

Dynamism in Pro-inflammatory Cytokines and Hormonal Changes of Mice Induced Endometritis Treated with Aqueous Extract of *Asphilia Africana*

A. O. Oladejo^{1*}, O. M. Obisesan¹, P.T. Abegunde¹, O.A. Adeniyi¹, O. D. Adelakun¹, A. A. Bakre², and S. Abdulganiy¹

¹Department of Animal Health Technology, Oyo State College of Agriculture and Technology, Igboora, Nigeria,

²Department of Veterinary Medicine, University of Ibadan Nigeria.

Corresponding author email: oladejoayodele85@gmail.com; +2348135542620

ABSTRACT

Reproduction is one of the key pillars of the production in many herds and individual animal in female animal, reproduction can be affected by uterine disease including metritis and endometritis. To this effect, this study investigated the effect of *Asphilia africana* on the treatment of endometritis. Thirty mice were randomly assigned into 5 groups: control group, infectious group, treatment A, treatment B, treatment C. After acclimatization, the 4 treatment were induced endometritis with the use of lipopolysaccharide. (LPS 0.114 b3e.coli) through the vulva. *Asphilia africana* were administered to the mice from the low dosage (0.01ml) medium dosage (0.03ml) and high dosage (0.05ml) for the treatment of endometritis. At the end of the induction, blood sample was collected randomly from the mice to analyse the inflammatory cytokines and hormonal change using specific Enzyme-linked immunosorbent assay (ELISA) kit; data collected were analyze using analysis of the variance (ANOVA). Base on the finding of the study, it was concluded that the high dosage of *Asphilia africana* had great effect on induced endometritis of mice, hormonal changes and pro inflammatory cytokines and it could be recommended that the varying dosage of *Asphilia africana* could reduce the rate of reproductive hormone abnormalities and postpartum inflammation. Therefore could be used cautiously in therapeutic procedures.

Keywords: Mice, endometritis, *Asphilia africana*, Hormones, and cytokine



Article information

Received 5 June 2025

Accepted 30 August 2025

Published 18 September 2025

<https://doi.org/10.26765/DRJVMAS196268750>

Citation: Oladejo, A. O., Obisesan, O. M., Abegunde, P.T., Adeniyi, O.A. Adelakun, O. D., Bakre, A. A., & Abdulganiy, S. (2025). Dynamism in Pro-inflammatory Cytokines and Hormonal Changes of Mice Induced Endometritis Treated with Aqueous Extract of *Asphilia Africana*. Direct Research Journal of Social Science and Educational Studies. Vol. 10(1), Pp. 28-31. This article is published under the terms of the Creative Commons Attribution License 4.0.

INTRODUCTION

Endometritis is the inflammation of the endometrial lining without systemic signs, extending no deeper than the stratum spongiosum. It is associated with chronic postpartum infection of the uterus with pathogenic bacteria. (Bondurant, 1999). Endometritis negatively impacts fertility and it has been associated with brucellosis, leptospirosis, campylobacteriosis, and trichomoniasis. Often, Endometritis is the result of nonspecific infections, but it has been associated with calving-related disorders such as abortion, dystocia, retention of fetal membranes, and metritis in animals (Fabio Lima, 2022).

Endometritis is a risk factor for cystic ovarian disease, anoestrus and other reproductive disorders (Grohn, 1990). Along with retained placenta, puerperal metritis, pyometra and other non-specific infections of the uterus, it is one of the significant causes of declining fertility in cattle (Noake and Parkinson, 2001).

Endometritis is histopathologically subdivided into two categories (Kiviat *et al.*, 1990). One is acute endometritis, which is characterized by micro abscess formation and neutrophil invasion in the endometrial superficial epithelium, gland lumina, and uterine cavity. The other is chronic endometritis (CE), the histopathology features of which are endometrial superficial edematous change, high stromal cell density, dissociated maturation between epithelium and stroma, and infiltration of endometrial stromal plasmacytes (Greenwood and Moran, 1981).

Asphilia africana is one of such medicinal plants, which are fast gaining recognition. It is known by various names among the Nigerian populace (*Orangila in Igbo, Tozalin in Hausa and Yunyun in Yoruba*). The plant has been reported in literature to possess antimicrobial (Mounir *et al.*, 2017), anti-inflammatory (Okoli *et al.*, 2007) and anti-fertility (Eweka, 2007) activity. Therapeutic phytochemicals in *A. africana* include flavonoids, alkaloids, tannins, saponins, terpenoids, sterols, phenolic compounds, and glycosides. Essential oils from the leaves of the plant are rich in monoterpenes, sesquiterpenes, α -pinene, and germacrene, which are important therapeutic ingredients (Komakech *et al.*, 2018), broad range of antimicrobial and biological activities, including anti-inflammatory, haemostatic, oxytocic, gastroprotective, antiulcer, wound healing, anticancer, antihypertensive, and antidiabetic potentials may be attributed to these groups of active therapeutic components of the plant (Ahuchaogu *et al.*, 2018). Typical secondary metabolites in *Asphilia africana* that exhibit great anti-inflammatory potential include monoterpenes such as ascarene, terpenes including α -pinene, and sesquiterpenes such as β -caryophyllene and tannins among others.

