Original Article

Histopathological And Biochemical Evaluation Of Piperine-Induced Changes In Ovarian Follicles And Hormonal Profile In Adult Albino Rats

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Abstract

Objective: This study aimed to evaluate the histopathological and biochemical alterations caused by piperine in the ovaries of adult female Wistar albino rats.

Methods: Thirty rats (3–4 months old, 200–250 g) were divided into three groups (n=10 each). Group A (control) received normal saline, Group B received Piperine at 5 mg/kg, and Group C received Piperine at 10 mg/kg body weight daily for 30 days. Serum oestrogen and progesterone levels were measured using ELISA. Histology of ovarian tissues was performed, and estrous cycle changes were monitored.

Results: Piperine exposure produced a dose-dependent increase in secondary follicle diameter (Group A: 210.4 ± 15.2 µm; Group B: 285.7 ± 18.9 µm; Group C: 342.1 ± 22.7 µm; $p < 0.001$). Serum oestrogen levels were significantly elevated in Groups B and C (62.7 ± 5.3 pg/ml and 75.4 ± 6.1 pg/ml) compared to those in the controls (48.2 ± 4.5 pg/ml), whereas progesterone levels declined (Group A: 1.45 ± 0.35 ng/ml; Group B: 1.01 ± 0.33 ng/ml; Group C: 0.41 ± 0.12 ng/ml; $p < 0.001$).

Conclusion: Piperine caused dose-dependent enlargement of secondary follicles with elevated oestrogen and reduced progesterone, leading to an arrest of the ovarian cycle at the proestrus stage. These findings suggest that piperine may be a reproductive toxicant in women of reproductive age.

Keywords: Piperine, Secondary Follicles, Estrogen, Progesterone, Estrous cycle.

Introduction

Piperine, a nitrogen-containing alkaloid, is the main component of black pepper, which provides a pungent flavour. Out of all spices, black pepper is known as the 'King of spices.' For centuries, it has been used as an essential spice in the human diet. For centuries, it has been used as an essential spice in the human diet and in traditional medicinal practices.[1] It was discovered by Hans Christian Ørsted in 1819.[2] Piperine is well recognised for its neuroprotective effects and its ability to slow neurodegenerative processes because of its diverse pharmacological properties. These effects are mediated through several physiological mechanisms, including its strong antioxidant activity, which reduces oxidative stress and neuronal damage, and its anti-inflammatory action, which suppresses microglial activation and the release of proinflammatory cytokines in the brain. Piperine also modulates key neurotransmitter systems, including serotonin, dopamine, and gamma-aminobutyric acid (GABA), thereby improving synaptic transmission, memory, and mood. In addition, piperine inhibits acetylcholinesterase, leading to increased availability of acetylcholine, which further supports cognitive function and neuronal survival. Through these combined mechanisms, piperine contributes to neuroprotection, memory enhancement, and antidepressant effects.[3] On the liver, piperine modulates detoxification enzymes, provides protection against oxidative stress, and is used in the treatment of diabetes and hepatic steatosis.[4] According to the study by Wei *et al*. on mice, piperine was found to have beneficial effects on the cardiovascular system. Their results showed that piperine decreases serum cholesterol levels and increases lipid metabolism.[5] It is also used as a painkiller and cough suppressant.[6,7] Piperine also possesses anti-inflammatory, antioxidant, and anticancerous properties.[7,8] Piperine increases the bioavailability of nutrients and many drugs with the help of intestinal absorption and enhances hepatic metabolism.[9]

The reproductive systems of both males and females are very sensitive to xenobiotics, such as environmental agents and dietary supplements. The functioning of female reproductive organs, especially the ovaries, is under the influence of both external and internal factors (hormones and local tissue factors).[10] Folliculogenesis is a primary function of the ovaries. It is a process of formation of ovarian follicles from different stages, e.g., Primordial to Graafian follicle. These ovarian follicles are the anatomical and physiological units of the ovaries. The histological morphology of these follicles at different stages can easily predict ovarian function along with its reproductive potential. This process is mainly controlled by the hypothalamic–pituitary–gonadal (HPG) axis.[11] pituitary gland releases gonadotropins, primarily follicle-stimulating hormone (FSH) and luteinizing hormone (LH). These hormones regulate the growth and function of female gonads (ovaries) and play an important role in the development and maturation of ovarian follicles. Successively, the ovaries produce hormones such as estrogen and progesterone (steroids in nature). These hormones are responsible for ovulation, the development of the endometrium, implantation of the embryo, and maintenance of pregnancy.[12] Hormonal imbalances from external agents can disrupt follicle morphology. Owing to this sensitivity, the ovaries are most commonly studied as target organs to evaluate reproductive toxicity due to the adverse effects of drugs and chemicals.[13] At the same time, some studies suggest that excessive usage alters the functions of the kidneys and pancreas.[14] Studies on piperine also suggest that it can influence the functioning of the female reproductive

