Lipid Profile and Antioxidant Status in Men's Sperm Fertility: A Cross-Sectional Study of Wistar Albino Rats Fed with Aqueous Extract of Brachistegia Eurycoma (Achi) Seeds

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ABSTRACT

Infertility is a common reproductive health issue, with male factors responsible for 40–50% of cases. This research investigated the impact of aqueous *Brachystegia eurycoma* (achi) seed extract on lipid levels, antioxidant capacity, and sperm parameters in adult male Wistar rats. Twenty rats (75–130 g) were divided into four groups (n = 5) to receive daily doses of the extract: 0 mg/kg (control), 200 mg/kg, 400 mg/kg, or 800 mg/kg for 14 days. Body weights were recorded and semen samples were collected to evaluate sperm motility, morphology, and count. Serum lipid profiles were analyzed, and antioxidant activity was assessed using DPPH and FRAP assays. The 800 mg/kg dose significantly improved sperm parameters, showing 45% progressive motility and 75% normal morphology. In contrast, the 400 mg/kg dose resulted in 25% motility and 45% normal morphology. Lipid profile analysis indicated increased total cholesterol (299 mg/dL) and triglycerides (442 mg/dL) at the highest dose, with HDL at 33 mg/dL and LDL at 178 mg/dL. The extract demonstrated strong antioxidant activity, achieving 94.42% DPPH inhibition at 80 mg/mL and a FRAP scavenging capacity of 87.99%. Brachystegia eurycoma seed extract may enhance sperm quality, likely due to its antioxidant properties and regulation of lipid metabolism.

Keywords: Brachystegia eurycoma, Lipid metabolism, Oxidative stress, Sperm motility.

INTRODUCTION

In many cultures, children are highly valued, making infertility a significant issue with psychological, social, economic, and medical consequences. The World Health Organization (WHO) and the International Committee for Monitoring Assisted Reproductive Technology (ICMART) define infertility as the inability to achieve a clinical pregnancy after at least 12 months of regular, unprotected sexual intercourse. Male factor infertility refers to a man's inability to impregnate a fertile female partner and contributes to approximately 40–50% of infertility cases, affecting about 7% of men globally (Singh *et al*., 2024). This condition is often associated with oligozoospermia, asthenozoospermia, or teratozoospermia, reflecting impairments in sperm count, motility, and morphology respectively (Aiyeyika *et al.,* 2025). Although several epidemiological studies have successfully established the burden of male infertility, many remain largely descriptive and fail to elucidate underlying biochemical mechanisms in detail. Recent reports indicate a concerning global decline in sperm quality, attributed to environmental pollutants, poor dietary habits, and socioeconomic stressors (Igwe *et al.,* 2024); however, these studies often lack controlled experimental validation, limiting causal inference.

Oxidative stress has been consistently identified as a central mechanism in the pathogenesis of male infertility (Eyeghre *et al*., 2023; Gaznee *et al*., 2023). It results from an imbalance between reactive oxygen species (ROS) production and antioxidant defense systems (Nnaoma and Okeke, 2024). Experimental studies have demonstrated that excessive ROS can induce lipid peroxidation, DNA fragmentation, and mitochondrial

dysfunction in sperm cells, thereby impairing fertilization capacity. Malondialdehyde remains a widely accepted biomarker for lipid peroxidation, while antioxidant enzymes such as catalase, superoxide dismutase, and glutathione peroxidase play protective roles in maintaining redox balance (Eyeghre *et al*., 2023). Despite these advances, many existing studies focus on isolated oxidative markers without integrating lipid metabolism or systemic biochemical indices, thereby providing a limited understanding of the multifactorial nature of male infertility.

Current therapeutic approaches, including hormonal therapy and assisted reproductive technologies, have shown measurable success in improving fertility outcomes. Their strengths lie in targeted intervention and relatively predictable clinical outcomes. However, these methods are often associated with high cost, technical complexity, and limited accessibility in low- and middle-income countries.

Consequently, there has been increasing scientific interest in traditional herbal medicine as an alternative or complementary approach. The World Health Organization reports that about 88% of member states recognize traditional medicine, with approximately 75– 80% of Nigerians relying on it for primary healthcare (Ohemu *et al.,* 2024). While several plant-based studies have demonstrated improvements in sperm parameters and antioxidant status, many lack standardization in extraction methods, dosage optimization, and reproducibility, which limits their translational potential (Akbaribazm *et al.,* 2024).

