

Original Article

Effect of Aqueous Extracts of *Gnetum africanum* and *Talinum fruticosum* on the Uterus of Adult Female Wistar Rats

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ABSTRACT: *Background:* The uterus is a hormonally responsive organ essential for female fertility. In many African communities, leafy vegetables such as *Gnetum africanum* and *Talinum fruticosum* are widely consumed for their perceived reproductive benefits. Despite their extensive dietary and ethnomedicinal use, scientific evidence on their direct effects on uterine structure remains limited. This study investigated the effect of aqueous extracts of *G. africanum* and *T. fruticosum* on the uterus of adult female Wistar rats when administered individually and in combination.

Materials and Methods: Twenty-four adult female Wistar rats weighing 150–200g were assigned to four groups (n = 6) based on their weights. Control was given distilled water and feed only, group 2 *T. fruticosum* extract, group 3 *G. africanum* extract, and group 4 a combination of *G. africanum* + *T. fruticosum* extracts. Extracts were administered orally for 12 weeks at a dose of 700mg per kg for the first 2weeks and thereafter increased to 1000mg per kg until the end of the 12weeks. The lethal dose was 5000mg per kg body weight. At the end of treatment, uteri were excised, weighed, and processed for histological analysis using hematoxylin and eosin staining. Histological assessment focused on endometrial thickness, uterine gland morphology, epithelial integrity, and inflammatory changes, while histomorphometry took into consideration parameters like cell count, total area, average size, percentage area, and perimeter of uterine cells.

Results: Animals in the control groups exhibited normal uterine histoarchitecture. While, *T. fruticosum*-treated group demonstrated mild uterine alterations, including reduced endometrial thickness and cytoplasmic vacuolation, suggestive of altered proliferative activity. The *G. africanum*-treated group showed visible uterine changes characterized by endometrial attenuation, uterine gland atrophy, and moderate inflammatory cell infiltration. Combined administration of both extracts resulted in moderate uterine responses, with preservation of glandular architecture and features indicative of a variant proliferative state.

Conclusion: Aqueous extracts of *G. africanum* and *T. fruticosum* exerted distinct effects on uterine histology in adult female Wistar rats. While *T. fruticosum* showed mild to moderate effects, *G. africanum* induced suppressive and inflammatory uterine changes. Combined exposure appeared to attenuate extreme structural alterations. From the histomorphometric study, the data lend scientific support to the claims that *Gnetum africanum* can act as a natural contraceptive agent in females, as evidenced by the reduced uterine glandularity. Extracts contain bioactive phytochemicals capable of reducing uterine glands and altering pituitary gonadotrophin signalling, a mechanism that could reduce fertility by impairing implantation or uterine support of early embryo. These findings provide experimental support for traditional claims and highlight the reproductive implications of dietary leafy vegetables, especially of *Gnetum africanum*.

KEYWORDS: Uterus, *Gnetum africanum*, *Talinum fruticosum*, Female Wistar rats, Fertility.

1. INTRODUCTION

The uterus is a muscular, hollow organ in the female pelvis that is approximately 5 cm wide, 8 cm long, and 4 cm thick with a volume of 80 to 200 mL (Gasner and Aatsha, 2023; Liedl et al., 2024). A physiologically normal uterus typically lies in a position of anteversion (tilts forward at the cervix) and anteflexion (tilts forward at the isthmus) (Gasner and Aatsha, 2023; Abdelrazek et al., 2025). The uterus plays a critical role in female fertility by supporting implantation, pregnancy, and parturition (Das et al., 2023). Its structural integrity and functional competence depend on the coordinated actions of estrogen and progesterone, which regulate cyclical changes in the endometrium, uterine glands, and myometrium. Disruptions in uterine morphology or hormonal signaling can impair endometrial receptivity and result in infertility, abnormal bleeding, or implantation failure (Vujovic et al., 2021; Shan et al., 2023). In many African communities, traditional leafy vegetables are consumed not only for nutritional value but also for their perceived medicinal and reproductive benefits (Moyo et al., 2021; Shayanowako et al., 2021). *Gnetum africanum* and *Talinum fruticosum* are two widely consumed indigenous vegetables, commonly eaten singly or in combination (Abdulrasheed-Adeleke et al., 2021; Ojmelukwe, 2022). *Talinum fruticosum* (waterleaf) is traditionally believed to enhance fertility by improving uterine blood flow and supporting ovulation (Khadim et al., 2024; Osawaru et al., 2024; Brahmi et al., 2025), while *Gnetum africanum* (afang/okazi) has been associated with menstrual irregularities and possible contraceptive effects. Despite their widespread use, scientific evidence validating these