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system; however, the findings are fragmented on its direct effects on ovarian follicular morphology and female hormones. This lack of information is concerning because many people use herbal medicines and dietary supplements that contain piperine and believe that they are completely safe and beneficial. Without clear scientific evidence, the regular use of piperine can be fatal for female reproductive health.
Therefore, the present study was designed to fill this knowledge gap. Specifically, we aimed to identify changes in follicular morphology and to measure alterations in serum oestrogen and progesterone levels following chronic piperine administration.

Materials And Methods

This animal-based randomised control trial was conducted over a period of six months (from January 2024 to July 2024) at the animal house and histology laboratory of the Niazi Medical and Dental College, Sargodha, Pakistan. Ethical approval for this study was obtained by the Ethical Review Committee of the institute vide letter no. NMDC; ERC/CER/31/23. Thirty adult female Wistar albino rats aged 3–4 months and weighing 200–250 g were obtained from the Veterinary Research Institute, Lahore. The sample size was selected in accordance with previous experimental studies evaluating ovarian histology and hormonal changes in rodents, where a group size of 8–10 animals was considered sufficient to detect biologically meaningful differences between treatment groups.[15,16] Animals were kept in the institutional animal house and acclimatised for one week before starting the experiment. Standard laboratory conditions were maintained: a 12-h light and dark cycle, optimum temperature (25±2 °C), and standard food and distilled water *ad libitum*. Rats were divided into three groups using a non-probability convenience sampling technique (n = 10):
Group A (control) received normal saline (10 ml/kg body weight) by orogastric gavage daily for 30 days along with a standard diet and distilled water *ad libitum*.
Group B (experimental group 1) received piperine mixed in normal saline at 5 mg/kg body weight by orogastric gavage daily for 30 days along with a standard diet and distilled water *ad libitum*.
Group C (experimental group 2): received piperine mixed in normal saline at 10 mg/kg body weight by orogastric gavage daily for 30 days along with a standard diet and distilled water *ad libitum*.
Every day, piperine suspensions were freshly prepared and administered at approximately the same time each morning. Piperine was extracted from black pepper by the Soxhlet method using ethanol as the solvent, purified, and quantified using gas chromatography–mass spectrometry (GC–MS). The purified compound was freshly prepared in normal saline before being administered to the animals.[17] On the 30th day of the experiment, the animals were anaesthetised with chloroform. Blood was collected by a single cardiac puncture with the help of sterile syringes. Approximately 3–4 mL of blood was withdrawn from each animal. The collected blood samples were transferred into clot activator tubes and allowed to clot at room temperature for 30 min. To separate the serum, the samples were centrifuged at 3000 rpm for 10 min. The separated serum was carefully pipetted into aseptic Eppendorf tubes, labelled, and stored at -80◦C for hormonal analysis. Serum oestrogen and progesterone levels were measured using commercially available rat ELISA kits.[18] After blood collection, the abdominal cavity was opened, and both ovaries were carefully dissected following the identification of the uterine tubes. Immediately after all that excised ovaries were fixed in 10% neutral buffered formalin. Fixed tissues were processed using routine histological methods (dehydration, clearing, paraffin infiltration, and embedding). Serial sections of 4–5 µm thickness were cut using a rotary microtome and stained with haematoxylin and eosin (H&E).[19]
Five sections of each ovary were prepared. The diameters of the secondary follicles were measured using an ocular micrometer under a light microscope. For each animal, the mean follicular diameter was calculated from measurements across the selected sections.[20] Group A (control) values were used as the reference for comparison with experimental groups B and C. All raw data, including follicle diameters and hormonal results, were recorded on standardised proformas for statistical analysis.
To analyze the data, SPSS version 24 was used. Results of biochemical parameters and diameter of secondary follicles were analyzed by Mean, $\pm$ S.D and one-way ANOVA was used to compare data among groups. Intergroup comparison was done by post hoc Tukey's HSD test. The *p*-value ≤ 0.05 was considered statistically significant.

