Investigational
Medicinal Chemistry & Pharmacology

Short Communication Open Access

Modulatory effects of hydromethanolic leaf extract of *Craterispermum schweinfurthii* on the female reproductive cycle in Wistar rats

Friday Saronee[1*], Karibo Amakiri Okari[2], Tolunigba Abisola Kolawole[3]

Abstract

Background: The estrous cycle is a set of recurring physiological changes induced by reproductive hormones in female animals. This study investigated the potential modulatory effects of the hydromethanol leaf extract of *Craterispermum schweinfurthii* on the female reproductive cycle using Wistar rats as experimental models.

Methods: A total of 20 female Wistar rats weighing 100–250g were randomly divided into four groups of five rats each and treated as follows: Group A: Negative control (received extract vehicle). Groups B, C, and D: received 250, 500, and 750 mg/kg body weight of the extract, respectively. Vaginal smears were collected daily from each rat between 7:00 and 9:00 a.m. for four weeks (two weeks before treatment and two weeks during treatment).

Results: Administration of graded doses (250, 500, and 750 mg/kg body weight) of the hydromethanol leaf extract of *Craterispermum schweinfurthii* caused a significant reduction in the durations of proestrus, estrus, metestrus, and diestrus phases in Groups B, C, and D compared with the control group (Group A) ($p < 0.05$). This suggests that the extract enhances fertility. The extract apparently caused a pro-fertility effect by shortening the reproductive cycle length in the treated rats.

Conclusion: These findings therefore provide scientific validation for the traditional use of this plant in folklore medicine for fertility-related purposes in our environment.

Keywords: C*raterispermum schweinfurthi*; Diestrus; Metestrus; Oestrus; Proestrus**.**

**Correspondence: Tel.: +2347066060696; E-mail address: saroneef@fuotuoke.edu.ng ; ORCID: https://orcid.org/0000-0002-3280-7963 (Friday Saronee)*

[1]Department of Human Physiology, Faculty of Basic Medical Sciences, College of Medical Sciences, Federal University Otuoke, Bayelsa State, Nigeria. [2]Department of Medical Biochemistry, Faculty of Basic Medical Sciences, College of Medical Sciences. Rivers State University. Port Harcourt. Rivers State. Nigeria. [3]Department of Physiology, Caribbean School of Medical Sciences, Kingston, Jamaica.

Other authors
kariboa.okari@ust.edu.ng; ORCID: https://orcid.org/0009-0008-9117-8037 (Karibo Amakiri Okari). Tolaueneok02@yahoo.com; ORCID: https://orcid.org/0000-0002-8495-9643 (Tolunigba Abisola Kolawole)

Citation on this article: Authors. Investigational Medicinal Chemistry and Pharmacology (2026) 9(2):140; Doi: https://dx.doi.org/10.31183/imcp.2026.00140

Invest. Med. Chem. Pharmacol. (IMCP) ISSN: 2617-0019 (Print)/ 2617-0027 (Online); © The Author(s). 2026 Open Access This article is available at https://investchempharma.com/

Background

The estrous cycle consists of recurring physiological changes induced by reproductive hormones in female rats [1]. These cycles begin shortly after the attainment of sexual maturity and are interrupted by pregnancy or periods of anoestrus. They continue throughout the animal's reproductive life until death. Estrous cycle length and frequency vary widely among animal species [2]. Some species exhibit bloody vaginal discharge during certain phases [3]. The four phases of the estrous cycle are proestrus, estrus, metestrus, and diestrus. Cycle variability differs markedly between species, with smaller animals generally displaying more frequent cycles. Even within the same species, significant variability can be observed [4]. Domestication and environmental changes can also influence estrous cycle patterns. In most species, the phases and duration of the estrous cycle can be determined through vaginal smear cytology [5]. Rats are polyestrous animals with a short cycle length of typically 4–5 days [1]. Although they ovulate spontaneously, they do not form a fully functional corpus luteum unless they receive coital stimulation [1]. Fertile mating results in pregnancy, whereas infertile mating induces a state of pseudopregnancy lasting approximately 10 days [6]. The events of the estrous cycle in rats are strongly influenced by the photoperiod (lighting periodicity) [7]. A cohort of follicles begins to develop near the end of proestrus and grows at a nearly constant rate until the onset of the subsequent estrus, at which point the growth rate increases approximately eightfold. Ovulation occurs about 109 hours after the initiation of follicular growth [6].