claims, particularly with respect to uterine structure, remains limited. Phytochemical analysis of both plants reveals the presence of bioactive compounds such as flavonoids, saponins, alkaloids, carotenoids, and phenolic compounds, many of which are known to possess estrogenic, anti-estrogenic, or anti-inflammatory properties (Dada et al., 2021; Madubogwu et al., 2025; Manikandan et al., 2025). These compounds have the potential to modulate uterine architecture either by promoting endometrial proliferation or by suppressing glandular development and inducing inflammatory changes. Given the uterus's sensitivity to hormonal and phytochemical modulation, it represents a key target organ for evaluating the reproductive effects of dietary plant extracts.

Animal models, particularly female Wistar rats, provide a reliable platform for investigating uterine responses due to their well-characterized estrous cycle and histological similarity to the human uterus. Therefore, this study was designed to evaluate the effect of aqueous extracts of *Gnetum africanum* and *Talinum fruticosum*, administered individually and in combination, on the uterine histology in adult female Wistar rats. Understanding these effects is essential for providing scientific insight into traditional dietary practices and their implications on female reproductive health.

2. METHODOLOGY

2.1. EXPERIMENTAL ANIMALS

Twenty-four (24) adult female Wistar rats weighing between 150 and 200 g were used for this study. Animals were obtained from the animal house of the Faculty of Pharmaceutical Sciences, University of Port Harcourt, Rivers State, Nigeria. Rats were housed under standard laboratory conditions with a 12-hour light/dark cycle and had access to food and water.

2.2. EXPERIMENTAL DESIGN

The rats were assigned to four groups based on similar weights (n = 6 per group)

- Group I: Control (distilled water)
- Group II: *Talinum fruticosum* aqueous extract only.
- Group III: *Gnetum africanum* aqueous extract only.
- Group IV: Combination of *G. africanum* + *T. fruticosum* aqueous extracts

Extracts were administered orally at 700mg per kg body weight, and 2weeks later increased to 1000mg per kg body weight daily for a period of twelve (12) weeks. The lethal toxic dose was 5000mg per kg, all consistent with the original study protocol.

2.3. ESTROUS CYCLE MONITORING

Estrous cycle phases were determined using the vaginal smear (cytology) method. Vaginal smears were collected daily, stained, and examined microscopically to identify proestrus, estrus, metestrus, and diestrus phases. This ensured proper assessment of reproductive cyclicity and uterine responsiveness.

2.4. TISSUE COLLECTION

At the end of the treatment period, rats were euthanized humanely. The uterus was carefully excised and immersed in a tube containing Bouin's fluid.

2.5. HISTOLOGICAL PROCESSING OF THE UTERUS

Fixed uterine tissues were dehydrated in graded alcohol, cleared in xylene, and embedded in paraffin wax. Sections of approximately 5 μ m thickness were cut using a microtome and stained with hematoxylin and eosin (H and E). Uterine sections were examined under a light microscope to assess uterine morphology, endometrial thickness, and glandular density.

2.6. HISTOMORPHOMETRY

Cell count histomorphometry of the uterine cells from different tissues in each group was counted and assessed using stereologic techniques.

2.7. ETHICAL APPROVAL

Ethical approval was obtained from the University of Port Harcourt Research Ethics Committee, with clearance number UPH/CEREMAD/REC/MM99/002.

