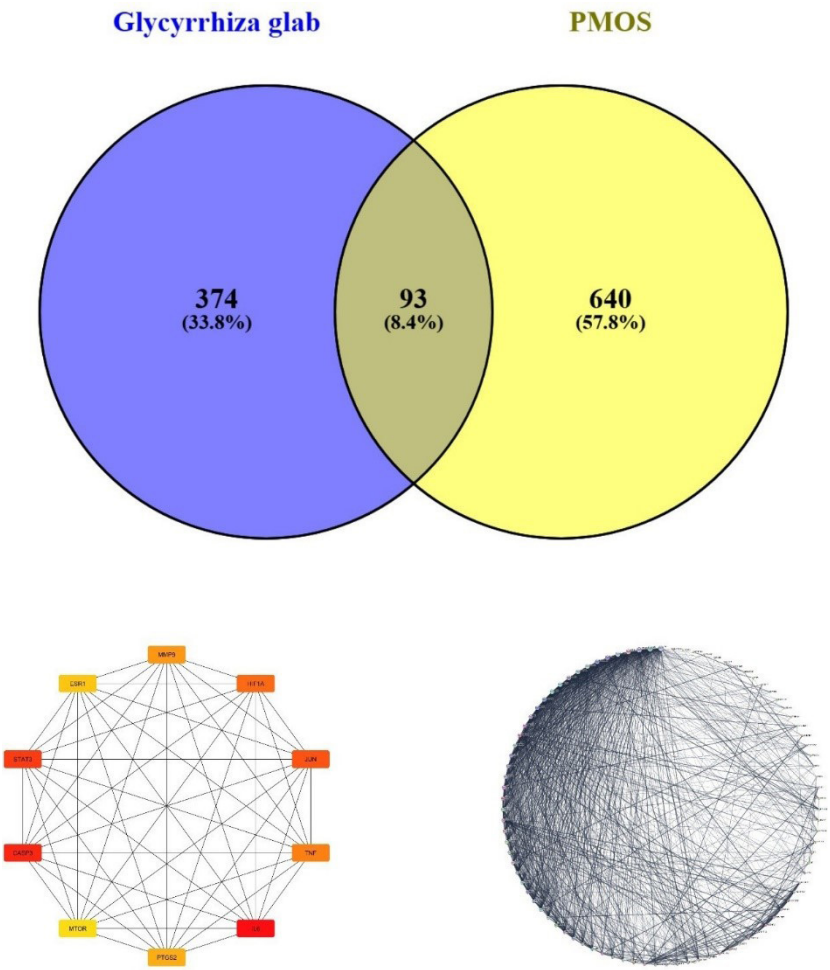

Figure 1: PPI, hub genes, and common targets of Yashtimadhu towards PMOS. Protein-protein interaction network showing the overlapping targets of *Glycyrrhiza glabra* phytoconstituents and PMOS-associated targets, including the identified hub genes. Reproduction size: full page width.