Results

In Group A (control), the mean diameters of secondary follicles were 210.4 ± 15.2 µm. In Group B (experimental group 1), it was 285.7 ± 18.9 µm and 342.1 ± 22.7 µm in Group C (experimental group 2) (Table 1). Statistical analysis demonstrated a significant difference among the groups ($p < 0.001$, Table 2).

Table 1: Mean diameter of secondary follicles (µm) in control and experimental groups (n=10)

Group A Mean ± SD	Group B Mean ± SD	Group C Mean ± SD	*p*-value
210.4 ± 15.2 µm	285.7 ± 18.9 µm	342.1 ± 22.7 µm	<0.001

Table 2: Post hoc Tukey's Analysis for inter-group comparison of mean follicular diameter

(I) Group	(J) Group	Sig. (p-value)
Group A (Control)	Group B (Experimental 1)	0.004
Group A (Control)	Group C (Experimental 2)	<0.001
Group B (Experimental 1)	Group C (Experimental 2)	0.004

Table 3: Serum Estrogen and Progesterone Levels in Control and Experimental Groups (n=10)

Hormone	Group A (Mean ± SD)	Group B (Mean ± SD)	Group C (Mean ± SD)	*p*-value (ANOVA)
Estrogen (pg/mL)	48.2 ± 4.5 Estrus-proestrous	62.7 ± 5.3 Pro-estrous	75.4 ± 6.1 Pro-estrous	< 0.001
Progesterone (ng/mL)	1.45 ± 0.35 corresponds to a low–moderate proestrus level, but still higher than suppressed groups	± 0.33 suppressed progesterone compared with controls	0.41 ± 0.12 the lowest level, indicating ovarian cycle arrest	< 0.001

Table 4: Post-hoc Multiple Comparisons of Serum Hormone Levels among Study Groups

Hormone	Comparison	*p*-value
Estrogen (pg/mL)	A vs B	< 0.001
	A vs C	< 0.001
	B vs C	< 0.001
Progesterone (ng/mL)	A vs B	< 0.001
	A vs C	< 0.001
	B vs C	0.002

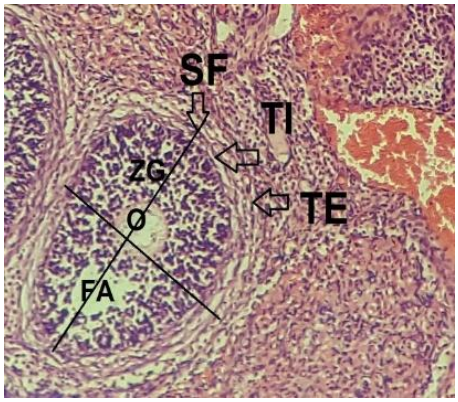

Figure 1: A cross section of a rat ovary from control group (A), demonstrating the secondary follicle surrounded by somatic cells (granulosa cells) and thecal cells. Diameter is 198.7± 12.4 µm. Original magnification at 10X

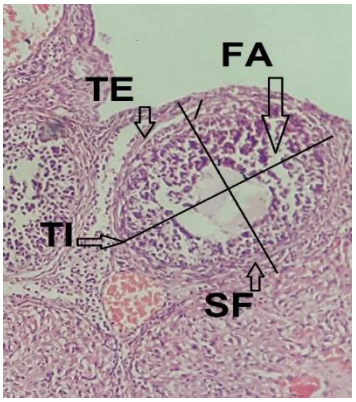

Figure 2: A cross section of a rat ovary from experimental group 1(B), demonstrating the secondary follicle surrounded by somatic cells (granulosa cells) and thecal cells. Diameter is 232.6 ± 16.8 µm. Original magnification at10X

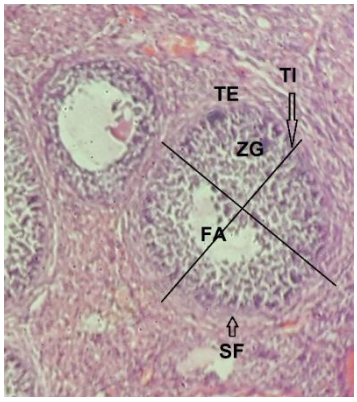

Figure 3: A cross section of a rat ovary from experimental group 2 (C), demonstrating the secondary follicle surrounded by somatic cells (granulosa cells) and thecal cells. Diameter is 360.6 ± 24.9 µm. Original magnification at 10X