3. RESULTS

TABLE 1 Descriptive Statistics of Uterine Histomorphometry (Stereology) Data

Groups	Source of variation between groups(ss)	Source of variation within groups (ss)	Df	Df	Ms	Ms	F	P-value	F-critical

RATS 1	266.31	4707.51	3	1687	88.77	2.79	31.831	5.312	i2.61
RATS 2	47.05	4901.79	3	1661	15.68	2.95	5.314	0.001*	2.61
RATS 3	72.09	6041.92	3	1984	24.03	3.05	7.891	3.118	2.61
RATS 4	16.07	5175.07	3	1917	5.36	2.69	1.984	0.114	2.61

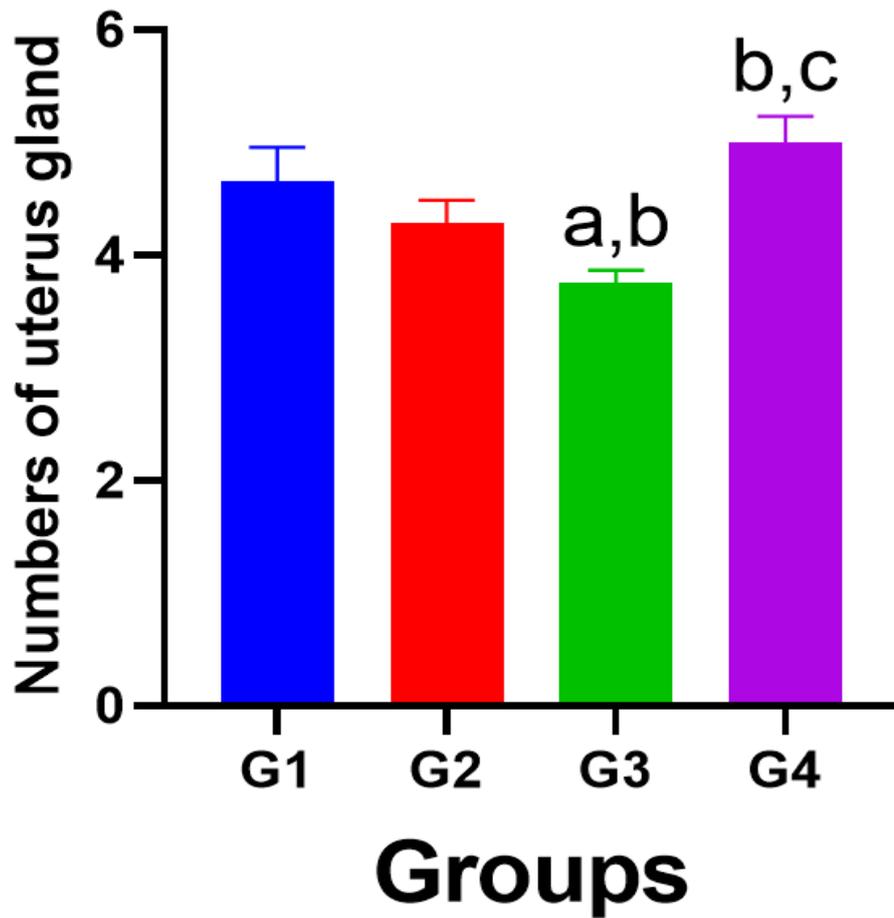


FIGURE 1 Histomorphometric quantification summary of cell count in the uterus

3.1. HISTOLOGY

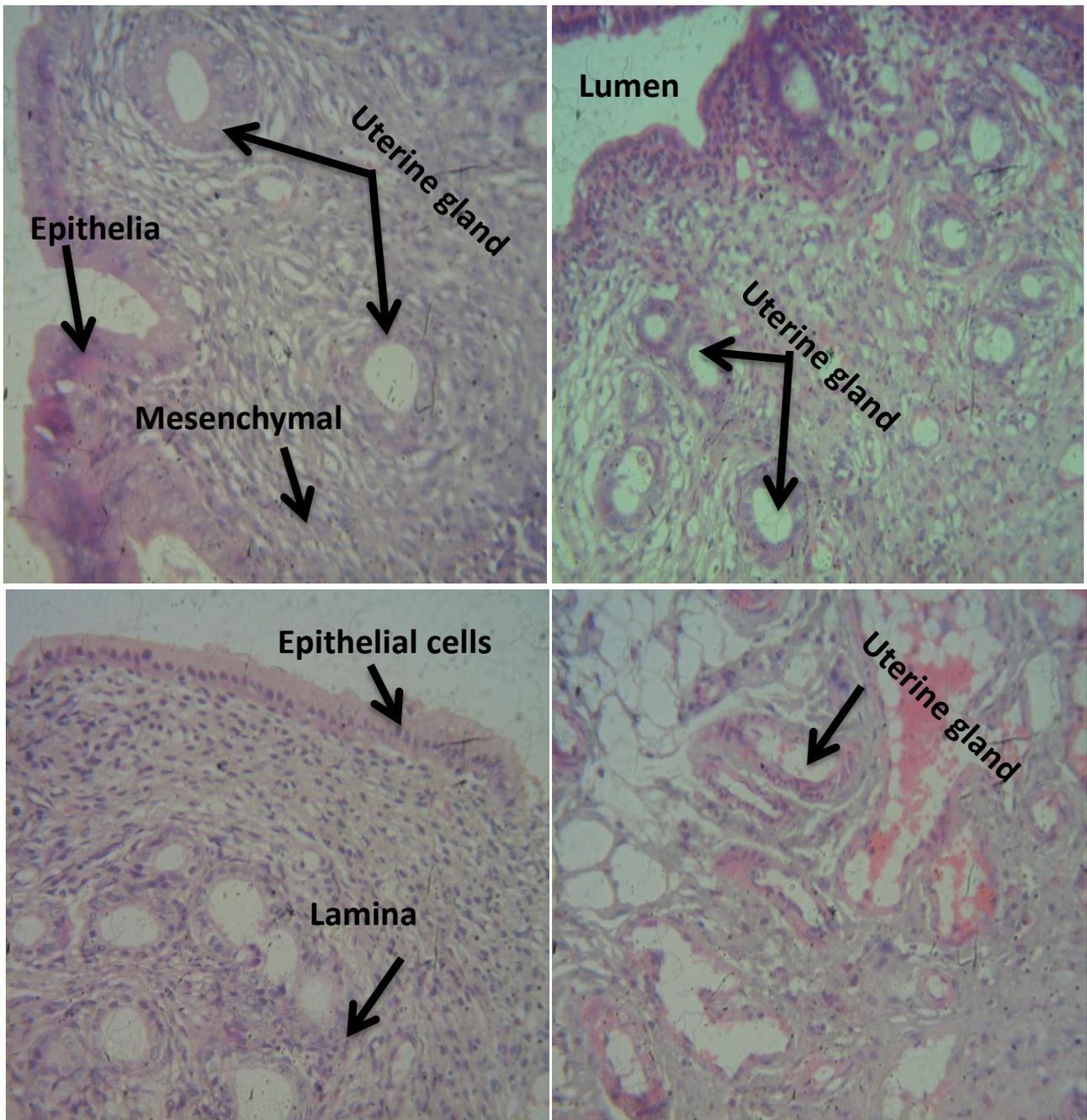


FIGURE 2 Day 21. Photomicrograph of the uterus (x400 Mag)

- (A) **Control:** Normal histology with the endometrium, uterine glands, supporting mesenchymal cells within the lamina propria, and epithelial cells lining the lumen.
- (B) ***T. Fructosum*:** Minimal distortion with reduced thickness of the endometrial wall.
- (C) ***G. Africanum*:** Decreased endometrial wall intensity with a well-delineated epithelial lining.
- (D) **Combination, *T. Fructosum* and *G. Africanum*:** normal variant proliferative stage with dilated uterine glands within the endometrium(arrows)