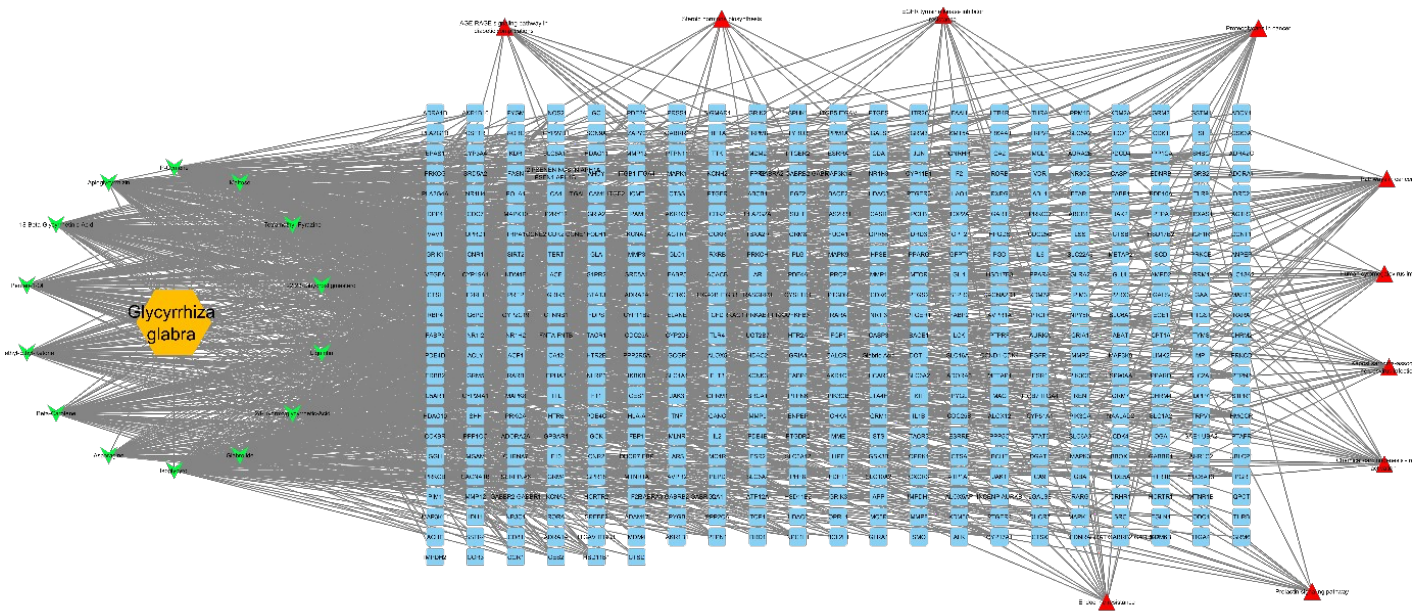

Figure 2: Topological analysis of Yashtimadhu in PMOS. Network showing the relationships among the selected phytochemicals, molecular targets, and KEGG pathways identified in the analysis. Reproduction size: full page width.

phytochemical such as liquiritin, 18-Beta-Glycyrrhetinic-Acid, 28-Hydroxyglycyrrhetic-Acid. The average binding affinity was there for the chemical Glabrolide with -7.5 kcal/mol. The lowest average binding affinity was there for 22,23-Dihydrostigmasterol, with an affinity of -4.5 kcal/mol. The 2D and 3D docking visualisation of Glabrolide towards IL6 is depicted in Figure 3 and Table 1.

DISCUSSION

Yashtimadhu is considered as one of the most potential drugs in ayurveda due to its wide range of therapeutic actions. It is known for its Chakshushya (Ocular health promoting), Bala(restorative), Varnikrita (complexion enhancing), Snigdha (lubricating), Shukrala (supporting fertility) and Keshya (hair health enhancing) properties. Yashtimadhu is a Rasayana (health-promoting) drug having Madhura Rasa (Sweet taste), Madhura Vipaka(Sweet post-digestive metabolic transformation), Sheeta Virya (Cold principal therapeutic potency) and Snigdha-Guru Guna, which are responsible for its Vata-Pitta shamana and Ojas vardhana guna.

These provide a credible basis for its application in PMOS, particularly in relation to stress-associated manifestations and disturbances of *Manas* (*mental and psychological functions*) (Ndefo *et al.*, 2013). *Yashtimadhu* constituents of phytochemicals, including glabridin, glycyrrhizin, and liquiritigenin, which are well known for their neuroprotective, antioxidant, and anti-inflammatory properties signifying their probable involvement in pathways regulating hormonal homeostasis, stress response, and metabolic function (Neven *et al.*, 2026). In a previous study conducted on female rats, phytochemicals such as Glabrene, Isoliquiritigenin and Glabridin showed a significant surge in LH and FSH levels and modulatory effects on the hypothalamic-pituitary-ovarian axis (Oliveros, 2015).

The present network analysis identified IL-6, CASP3 and ESR1 as the key targets which with phytoconstituents of *Yashtimadhu*, may engage with PMOS pathophysiology. Molecular docking revealed significant binding interactions between the principal constituents liquiritin, Glabrolide suggesting a multi-target, multi-pathway mode of action. The Endocrine resistance pathways are hampered in PMOS mainly through altered reproductive hormone signalling, insulin resistance and inflammation. In Insulin resistance pathway impairment hyperinsulinemia and increased androgen synthesis in ovarian theca cells is observed mainly due to defects in PI3K/AKT metabolic pathway (Pastorino *et al.*, 2018; Riestenberg *et al.*, 2022).

Chronic inflammation through TNF-α, NF-kB and IL-6 signalling further intensifies insulin resistance (Shannon *et al.*, 2003). Further complications are created through hormonal imbalance and progesterone resistance contributing to endometrial dysfunction and related complications (Sherman *et al.*, 2022). Among the hub targets IL-6 emerged as a central node linking pro inflammatory cytokine causing endocrine resistance mainly due to its chronic low-grade inflammation in PMOS (Stelzer *et al.*, 2016).

Increased IL-6 is linked to activation of NF-kB and pro-apoptotic markers such as Caspase-3 and Bax and decreased activity of Bcl-2, signifying its role in follicular atresia and granulosa cell apoptosis (Stener-Victorin *et al.*, 2024). Increased levels of IL-6 may further lead to cardiovascular risk and insulin resistance (Szklarczyk *et al.*, 2023). CASP3 was identified as a critical effector node connecting granulosa cell apoptosis with the

follicular dysfunction characteristics of PMOS. CASP3 plays a significant role in follicular atresia and granulosa cell apoptosis in PMOS. Further this granulosa cell damage is promoted by Insulin resistance, hormonal imbalance, cytokines, inflammation, oxidative stress and mitochondrial damage encourage granulosa cell apoptosis (Tang *et al.*, 2023). Polycystic ovarian morphology and disturbed follicular turnover are linked to altered CASP3 expression which is seen in PMOS (Teede *et al.*, 2023).