Medicinal plants are rich in bioactive phytochemicals such as flavonoids, phenolics, alkaloids, and saponins, which have been linked to antioxidant and fertility-enhancing effects (Nnaoma *et al*., 2024; Nnaoma *et al*., 2025; Nath *et al*., 2026). Previous research has highlighted the pharmacological potential of several plant species in ameliorating oxidative stress-induced reproductive damage.

However, most studies have focused on well-known medicinal plants, leaving several indigenous species underexplored. Also, existing studies often evaluate reproductive parameters or antioxidant status independently, without examining their interaction with lipid metabolism, which is increasingly recognized as a critical factor in reproductive health.

Brachystegia eurycoma, a leguminous plant indigenous to tropical Africa (Anyachukwu *et al*., 2025), is widely consumed as a dietary thickener and possesses a diverse phytochemical profile. Its reported pharmacological properties include antioxidant, anti-inflammatory, antimicrobial, hypoglycemic, and hepatoprotective activities (Osiobe & Eraga, 2025). While these findings shows its therapeutic potential, there is a paucity of experimental data specifically investigating its role in male reproductive function. In particular, no comprehensive study has simultaneously evaluated its effects on both lipid profile and oxidative stress parameters in the context of male fertility, representing a significant gap in the literature.

This study therefore investigates the effects of *Brachystegia eurycoma* seed extract on male reproductive health, with a specific focus on serum lipid profile and antioxidant parameters in male Wistar albino rats. The key contribution of this work lies in providing an integrated biochemical assessment that links oxidative stress modulation with lipid metabolism, thereby offering a more holistic understanding of its fertility-enhancing potential.

By addressing the limitations of previous studies, this research contributes to the growing body of evidence supporting plant-based therapeutic strategies for male infertility. Also, this paper is organized as follows: the introduction outlines the background and research gap; the materials and methods describe the experimental procedures; the results present the findings; the discussion critically interprets the results; and the conclusion summarizes the implications and future research directions.

MATERIALS AND METHODS

This study was designed to evaluate the effects of *Brachystegia eurycoma* seed extract on male reproductive parameters, lipid profile, and antioxidant status in Wistar albino rats. Standard experimental procedures were employed, including plant extraction, animal handling, biochemical assays, and statistical analysis. All protocols were conducted under controlled laboratory conditions and in accordance with established ethical guidelines for animal research. The methodologies adopted in this study are described in detail under the following subsections.

Plant collection and preparation

Dried *Brachystegia eurycoma* seeds were bought from Eke-Onuwa market in Owerri, and were identified by a Botanist. The seeds were ground into fine powder using an electric blender. They were labelled and kept in an airtight container, after which they were subjected to extraction.

Extraction of Plant Material

Extraction was performed using the maceration method. 1 kg of the ground plant material was soaked in 3 L of distilled water for 3 days. It was then filtered using a clean muslin cloth to get the crude. The crude extract was concentrated on a vacuum rotary evaporator at 40 ºC, and evaporation was completed to dryness using a water bath at 60 ºC.

Animal Ethics Declaration

We hereby declare that all experimental procedures involving animals were conducted in accordance with the ethical standards and guidelines for the care and use of laboratory animals.

The study protocol was approved by the Committee of the Department of Biochemistry, Federal Polytechnic Nekede, Owerri, Imo State (approval reference no: FPN/SIAS/BCH/032) in compliance with the principles of the Protection of Animals used for Scientific Purposes.

Experimental animals

For this study, adult male Wistar rats weighing 75-130 g were used. The animals were procured from a local breeder in Aba and were acclimatised in an airy pen at room temperature, fed with standard rat feed, and given water *ad libitum* for two weeks. The NIH recommendations for the care and use of laboratory animals were followed in the experimental procedures.

Experimental design

Groups	Group label	Number of Rats	Treatment	Duration of treatment
1	Normal control	5	Feed and water	14 days
2	Test group 1	5	200 mg/kg b.w. plant extract	14 days
3	Test group 2	5	400 mg/kg b.w. plant extract	14 days
4	Test group 3	5	800 mg/kg b.w. plant extract	14 days

Body weight measurements and Sacrifice of animals

The body weights of the rats were taken before treatment and at the end of the experiment, using a Toploader weighing balance. At the end of fourteen days, a transverse incision was made through the ventral wall of the abdomen of each rat following a cervical dislocation. Blood samples were obtained through cardiac puncture and homogenised in plain bottles for hormonal assay estimation.