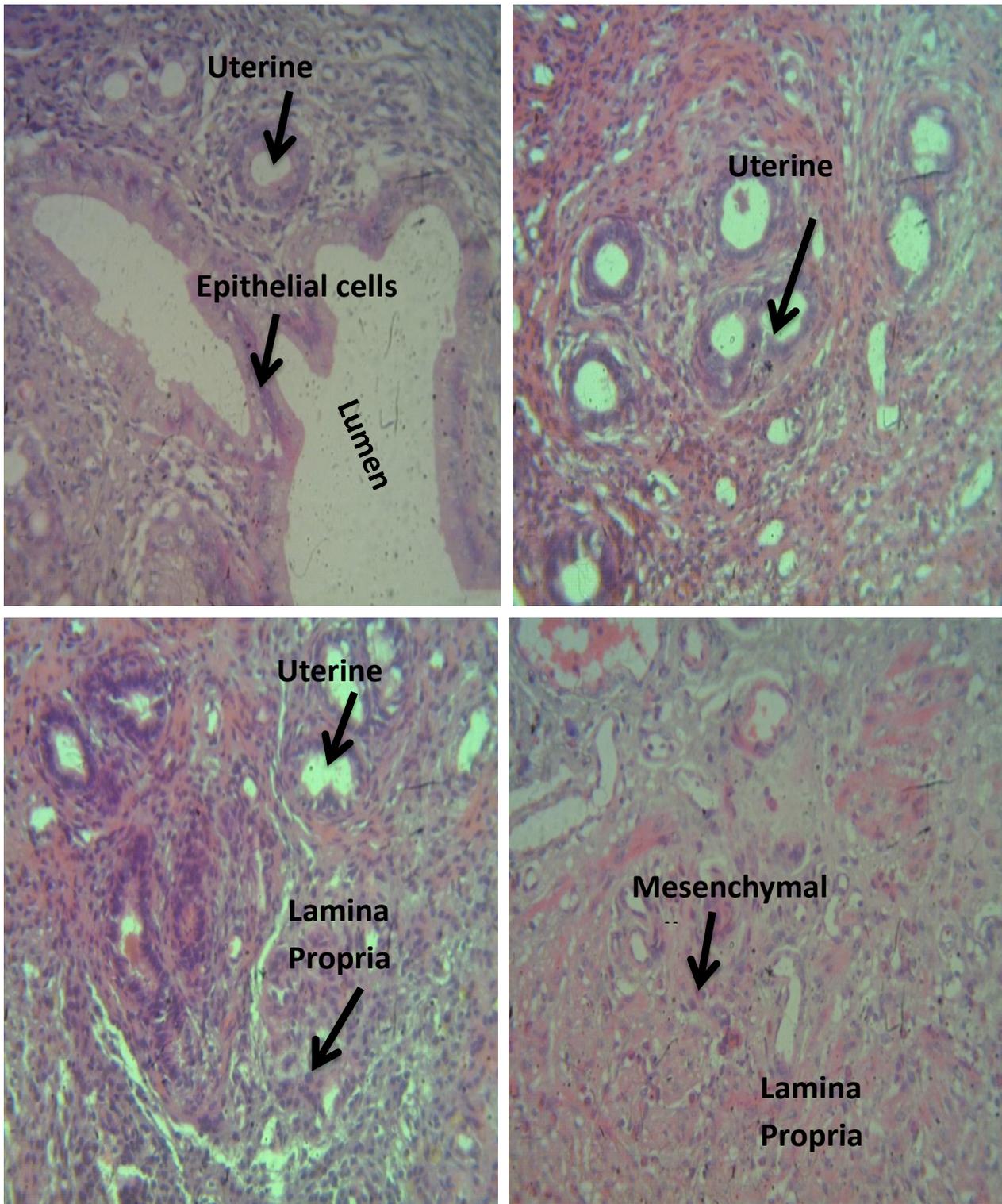


FIGURE 3 (Day 42) Photomicrograph of the Uterus (x400 Mag)

- (A) **Control:** Normal appearance with the epithelial lining of the lumen of the uterus, and a well-delineated uterine gland (arrows).
- (B) ***T. Fructuosum*:** Mild distortion with cytoplasmic vacuolation in the early proliferative stage.
- (C) ***G. Africanum*:** Moderate inflammation with mononuclear activities and vacuolation.
- (D) **Combination, *T. Fructuosum* and *G. Africanum*:** Mild distortion with reduced uterine glands.