There is a growing global dependence on native medicinal plants for the treatment of varying ailments [8, 9]. It is estimated that a large proportion of the population in developing countries relies heavily on medicinal plants and the services of traditional medicine practitioners [8, 9, 10]. The scientific value of medicinal plants stems from their accessibility, affordability, relatively fewer side effects, and other advantages compared to conventional orthodox medicines [11, 12]. Many medicinal plants contain bioactive compounds that are effective in the treatment, management, and prevention of various disease conditions [13]. These compounds are frequently isolated and characterized, serving as raw materials for the development of pharmaceutical drugs [8, 13]. C*raterispermum schweinfurthii* is distributed widely across Africa [14, 15]. The species consists of shrubs or small trees characterized by axillary inflorescences that are paired at the nodes. In traditional medicine, the seeds, leaves, and inner bark of C*raterispermum schweinfurthii* are used to treat stomach ailments, ulcers, infertility, anaemia, diabetes, and fever [16]. Despite these widespread anecdotal claims, only a few scientific studies have experimentally investigated the pharmacological effects of its leaves. Therefore, this study aims to evaluate the potential modulatory effects of the hydromethanol leaf extract of C*raterispermum schweinfurthii* on the female reproductive cycle using Wistar rats as experimental models.

Methods

Collection, identification and extraction of plant materials

Fresh leaves of *Craterispermum schweinfurthii* were obtained from the University of Port Harcourt Botanical Garden. Dr. Chimezie Ekeke of the Department of Plant Science and Biotechnology, University of Port-Harcourt, Nigeria identified and authenticated the specimen and assigned a reference code; UPH/V/296. Voucher specimen was subsequently deposited in the University Herbarium for future reference. The plant leaves were gathered, and all extraneous materials carefully removed. Leaves were air dried at room temperature for about 7 days and subsequently pulverized into powder and a weighed quantity of 670.6 g dissolved using Soxhlet device in 390ml of water-methanol mixture (25:75% v/v BDH) for three days in a jar. The solution was filtered and concentrated using a rotary evaporator at 40°C with a percentage yield of 73. Obtained extract was preserved in airtight containers and stocked at room temperature before administration.

Procurement and handling of experimental animals

Female Wistar rats weighing between 100-250 g were used for the study, which were acquired from the Faculty of Basic Medical Sciences Animal House, College of Health Sciences, University of Port Harcourt. Rats were placed in different compartments of the cage, one for each experimental group and cared for under standard laboratory conditions. Wood shavings and beddings were changed routinely to prevent possible infection due to untidy beddings.

Statement of ethics and acute toxicity studies

Ethical approval was obtained from the University of Port Harcourt Ethical Committee through a communication referenced: UPH/CEREMAD/REC/MM82/024 and dated 23rd November 2021. Acute toxicity was determined using Karber's method as modified by Aliu and Nwude [17]. Experimental rats were randomly divided into five groups of five animals each. The animals received oral doses of 0.5 g, 1 g, 2 g, 3 g, and 4 g of the extract, respectively, and were observed for 3 days for various physiological and clinical signs. Animals in the first two groups (0.5 g and 1 g) remained normal and active throughout the observation period. Slight weakness was noted in group 3 (2 g). In groups 4 and 5 (3 g and 4 g), the rats exhibited depression, sluggishness, anorexia, weakness, neurological deficits, and significant mortality. Following probit analysis, the median lethal dose (LD_{50}) of the extract was determined to be 3968 mg/kg body weight. Doses between 0.1 g and 2 g were considered safe. The study was carried out following standard guidelines for the care and use of laboratory animals [18].