Discussion

Piperine is an organic alkaloid and pungent in taste found in piper nigrum (black pepper) and piper longum (long pepper). Its hepatoprotective, immunomodulatory, and anti-tumor effects have already been discovered, and it is used as a natural medicine for the management of asthma and allergies, showing promising results.[21] On the other hand, some other studies have reported negative effects of piperine on oxidative defense mechanisms, drug-drug interactions, bioavailability of various medicinal compounds, and biochemical activity of many enzymes, e.g., superoxide dismutase, catalase, CYP P450 monooxygenase, and glucoronyl transferase. This diverse data has shown that piperine is biologically a versatile compound that imposes diverse biochemical effects on different systems and functions of the body.[22]

This laboratory-based animal study observed the effects of low and high consecutive doses of piperine (5 and 10 mg/kg, respectively) for 30 days on the diameter of secondary follicles of adult Wistar albino rats and female reproductive hormones (serum oestrogen and progesterone) to gain a comprehensive understanding of its impact. Laboratory female rats have four phases in their estrus cycle: proestrus, estrus, metestrus, and diestrus. Reproductive hormone levels vary in these stages. The pro-estrus phase is similar to the human female follicular phase, characterised by the proliferation of primordial follicles into secondary and graafian follicles under the influence of follicle stimulating hormone(FSH), preparing it for ovulation associated with elevated oestrogen and reduced progesterone levels. In the oestrus phase, the largest follicle ruptures, called ovulation, and it occurs due to a luteinizing hormone surge. Oestrogen levels drop immediately after ovulation, whereas progesterone levels are at their lowest. In the metestrus phase, the formation of the corpus luteum begins, which starts to release progesterone. Serum oestrogen levels are lower in this phase. In the diestrus phase, the corpus luteum is fully active, serum progesterone is highest, and oestrogen is lowest.[23]

Our results demonstrated a significant increase in serum oestrogen levels in both experimental groups (B and C) with a dose-dependent increase and corresponding reductions in serum progesterone. Control rats maintained normal oestrogen (48.2 ± 4.5 pg/ml) and progesterone (1.45 ± 0.35 ng/ml) levels, whereas the experimental groups showed elevated oestrogen (62.7 ± 5.3 pg/ml in Group B and 75.4 ± 6.1 pg/ml in Group C) and markedly suppressed progesterone (1.01 ± 0.33 ng/ml in Group B and 0.41 ± 0.12 ng/ml in Group C). These findings indicate a clear disruption of the estrous cycle, with consistent arrest in the proestrus phase, where oestrogen is persistently high, and progesterone remains suppressed. These values of serum oestrogen and progesterone were statistically significant (p <0.001). Post hoc Tukey's Honest Significant Difference (HSD) test, when applied for intergroup comparison of serum oestrogen and progesterone, showed statistically significant differences within group A and B, group A and C, and group B and C ($p \leq 0.001$). Similar hormonal alterations have been described by Daware *et al.,* but interestingly, their findings are different from ours. He reported that the administration of piperine at 10–20 mg/kg in Swiss albino rats caused prolongation of the diestrus phase with persistent corpus luteum activity and elevated progesterone levels, decreased mating behaviour, reduced implantation, and ultimately led to subfertility.[24] This can be explained by differences in rat strain (Wistar vs. Swiss albino), dosage (5–10 mg/kg vs. 10–20 mg/kg), duration of exposure, and the baseline phase of the estrous cycle at the start of treatment. It is possible that lower doses of piperine predominantly interfere with folliculogenesis, resulting in proestrus arrest, while higher doses disrupt luteal physiology and prolong diestrus. Thus, the reproductive effects of piperine can be dose-dependent and phase-specific. Radic *et al.* also emphasised that low doses of piperine, comparable to those used in the current study, can alter the hypothalamic–pituitary–gonadal (HPG) axis, elevate gonadotropins (FSH and LH), and disrupt germ cell development in both male and female rats.[7] These reports align with our biochemical data, supporting the study by Jibira

et al. that piperine-induced endocrine disruption is mediated through interference with steroidogenic enzymes such as 11α-hydroxylase and 21α-hydroxylase. The inhibition of these cytochrome P450-dependent enzymes interferes with the biosynthesis of steroids and the normal ovarian cycle.[25]