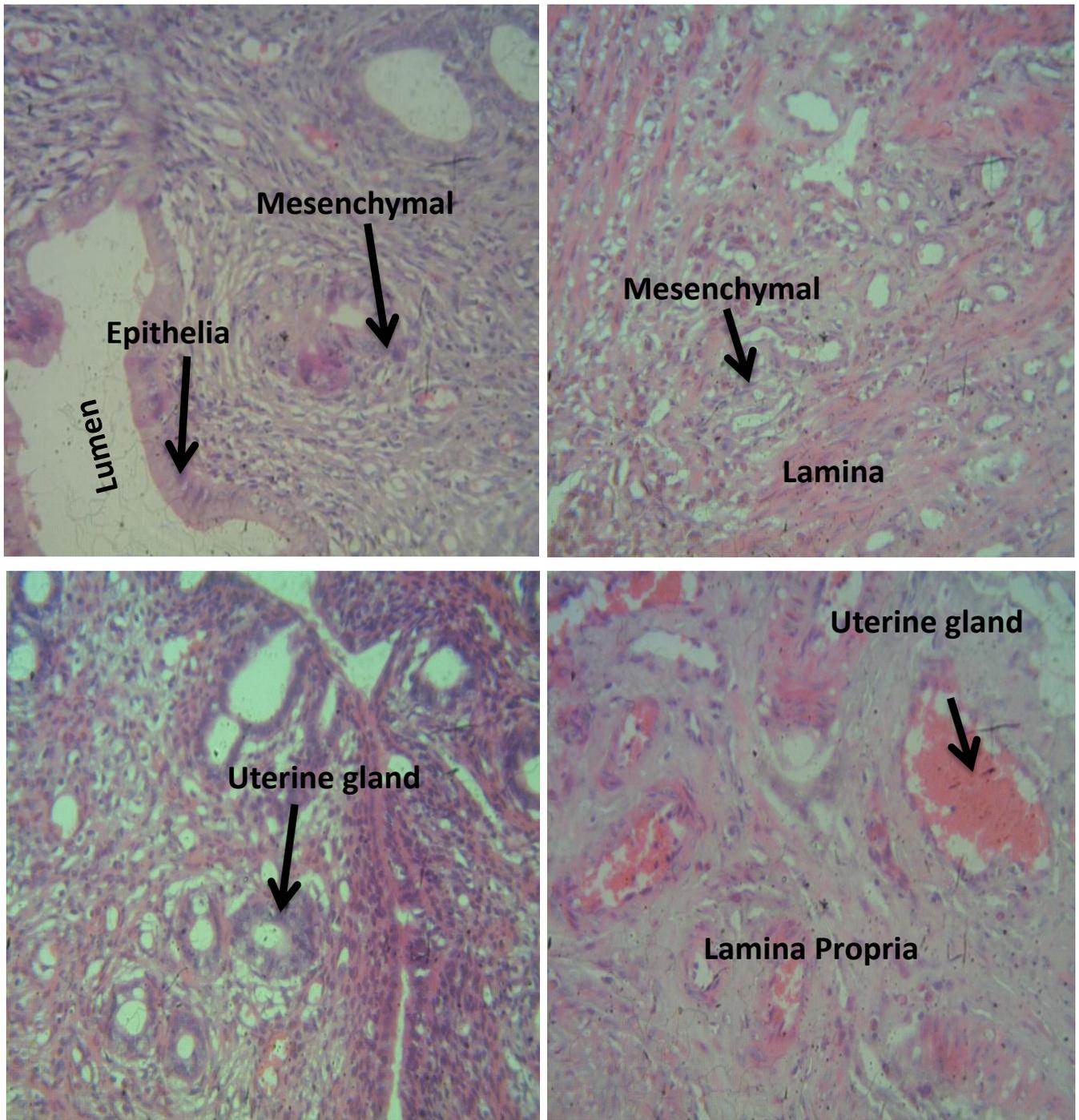


FIGURE 4 (Day 63) Photomicrograph of the Uterus. (x400 Mag)

- (A) Control:** Normal histology of the endometrium with the uterine glands, and supporting mesenchymal cells within the lamina propria and epithelial cells lining the lumen.
- (B) *T. Fructicosum*:** Mild distortion with cytoplasmic vacuolation and decreased thickness of the endometrial wall.
- (C) *Africanum*:** Moderate inflammation with cytoplasmic vacuolation of the endometrium (arrows).
- (D) Combination, *T. Fructicosum* and *G. africanum*:** proliferative stage with dilated uterine glands within the endometrium wall (arrows).