Research design

A total of 20 female Wistar rats weighing between 100-250g were used for the study. After two weeks of acclimatisation, the rats were randomly divided into 4 groups of 5 rats each designated Groups A to D and treated as follows:

Group A: Negative control; received extract vehicle only.

Group B: Low dose extract; received 250mg/kgbw of the leaf extract of C*raterispermum schweinfurthi.*

Group C: Medium dose extract; received 500mg/kg bw of the leaf extract of C*raterispermum schweinfurthi.*

Group D: High dose extract; received 750mg/kg bw of the leaf extract of C*raterispermum schweinfurthi.*

Experimental animals were allowed free access to standard rat pellet and clean drinking water ad-libitum.

Determination of estrus cycle

Vaginal smears of each experimental rat were collected morning of every day of the study between 7 - 9 am West African Time for four weeks (2 weeks before the commencement of treatment and 2 weeks during treatment) as previously described by Mclean et al.

[19], Goldman et al. [20], Tropp and Markus [21] and Saronee et al. [22].

Statistical analysis

Statistical analysis was performed using SPSS to determine mean values and standard error of mean (SEM). Furthermore, an ANOVA test was conducted to determine the mean differences among all treatment groups, and finally, a post hoc test using LSD. Data were expressed as mean ± standard error of mean and a p-value (<0.05) was considered significant.

Results

In the table below, Significant reduction in proestrus, estrus, metestrus and diestrus were observed following administration of graded doses (250mg/kg, 500mg/kg and 750mg/kg body weight) of the leaf extract of C*raterispermum schweinfurthi* amongst Groups B, C and D rats relative to control (Group A) rats ($p<0.05$). Demonstrating a possible fertility enhancing potentials of the extract in female Wistar rats

Discussion

The estrous cycle in mice and rats lasts 4–5 days, making them highly suitable models for studies on reproductive physiology [20]. The different phases of the cycle are characterized by specific vaginal epithelial cell types: nucleated epithelial cells predominate during proestrus, cornified cells during estrus, leukocytes during diestrus, and a mixture of these cell types during metestrus [20, 23, 24]. Results from the present study revealed a significant reduction ($p < 0.05$) in the mean duration of the estrous cycle in all treatment groups compared with the control group. The mean estrous cycle duration in the control group was notably longer than that observed in the treatment groups. This finding is consistent with previous reports by Saronee et al. [22], Marcondes et al. [25], Saronee et al. [26], and Nwafor et al. [27]. Groups B, C, and D, which received 250 mg/kg, 500 mg/kg, and 750 mg/kg body weight of the extract, respectively, showed a significant and largely dose-dependent reduction in both the duration and mean values of the estrous cycle. These results suggest potential therapeutic efficacy of the intervention. Furthermore, analysis of the data indicated notable alterations in the phases of the estrous cycle across the various treatment groups, pointing to a highly significant overall effect. Shortening of the cycle implies modulation of the hypothalamic–pituitary–ovarian axis, ovarian steroidogenesis, follicular dynamics, or local uterine/ovarian signalling. Similar effects have been reported for other plant extracts that regularize or accelerate cyclicity and improve fertility indices in rodents [25, 26, 27].

Table 1. Values of proestrus, estrus, metestrus and diestrus following administration of C*raterispermum schweinfurthi* leaf extract

Groups	Proestrus	Estrus	Metestrus	Diestrus
A: Control	1.14±0.13	3.34±0.08	2.75±0.01	5.10±0.01
B: 250mg/kg Extract	0.93±0.00[a]	2.89±0.02	2.24±0.16[a]	2.60±0.20[a]
C: 500mg/kg Extract	0.44±0.72[a]	2.07±0.12[a]	0.70±0.10[a]	3.51±0.03[a]
D: 750mg/kg Extract	0.34±0.01[a]	1.06±0.13[a]	0.95±0.01[a]	1.80±0.02[a]

Values are shown as Mean ± SEM; n=5; [a] Significant at P<0.05 compared to control.