Histological findings further corroborated these observations. Compared to the control, the ovaries of piperine-treated rats showed an increased mean diameter of secondary follicles (Group A: 210.4 ± 15.2 µm; Group B: 285.7 ± 18.9 µm; Group C: 342.1 ± 22.7 µm; $p < 0.001$). Instead of developing into mature Graafian follicles, these follicles failed to progress. The failure to form the corpus luteum suggests an arrest of follicular maturation at the secondary stage. The combination of enlarged secondary follicles, high oestrogen levels, and low progesterone levels is strongly indicative of proestrus arrest. These changes in ovarian tissue are in accordance with the hormonal results. When oestrogen levels are high but there is no proper increase in LH, ovulation does not occur, and the follicles cannot develop into a corpus luteum. Similar observations have been reported by Singh, who noted increased follicular atresia and disrupted ovarian histomorphology in rats with prolonged exposure to oestrogen and persistent estrus conditions, similar to a hormonal imbalance (proestrus/diestrus arrest).[26]

These findings are very important, especially for those who are using Piperine-enriched supplements in human populations for weight loss and metabolic regulation. Although the doses used in animal studies may not directly impact human consumption, the mechanism of action of piperine in humans is of great concern. Chronic exposure to piperine can predispose women to menstrual irregularities, ovulatory dysfunction, and infertility, especially with long-term use or high doses.

Conclusions

The findings revealed a dose-dependent increase in the diameter of secondary follicles, with persistently elevated oestrogen and markedly reduced progesterone levels preventing normal ovulation. These results raise important concerns about its reproductive safety, especially in women of reproductive age.

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References

1. Umapathy VR, Dhanavel A, Kesavan R, Natarajan PM, S B, P V. Anticancer Potential of the Principal Constituent of Piper nigrum, Piperine: A Comprehensive Review. Cureus. 2024;16(2):1–8. https://doi.org/10.7759/cureus.54425
2. Tiwari A, Mahadik KR, Gabhe SY. Piperine: A comprehensive review of methods of isolation, purification, and biological properties. Med Drug Discov [Internet]. 2020;7:100027. https://doi.org/10.1016/j.medidd.2020.100027
3. Balakrishnan R, Azam S, Kim IS, Choi DK. Neuroprotective Effects of Black Pepper and Its Bioactive Compounds in Age-Related Neurological Disorders. Aging Dis 2023 Jun 1;14(3):750–77. https://doi.org/10.14336/AD.2022.1022
4. Adeyemo OA, Ore A, Ajisafe EO. The protective effect of piperine on oxidative stress and hepatic damage induced by diisononyl phthalate in rats. Egypt J Basic Appl Sci [Internet]. 2021 Jan 1;8(1):293–301. https://doi.org/10.1080/2314808X.2021.1983746
5. Wei M, LYu P, Li P, Hu J, Wu R, Ouyang Q, et al. Baolier Capsule's Secret Weapon: Piperine Boosts Cholesterol Excretion to Combat Atherosclerosis. Drug Des Devel Ther [Internet]. 2024;18:6427–46. https://doi.org/10.2147/DDDT.S499598
6. Azam S, Park JY, Kim IS, Choi DK. Piperine and ItsMetabolite's Pharmacology in Neurodegenerative and Neurological Diseases. Biomedicines. 2022;10(1):1–16. https://doi.org/10.3390/biomedicines10010154
7. Wu Z, Hu Y, Hao R, Li R, Lu X, Itale MW, et al. Research Progress of Genomics Applications in Secondary Metabolites of Medicinal Plants: A Case Study in Safflower. Int J Mol Sci [Internet]. 2025 Apr 19;26(8):3867. https://doi.org/10.3390/ijms26083867
8. Hasan R, Bhuia MS, Chowdhury R, Khan MA, Mazumder M, Yana NT, et al. Piperine exerts anti-inflammatory effects and antagonises the properties of celecoxib and ketoprofen: in vivo and molecular docking studies. Nat Prod Res 2024 Oct 11;1–16. https://doi.org/10.1080/14786419.2024.2413039
9. Srinivasan K. Black Pepper and its Pungent Principle-Piperine: A Review of Diverse Physiological Effects. Crit Rev Food Sci Nutr [Internet]. 2007 Oct 25;47(8):735–48. https://doi.org/10.1080/10408390601062054
10. Amir S, Shah STA, Mamoulakis C, Docea AO, Kalantzi OI, Zachariou A, et al. Endocrine Disruptors Acting on Estrogen and Androgen Pathways Cause Reproductive Disorders through Multiple Mechanisms: A Review. Int J Environ Res Public Health [. 2021 Feb 4;18(4):1464. https://doi.org/10.3390/ijerph18041464
11. Orozco-Galindo BV, Sánchez-Ramírez B, González-Trevizo CL, Castro-Valenzuela B, Varela-Rodríguez L, Burrola-Barraza ME. Folliculogenesis: A Cellular Crosstalk Mechanism. Curr Issues Mol Biol. 2025;47(2):1–16.https://doi.org/10.3390/cimb47020113
12. Marques P, De Sousa Lages A, Skorupskaite K, Rozario KS, Anderson RA, George JT. Physiology of GnRH and Gonadotrophin Secretion Endotext. 2000.
13. Land KL, Miller FG, Fugate AC, Hannon PR. The effects of endocrine-disrupting chemicals on ovarian- and ovulation-related fertility outcomes. Mol Reprod Dev. 2022;89(12):608–31. https://doi.org/10.1002/mrd.23652
14. Stojanović-Radić Z, Pejčić M, Dimitrijević M, Aleksić A, V. Anil Kumar N, Salehi B, et al. Piperine-A Major Principle of Black Pepper: A Review of Its Bioactivity and Studies. Appl Sci [Internet]. 2019 Oct 12;9(20):4270.https://doi.org/10.3390/app9204270
15. Charan J, Kantharia N. How to calculate sample size in animal studies? J Pharmacol Pharmacother. 2013;4(4):303–6. https://doi.org/10.4103/0976-500X.119726
16. Okafor IA, Nnamah US, Nnaka J. The fertility assessment of normal cyclic Wistar rats following the administration of methanolic extract of Portulaca oleracea: an experimental study. Middle East Fertil Soc J. 2021;26(1). https://doi.org/10.1186/s43043-020-00048-x