MATERIALS AND METHOD

Experimental Site

The experiment was carried out at the Teaching and

Research Farm, Oyo State College of Agriculture and Technology, Igboora, which is located at 7°15'N and 3°30'E.

Experimental Design

The experimental design was a complete randomized design.

Procurement of Test Ingredient

The test ingredient was procured from a reputable pharmacy.

Experimental Animals and Management

A total of twenty-five mice was purchased from a reputable farm and kept in a plastic cage and fed for one week. The mice were allotted to four treatments and five mice each for other three treatments and the control.

Establishment of Endometritis

The mice were allotted to four treatments and one control; there were five mice for treatment one and five mice each for other three treatments and the control. They were placed in a plastic cage and allowed three (3) weeks acclimatization period. After acclimatization period, the four treatments were induced with lipopolysaccharide (LPS 0.114B3 *E. coli*), which will be deposited into the endometrium through the vulva and was left for 24 hours. After the 24th hour, at the first twelve (12) hour, incision was made through the endometrium in five (5) mice to check for improvement, development and growth of the induced lipopolysaccharide and also at the twenty-fourth (24th) hour, another incision was made through the endometrium in another five (5) mice to check for improvement, Development and Growth of the induced lipopolysaccharide in the first (1st) treatment.

Plant Extract and Animal Treatment

The leaf of *Asphilia Africana* was soaked and boiled for 48hrs to the required doses. Rotary evaporation and fractional sieving will be done. The filtrate was administered to the mice in low, medium and high doses range for seven days orally post endometritis establishment. Treatment two (2) was given a low dosage, treatment three (3) was given a medium dosage and treatment four was given a high dosage of aqueous extract of *Asphilia Africana*.

Sample collection

Blood sample was collected randomly from the ear veins and tails of the mice after the experiment. The blood sample was dropped in ETA bottle.

Parameters measured

The ELISA Techniques was used to measure both the hormones and cytokines.

Statistical Analysis

All data were analyzed and shown as mean \pm SEM based on at least three experiments. Data from different groups were tested for normality and homogeneity of variance. Multiple comparison analysis was performed on the significant factors to determine the optimal value. The differences between groups were compared by Student's t-test or one-way analysis of variance (ANOVA). GraphPad Prism 8.4 (GraphPad Software, USA) was applied for statistical analysis and plotting. A p-value <0.05 or <0.01 was considered statistically significant.

RESULTS AND DISCUSSION

The specific pro-inflammatory cytokines of the Endometritis induced mice in different experimental groups (Mean \pm SE)

Table 1 shows the result of specific pro inflammatory cytokines of the endometritis induced in different experiment group (mean \pm se); control, infectious group, treatment A, treatment B, treatment C. There were no significant different, the IL $-\beta$ value / amount was higher in infectious group, it have lowest value in treatment C but lower in treatment D and low in treatment A. The value of IL-6 in control is lower than the value in infectious group. In treatment A with low in dosage, the value of IL-6 reduced and also reduced in treatment b treated medium dosage but in treatment C treated with high dosage, the value of IL-6 is higher compared to the value in treatment A and B. IL-8 have value of 56.55 ± 2.63 in infectious group the value of 42.34 ± 1.32 in control, the value /amount of IL-8 appear to be lowest in treatment C IL-8 increased in infectious group compared to the value in treatment A of 45.34 ± 1.02 IL-8 decreased as there is increase in dosage of experiment treatment (*Asphilia africana*). TNF $-\alpha$ value in control is lower than the value in infectious group, TNF $-\alpha$ increased in treatment A has the lowest value in treatment C. TNF $-\alpha$ has equal value in treatment C and in the control despite the fact the treatment C was treated with high dosage but it has highest value in treatment A treated with low dosage (Table 1).