Conclusion

In conclusion, the extract appeared to have caused a pro-fertility effect by shortening the reproductive cycle in the experimental rats. These findings therefore support the traditional use of this plant in folk medicine within our region. The extract could be explored as a candidate for conditions involving irregular or prolonged cycles, anovulation, or reduced fertility. It contributes to the broader search for plant-derived agents that influence ovulation frequency or ovarian function. Though, our observation is preliminary, but it encourages systematic phytochemical work and rational prioritization of the plant for further study.

Abbreviations

LD_{50}: Median Lethal Dose (Lethal Dose Required to Kill 50% of the Test Population)
LSD: Least Significant Difference
ORCID: Open Researcher and Contributor ID
SEM: Standard Error of the Mean
SPSS: Statistical Package for the Social Sciences
UPH: University of Port Harcourt
REC: Research Ethics Committee
CEREMAD: Centre for Research Management and Development

Authors' Contribution

This work was carried out in collaboration among all authors. SF designed the study, performed statistical analysis and wrote the protocol. KAO and TAK ran all laboratory analysis, managed the literature searches and wrote the first draft of the manuscript. All authors cross-checked and approved the final manuscript

Acknowledgments

We acknowledge the guidance and technical assistance given to us throughout the study, courtesy of Profs. D. V. Dapper and F. S. Amah-Tariah.

Conflict of interest

The authors declare no conflict of interest

Article history:

Received: 23 April 2026
Received in revised form: 07 August 2026
Accepted: 09 November 2026
Available online: 09 November 2026