17. Sander A, Bival Štefan M, Sander T, Kučić Grgić D, Parlov Vuković J, Blažević I, et al. Characterization of Essential Oils and Ethanolic Extracts from Nine Pepper Species: Antioxidant and Antimicrobial Activity and Spectroscopic Analysis. Molecules. 2025;30(20):1–38. https://doi.org/10.3390/molecules30204140
18. Jahangir MA. Serum & Plasma Sampling from Small Animal Species Internationale Pharmaceutica Sciencia Serum & Plasma Sampling from Small Animal Species. 2025;(July):16–9. https://doi.org/10.31531/2231-5896.1000133
19. Al-Sabawy HB, Rahawi AM, Al-Mahmood SS. Standard techniques for formalin-fixed paraffin-embedded tissue: A Pathologist's perspective. Iraqi J Vet Sci. 2021;35(1–3):935–43. https://doi.org/10.33899/IJVS.2021.131918.2023
20. 20. Giovannopoulou E, Karakasi MV, Kouroupi M, Ieronimaki AI, Papakonstantinou E, Giatromanolaki A, et al. Ovarian Morphometric and Histologic Characteristics and Correlation with Clinical Factors: A Cross-Sectional Study. J Pers Med. 2023;13(2). https://doi.org/10.3390/jpm13020232
21. Siddiqui S, Khushtar M, Zafar A, Hasan SM, Arshad M, Ahmad MA, et al. Mechanism-Based Physiological Effects of Piperine: A Review. Curr Pharmacol Reports [Internet]. 2023 Mar 14;9(3):117–27. https://doi.org/10.1007/s40495-023-00314-2
22. Ziegenhagen R, Heimberg K, Lampen A, Hirsch-Ernst KI. Safety Aspects of the Use of Isolated Piperine Ingested as a Bolus. Foods [Internet]. 2021 Sep 8;10(9):2121. https://doi.org/10.3390/foods10092121
23. Ajayi AF, Akhigbe RE. Staging of the estrous cycle and induction of estrus in experimental rodents: an update. Fertil Res Pract. 2020;6(1):1–15. https://doi.org/10.1186/s40738-020-00074-3
24. Daware M, Mujumdar A, Ghaskadbi S. Reproductive Toxicity of Piperine in Swiss Albino Mice. Planta Med [Internet]. 2000 Dec 31;66(03):231–6.https://doi.org/ 10.1055/s-2000-8560
25. Yakubu J, Natsaridis E, du Toit T, Sousa Barata I, Tagit O, Pandey A. Curcumin and Piperine Nanoparticles inhibit CYP17A1 activity to Regulate Steroid Biosynthesis in Prostate Cancer. 2024. https://doi.org/10.20944/preprints202410.2072.v1
26. Singh KB. Persistent estrus rat models of polycystic ovary disease: an update. Fertil Steril 2005 Oct;84:1228–34. https://doi.org/10.1016/j.fertnstert.2005.06.013