The identification of ESR1 as a hub target in the present analysis depicts its role as a key mediator of oestrogen signalling disruption in PMOS. Relatively reduced ERβ and GPER1 activity relative to ERα leads to metabolic, neuropsychiatric and reproductive abnormalities in PMOS. This further leads to increased androgen production, contributing to hyperandrogenism (Thompson *et al.*, 2008). ESR1 polymorphisms are also related to inflammatory and metabolic factors in PCOS (Thul and Lindskog, 2018). Among the phytoconstituents of *Yashtimadhu*, liquiritin demonstrated the most extensive interactions across the identified hub targets. Liquiritin enhances miR-206 and suppresses the PI3K/AKT function, which in turn regulates ovarian granulosa cell function (Tong *et al.*, 2022). It interacts with CYP17A1, an enzyme associated in androgen biosynthesis and modulates steroidogenic enzymes, which helps to normalise the elevated testosterone (Xue *et al.*, 2024).

In animal models, liquiritin improves insulin and glucose sensitivity and improves ovarian follicular changes fractionally by regulating bile acid metabolism and reducing chronic inflammation (Zeb and Gul, 2025). Glabrolide, another phytoconstituent retrieved, showed predicted interaction with neuroinflammatory pathways, as no direct evidence of how Glabrolide acts in PMOS is available, but an indirect reference suggests something about BACE-1 inhibitory activity, which suggests relevance through its action on neuroinflammatory and insulin resistance conditions associated with PMOS.

Figure 3: 2D and 3D visualization of Glabrolide towards IL6. Molecular docking visualization showing the interaction of Glabrolide with the IL6 target. Reproduction size: column width.

Table 1: Compounds of *Glycyrrhiza glabra* towards hub gene targets. Molecular docking binding affinity scores (kcal/mol) of the selected *Glycyrrhiza glabra* phytochemicals against CASP3, STAT3, and IL6.

Targets	CASP3	STAT3	IL6
Phytochemical			
P-Cymene	-6.3	-6.2	-6.1
Maltose	-5.7	-5.7	-5.4
Isoglycyrol	-6.1	-6.2	-6.5
Glabrolide	-6.8	-7.8	-7.9
22,23-Dihydrostigmasterol	-4.7	-4.5	-4.3
18-Beta-Glycyrrhetinic-Acid	-6.2	-6.3	-6.5
Tetramethyl-Pyrazine	-5.4	-4.8	-5.6
Liquiritin	-7.1	-7.3	-6.7
Apioglycyrrhizin	-6.2	-6.5	-6.3
28-Hydroxyglycyrrhetic-Acid	-6.8	-6.6	-6.3

Note: Values represent molecular docking binding energies in kcal/mol. CASP3 = Caspase-3; STAT3 = Signal Transducer and Activator of Transcription 3; IL6 = Interleukin-6.

This integrative approach provides a mechanistic basis for the potential multi-target action of Yashtimadhu in PMOS. However, the identified interactions need further experimental validation primarily for the less studied phytoconstituents such as Glabrolide. Further *in vitro, in vivo* and clinical studies are required to validate these targets and their therapeutic relevance in PMOS.

CONCLUSION

According to the current study, *Yashtimadhu* (*Glycyrrhiza glabra*) may have potential in PMOS through multi-target and multi-pathway activities involving hormonal, inflammatory, metabolic, and apoptotic pathways. Glabrolide had the highest docking affinity; however, IL6, CASP3, and ESR1 were identified as significant targets. The traditional *vrushya* and *Rasayana* role of *Yashtimadhu* has a basic mechanistic basis thanks to these discoveries, but more *in vitro, in vivo*, and clinical research is needed for validation.

ABBREVIATIONS

PMOS: Polyendocrine Metabolic Ovarian Syndrome; **PCOS:** Polycystic Ovarian Syndrome; **ADMET:** Absorption, Distribution, Metabolism, Excretion, and Toxicity; **SMILES:** Simplified Molecular Input Line Entry System; **KEGG:** Kyoto Encyclopedia of Genes and Genomes; **PPI:** Protein-Protein Interaction; **IL-6:** Interleukin-6; **CASP3:** Caspase-3; **ESR1:** Estrogen Receptor 1; **TNF:** Tumor Necrosis Factor; **STAT3:** Signal Transducer and Activator of Transcription 3; **PTGS2:** Prostaglandin-Endoperoxide Synthase 2; **MMP9:** Matrix Metalloproteinase 9; **MTOR:** Mechanistic Target of Rapamycin; **HIF1A:** Hypoxia-Inducible Factor 1-Alpha; **PPARG:** Peroxisome Proliferator-Activated Receptor Gamma; **LH:** Luteinizing Hormone; **FSH:** Follicle-Stimulating Hormone; **CYP17A1:** Cytochrome P450 Family 17 Subfamily A Member 1; **NF-κB:** Nuclear Factor Kappa B; **BACE-1:** Beta-Secretase 1; **PI3K/AKT:** Phosphoinositide 3-Kinase/Protein Kinase B.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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