The specific hormonal changes of the Endometritis induced mice in different experimental groups (Mean \pm SE)

Table 2 shows the result of specific hormonal changes of the endometritis induced in different experiment group (mean \pm se); control, infectious group, treatment A, treatment B, treatment C. In Follicle stimulating hormone, FSH have the highest value in infectious group thus, it value reduce to 6.2 ± 1.00 in treatment A FSH value

increased in value in treatment B with medium dosage and appear to be low in treatment C with higher dosage, the treatment c were significant difference to the control. Prolactin has highest value in treatment A but significant difference from infectious group. The value of prolactin in treatment A is higher than treatment B and has the lowest value in treatment C. The value of prolactin reduces in three treatments as the dosage increase. There is increase in progesterone in treatment B compared to the treatment A and in control. Progesterone appears to have lowest value in treatment C compared to value in infectious group. The value of estrogen in treatment C and infectious group slightly equal, thus, the level of oestrogen increased in treatment C reduce consequently in treatment A and B. The level of oestrogen in control is slightly equivalent to the value in treatment B. The level of luteinizing hormone in treatment A reduced compare to the infectious group which has the highest value. Luteinizing hormone reduced in treatment A compare to the control and infectious group, the level of luteinizing hormone reduced in treatment B compared to treatment A and also got increased in treatment C, there are significant different in treatment C from control (Table 2).

Endometritis negatively impacts fertility and it has been associated with brucellosis, leptospirosis, campylobacteriosis, and trichomoniasis. Often, Endometritis is the result of nonspecific infections, but it has been associated with calving-related disorders such as abortion, dystocia, retention of fetal membranes, and metritis in animals (Fabio Lima, 2022).

The extract of *Asphilia africana* has been reported to have an anti-inflammatory activity (Okoli *et al.*, 2007). Some studies have revealed that the process of hormonal changes is comparable to an inflammatory process (Epsy, 1994). Anti-inflammatory drugs have been employed in blocking some hormones and affect cytokines (Gaytan *et al.*, 2002). The anti-inflammatory property of *Asphilia africana* may be responsible for its observed effect in balancing FSH hormone, reducing prolactin hormone, increasing progesterone, increasing Estrogen and reducing luteinizing hormone.

It was revealed that all traditional non-steroidal anti-inflammatory drugs produce most of their effects by blocking some cytokines such as IL-8, IL-6 and IL- 1β . The cytokines deficient in mice suffer from make defect in reproductive functions such as prolactin that stimulates ovulation and progesterone that stimulates fertilization (Arunsi *et al.*, 2020) since, Prolactin is important in ovulation through its role as an essential enzyme for follicular rupture.

Falodun *et al.*, (2006) reported the oxytocic effects of a plant that has saponin, saponin glycosides, steroid, tannins, volatile oils, and alkaloids on the function of isolated uterus from female rats. Most of these compounds are also present in *Asphilia africana*. These effects are most probably due to imbalances in hormonal level caused by high level of saponins and other phytoestrogens. The thickness of endometrium varies considerably according to the individual's hormonal state (Spornitz, 1992).

Table 1: The specific pro-inflammatory cytokines of the endometritis induced mice in different experimental groups (Mean ± SE).

Parameters (pg/ml)	Control	Infected	Treatment A	Treatment B	Treatment C
IL-1B	32.13± 2.08	56.55± 2.63*	35.60± 1.72	33.34± 2.49	31.88± 2.43
IL-6	58.34± 1.32	75.25± 2.42*	55.34± 2.54	53.34± 2.19	57.86± 2.05
IL-8	42.34± 1.32	59.55± 1.32*	45.34± 1.02	43.34± 2.19	41.94± 2.22
TNF-a	100.36± 3.35	172.46± 9.24*	130.28± 4.52	101.47± 6.71	100± 8.32

Table 2: The specific hormonal changes of the Endometritis induced mice in different experimental groups (Mean ± SE).

Parameters	Control	Infected	Treatment A	Treatment B	Treatment C
FSH(iu/l)	6.6±0.51	7.0±2.35	6.21±1.00	7.1±2.09	6.5±0.89
PROL(iu/l)	10.0±1.00	11.0±1.87	12.0±1.64	10.0±1.64	7.0±2.13*
PROG(iu/l)	5.0±0.45	4.8±0.37	5.3±0.49	5.8±0.37	3.3±0.51*
ESTRO(iu/l C)	29.6±1.66	30.0±1.38	28.6±1.5	29.8±0.37	30.3±0.49
LU (iu/l)	3.4±0.24	3.8±08	2.9±0.9	3.3±0.32	3.3±0.32

The results of this research showed that methanolic extract of *Asphilia africana* leaves possess negative influences on the estrous cycle and histo-architecture of the uterus of rats suggesting negative influences on the reproductive health of the animals. The result of the study suggest that *Asphilia africana* may affect some reproductive hormones (FSH, Estrogen, Progesterone and Luteinizing hormone) by reducing their concentration at certain level in administering extract of *Asphilia africana*. The concentration we administer may have been much enough to affect the level of concentration of reproductive hormones especially prolactin hormone that stimulates ovulation.