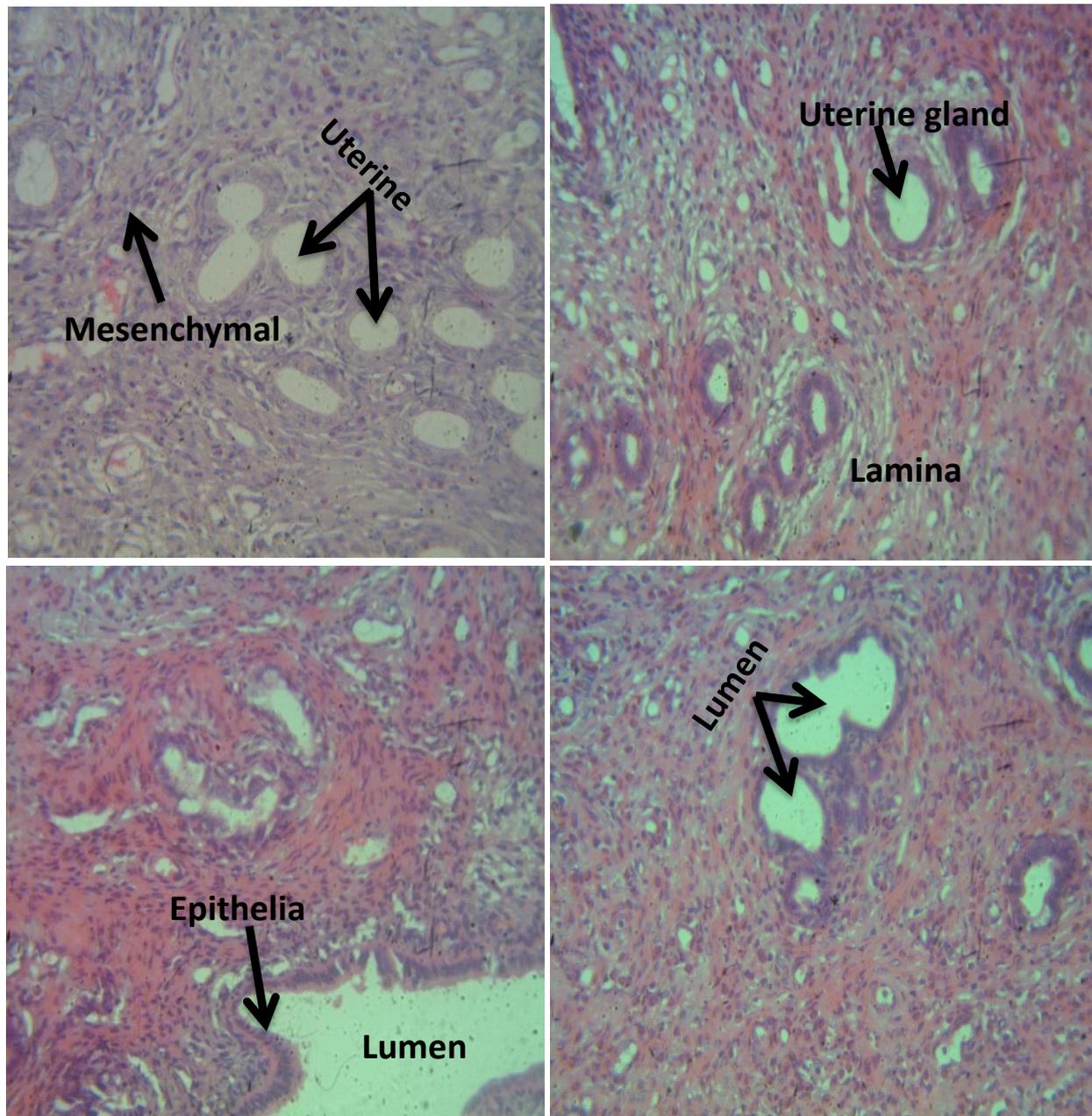


FIGURE 5 (Day 84) Photomicrograph of the Uterus. (x400Mag)

- (A) Control:** Normal structure of the endometrium with the uterine glands, mesenchymal cells within the lamina propria.
(B) *T. Fructuosum*: Minimal distortion with reduced thickness of the endometrial wall.
(C) *G. Africanum*: Decreased endometrial wall intensity with uterine gland atrophy.
(D) Combination, *T. Fructuosum* and *G. Africanum*: Mild distortion with dilated uterine glands within the endometrium (arrows).