References

1. Hill MA. 2021. Embryology *Estrous Cycle*. Retrieved from https://embryology.med.unsw.edu.au/embryology/index.php/Estrous_Cycle.
2. Bronson FH. 1989. Mammalian Reproductive Biology. University of Chicago Press, Chicago, IL, USA.
3. Llera R, Yuill C. 2021. Estrous Cycles in Dogs. *VCA Hospitals*. Retrieved April 6.
4. Griffin B. 2001. Prolific Cats: The Oestrous Cycle (PDF). Compendium on Continuing Education for the Practicing *Vet*. 23(12): 1049–1056.
5. Marcondes FK, Bianchi FJ, Tanno AP. 2002. Determination of the oestrous cycle phases of rats: some helpful considerations. *Bra J Bio.* 62 (4A): 609–614.
6. McCracken JA, Custer EE, Lamsa JC. 1999. Luteolysis: A neuroendocrine- mediated event. *Physio Rev.* 79(2): 263– 323.
7. Freeman Marc E. 1994. The Neuroendocrine control of the ovarian cycle of the rat". In Knobil, E.; Neill, J. D. (eds.). The Physiology of Reproduction. Vol. 2 (2nd ed.). Raven Press.
8. Iwu M. 1993. Handbook of African Medicinal Plants. (32-38). CRC Press Boca Raton, FL.
9. Saronee F, Kolawole TA, Amieye BD, Amadi NG, Amadi JE, Buduburisi RB and Dapper DV. 2024. Antioxidant and anti-inflammatory functions of cucumber juice following lead-induced toxicity in male Wistar rats. turmeric and *Res Bio Sci.* 7(10): 21- 31.
10. Saronee F, Okari K, Nwikue G, Dimkpa VO, & Ante IA. 2025. Influence of Craterispermum schweinfurthi Leaf Extract on Glucose Homeostasis in male Wistar rats. GPH-Inter J Health Sci Nur, 8(2), 1-9.
11. Chilpa R, Baggio CH, Solano AD, Muniz E, Kauffman FC, Sanchez RI. 2006. Inhibition of gastric H+, K+-ATPase activity by flavonoids, coumarins and xanthones isolated from Mexican medicinal plants. *J Ethnopharmacol.* 10(5):167-172.
12. Saronee F, Bekinbo MT, Ojeka SO, Dapper D.V. 2019. Comparative assessment of methanolic extracts of hog plum (Spondias mombin Linn.) leaves and turmeric (Curcuma long L.) rhizomes on blood glucose and glycosylated haemoglobin in male wistar rats. *J Appl Sci Environ Mgt.* 23(9):1631- 1636.
13. Saronee F, Amah-Tariah FS, Chinko BC and Dapper DV. 2023. GC-MS and proximate analysis of the hydromethanol extract of *Craterispermum schweinfurthi* leaves. *South Asian Res J Nat Pro.* 6(2):101-109.
14. Taedoumg HBP, Hamon P, Sonké B. 2012. Craterispermum parvifolium and C. robbrechtianum spp. nov. (Rubiaceae) from west central Africa. *Nord J Bot.* 29(9):700–707.
15. Taedoumg HBP. 2017. Craterispermum capitatum and C. gabonicum (Rubiaceae): two new species from the Lower Guinean and Congolian Domains.*Phytokeys.* 57(3):236-242.
16. Sofowora A. 1993. Screening Plants for Bioactive Agents. In: Medicinal Plants and Traditional Medical in Africa. 2nd Edition, (pp134-156). Spectrum Books Ltd., Sunshine House
17. Aliu AY. and Nwude N. 1982. Vet. Pharmacology and Toxicology Experiments. (pp104 – 109). Baraka Press, Nigeria Ltd, Zaria
18. (US) NRC. Committee for the Update of the Guide for the Care and Use of Laboratory Animals2019. Available from: https://www.ncbi.nlm.nih.gov/books/NBK54050/ downloaded on the 20th of June, 2022.
19. Mclean AC, Valenzuela N, Fai S, & Bennett SAL. 2012. Performing Vaginal Lavage Crystal Violet Staining, and Vaginal Cytological Evaluation for Mouse Estrous Cycle Staging Identification. 15;(67):4389.
20. Goldman JM, Murr AS, Cooper RL. 2007. The rodent estrous cycle: characterization of vaginal cytology and its utility in toxicological studies. Birth Defects Research Part B. *Dev Repro Tox.* 80(2):84-97.
21. Tropp J, & Markus EJ. 2001. Effects of mild food deprivation on the estrous cycle of rats. *Physio Behav.* 73(4):553-559.
22. Saronee F, Okari K, Kolawole TA. 2026. Phytosterol Promotes Ovulatory Functions in Wistar Rats by Shortening the Estrous Cycle. *GPH-Inter J Bio Med Sci.* 9(2):1-8.
23. Maeda SK, Ohkura S, Tsukamura H. 2000. Physiology of reproduction. In: Krinke GJ, editor. The Laboratory Rat: Handbook of Experimental Animals. London, UK: Academic Press. (pp145-176).
24. Westwood FR. 2008. The female rat reproductive cycle: A practical histological guide to staging. *Tox Path.* 36(3):375-384.
25. Marcondes FK, Bianchi FJ, Tanno AP. 2002. Determination of the estrous cycle phases of rats: Some Helpful Considerations. *Bra J Bio.* 62(4A):609- 614.
26. Saronee F, Dan-Jumbo D, Azosibe P, Amadi JE. 2024. Modulatory Functions of Craterispermum schweinfurthi on the Hypothalamic-Pituitary-Gonadal Axis of Male Wistar Rats in Phenyl Hydrazine Induced Testicular Toxicity. *J Comp Alter Med Res.* 25(4): 31-39.
27. Nwafor CC, Ifedi IC, Inyang EP, Uvoh SM, OgwihI OB, and Ebiyemzi AB. 2021 Fleurya Aestuans Leaves and Tetrahydroxyflavone Mitigate Lead Induced Testicular Toxicity in Wistar Rats. *J Adv Med Pharm Sci*. 23(10): 35-47.