In conclusion, these studies suggest that administration of aqueous extract of *Asphilia africana* alters the reproductive hormones and pro-inflammatory cytokines. Based on this finding, it could be recommended that the varying dosage of *Asphilia africana* could reduce the rate of reproductive hormone abnormalities. The farmers are encouraged to use ethnoveterinary plant matter to treat endometritis with rapt precaution

REFERENCES

- Ahuchaogu, B. C. Onyekwere, E. U. G. I. Ogbuehi, G. U. Ihenetu, and J. B. O., (2018). Echeme, "Chemical constituents of methanol leaf extract of *Asphilia africanac*. d. adams by GC MS," *International Journal of Advanced Research in Chemical Science*, vol. 5, no. 10, pp. 21–29, 2018.
- Arunsi, U. O., Chinyere, G. C., Ngwogu, K. O., Ngwogu, A. C., Atasie, O. C., Oti, U. A., Akujuobi, J. K., Udeogu, C., & Ibe, C. (2020). Evaluation of the biochemical, haematological and histopathological parameters of female Wistar rats fed with aqueous and ethanol extracts of *Asphilia africana* leaves. *Journal of Herbal Medicine and Pharmacology*, 9(3), 257–267. <https://doi.org/10.34172/jhp.2020.33/jhp.2020.33>
- Bondurant, R.H. (1999). Inflammation in bovine female reproductive tract. *Journal of Animal Science*. 77: 101-110.
- Epsley, L.L., (1994). Current status of hypothesis that mammalian ovulation is comparable to and inflammatory reaction Biol. Reprod., 50:233-238.
- Eweka, A. O. (2008): Histological Studies of the Teratogenic Effects of Oral Administration of *Asphilia africana* (Asteracea) Extract on the Developing Kidney of Wistar Rats. *The inter. Journal of Toxicology*. 4 (2).
- Fabio Lima, (2022). Endometritis in Production Animals. DVM, MS, PhD, DACT, University of California, Davi. Last review/revision Aug 2022
- Modified Oct 2022.
- Falodun, A.; Nworgu, Z. A. & Ikponmwonso, M. O., (2006). Phytochemical Components of *Hunteria umbellata* (K. Schum) and Its Effect on Isolated Non-Pregnant Rat Uterus in Oestrus. *Pak. J. Pharm Sci*, 19(3):256-8, 2006.
- Gaytan, E. (2002). Morphological evidence for uncontrolled proteolytic activity during the ovulatory process in indomethacin-treated rats reproduction, 123:639-649
- Greenwood SM, Moran J J. (1981). Chronic endometritis: morphologic and clinical observations. *ObstetGynecol* 1981; 58:176–84.
- Grohn, Y.T. (1990). Epidemiology of reproductive disorders in dairy cattle. *Preventive Veterinary Medicine*. 8: 25-39.
- Kiviat NB, Wølner-Hanssen P, Eschenbach DA, Wasserheit JN, Paavonen JA, Bell TA, (1990). Endometrial histopathology in patients with culture-proved upper genital tract infection and laparoscopically diagnosed acute salpingitis. *Am J SurgPathol* 1990; 14:167–188,
- Komakech, R., M. G. Matsabisa, and Y.-M. Kang., (2018). "A review on the botanical aspects, phytochemical contents and pharmacological activities of *Warburgiaugandensis*," *Journal of Medicinal Plants Research*, vol. 12, no. 27, pp. 448–455, 2018.
- Mounir A, Rachid K, Christian H, Gary CW., (2017). Risk factors of clinical and Subclinical endometritis in cattle: A review. *Turkish Journal of Veterinary and Animal Sciences*. 2017; 41: 1-11. doi: 10.3906/vet-1603-63.
- Noakes, D.E. and Parkinson, T.J. (2001). *Arthurs Veterinary Reproduction and Obstetrics*. Harcourt Publishers Limited, the United Kingdom.
- Okoli, C. O., Akah, P. A. and Okoli, A. S. (2007). Potential of leaves of *Asphilia africana* (Composite) in wound care: an experimental evaluation. *Biomedical Centre Complimentary and Alternative Medicine*, 7(24): 101-109.
- Spornitz U. M., (1992). The Functional morphology of the human endometrium and deciduas. *Adv. Anat. Embryol. Cell.Biol.*, 124:1-99, 1992.