4. DISCUSSION

The present study investigated the effect of aqueous extracts of *Gnetum africanum* and *Talinum fruticosum*, administered individually and in combination, on uterine histology in adult female Wistar rats. The findings demonstrate that both extracts exert distinct and duration-dependent effects on uterine architecture, affecting endometrial thickness, uterine gland morphology, epithelial integrity, and inflammatory status. These observations confirm that the uterus is a sensitive target organ for dietary phytochemicals, as evidenced from previous studies (Ayesha et al., 2023; Seetharaman et al., 2024).

At the early stage of treatment, uterine tissues from control groups demonstrated normal histological organization, which showed complete luminal epithelium, well-defined endometrium, and identifiable uterine glands. Treatment with *T. fruticosum* showed minimal histological alteration, thinning of the endometrium, reflecting an early modulatory effect on

endometrial proliferation. Furthermore, administration of *G. africanum* showed a reduction in the endometrial wall intensity with maintenance of epithelial continuity, reflecting possible inhibition of estrogen-driven growth of the uterus. Also, the co-administration of the two plant extracts showed characteristics of a normal variant proliferative stage, indicating a modulatory effect of the combination of the two plant extracts. With prolonged exposure, the uterine cells of the control group retained their normal epithelial and glandular structures. While in the *T. fruticosum*-treated group, there was a slight distortion with cytoplasmic vacuolation and changes indicative of altered proliferative activity. These might be attributed to the induction of uterine cellular metabolism and hormonal sensitivity by the uterus to phytochemicals present in the leaf extract. Administration of *G. africanum* catalyzed moderate inflammation, indicated by mononuclear cells and cytoplasmic vacuolation of the endometrium. Following an increase in treatment, the uterine responses of *T. fruticosum* became visible, with a consistent decrease in endometrial thickness and the onset of cellular vacuolation, indicating that with continued treatment, optimal endometrial development could be impaired. *G. africanum* persisted in stimulating inflammatory and suppressive uterine responses, such as endometrial thinning and decreased glandular activity. The combined extract group showed consistent moderation of uterine stimulation with maintained glandular structure and indicative activity of maintained proliferative activity. Results from the study showed that *G. africanum* produced marked endometrial reduction and uterine gland atrophy, changes commonly associated with impaired uterine functional capacity. *T. fruticosum* caused comparatively mild alterations. These findings suggest that phytochemical interactions between the two plants may attenuate adverse uterine effects when consumed together. This study provides experimental evidence that *T. fruticosum* exerts mild uterine modulatory effects, *G. africanum* induces suppressive and inflammatory changes, and combined exposure moderates these outcomes. The results further demonstrate the significance of evaluating traditional dietary plants within a reproductive health context and considering combined dietary exposure when assessing fertility-related outcomes.

5. CONCLUSION

This study demonstrates that aqueous extracts of *Gnetum africanum* and *Talinum fruticosum* significantly influence uterine histology in adult female Wistar rats. *Talinum fruticosum* administration was associated with mild structural alterations, including reduced endometrial thickness and cytoplasmic vacuolation, suggesting modulation of normal uterine proliferative activity. In contrast, *Gnetum africanum* induced more marked uterine changes characterized by endometrial attenuation, glandular atrophy, and inflammatory responses, supporting traditional assertions of its potential anti-fertility effects. Notably, combined administration of both extracts resulted in moderated uterine responses, with histological features indicative of a preserved or variant proliferative state. This suggests possible phytochemical interactions that may balance individual plant effects, reflecting real-life dietary practices in many African communities. These findings provide scientific evidence that commonly consumed leafy vegetables can alter uterine structure and potentially influence fertility outcomes. The study highlights the importance of careful dietary consideration for women of reproductive age and supports the need for further investigations into dose-dependent effects, hormonal correlations, and translational relevance to human reproductive health.

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