Semen analysis

Sperm samples collection

Each rat was euthanised by cervical dislocation, and its epididymis was harvested. Sperm samples were collected from the epididymal reserve at the caudal portion of the epididymis, and a smear of the same was prepared on the preheated glass slides for evaluation.

Microscopic Examination

The sperm quality of the rats was examined by counting the sperm cells, determining the percentage sperm motility and carrying out the morphological assessment of the sperm. The movement and swimming ability of

sperm (motility) was determined using a microscope, and the sperm cells were counted using a haemocytometer (Ashidi *et al.*, 2019).

Lipid profile analysis

Lipid analysis was performed on a fully automated analyser based on spectrophotometric principles using kits. The serum lipid profile was analysed on the same day as the collection of blood samples. Three laboratory measures were determined: Total cholesterol, HDL, and Triglycerides. From these three data points, LDL was calculated. According to Friedewald's equation (Friedewald *et al.*, 1972), LDL = Total cholesterol - HDL – (Triglycerides/5).

The blood samples were centrifuged at 3000rpm for five minutes to obtain the serum, which was then analysed for the lipid profile. The analysis of the different samples was performed using the auto-analyser according to the manufacturer's operational guidelines. Three different reagents were employed in the determination of the lipids: TC reagent for Total cholesterol, TGL reagent for Triglyceride and HDL precipitating reagent for highdensity lipoprotein determination. All the samples were analysed within thirty minutes of collection. *2.8. Antioxidant analysis*

DPPH scavenging activity

The modified method by Brand-Williams *et al*. (1995), reported by Nnaoma and Okeke (2024), was used to determine the DPPH scavenging ability of the extract. The parent solution was prepared by dissolving 24 mg of DPPH in 100 mL of methanol and stored at 20°C until required. The working solution was obtained by diluting the DPPH solution with methanol. A 3 ml aliquot of this solution was mixed with 100 µl of the sample at various concentrations. The reaction mixture was thoroughly shaken and stored in the dark for 15 minutes at room temperature. Absorbance was measured at 517 nm. The control (BHT) was prepared in the same manner, except that no crude extract was added. The scavenging activity was determined using the following equation:

DPPH scavenging activity = 100 x (AC – AS)/ AC.

AC = Absorbance of control

AS = Absorbance of sample

Ferric reducing antioxidant power (FRAP) assay

The aliquots of various concentrations of the standard and test sample extracts (10 to 80 mg/ml) in 1.0 ml of deionised water were mixed with 2.5 ml of (pH 6.6) phosphate buffer and 2.5 ml of (1%) potassium ferricyanide. The mixture was incubated at 50°C in a water bath for 20 min after cooling. Aliquots of 2.5 ml of (10%) trichloroacetic acid were added to the mixture, which was then centrifuged at 3000 rpm for 10 min. The upper layer of solution 2.5 ml was mixed with 2.5 ml distilled water and a freshly prepared 0.5 ml of (0.1%) ferric chloride solution. The absorbance was measured at 700 nm in a UV spectrometer (Systronic double beam-UV-2201). A blank was prepared without adding extract. Gallic acid at various concentrations (10 to 80 mg/ml) was used as a standard (Bhalodia *et al.*, 2013).

Determination of Total Phenolic Contents

The total phenolic content of the extracts was measured according to the Folin-Ciocalteu method adapted from Guchu *et al.* (2020). Briefly, the extract (1 ml) was mixed with 2 ml of Folin-Ciocalteu reagent, which was prepared by dilution with distilled water in a ratio of 1 :10 v/v, after which 1 ml of 20% sodium carbonate (Na2CO3) was added. The mixture was shaken for 20 seconds and incubated at 40°C for 30 minutes. Absorbance was measured at 765 nm. Gallic acid was used to generate the standard curve. The total phenolic content was expressed as mg of gallic acid equivalents (GAE) per ml of the studied extracts.

Determination of Total Flavonoid Contents

The total flavonoid content of the extracts was evaluated through a technique described by Guchu *et al.* (2020). In a 10 ml test tube, 0.3 ml of the extracts, 3.4 ml of 30% methanol, 0.15 ml of NaNO2 (0.5 M), and 0.15 ml of $AlCl_3$ (0.3 M) were mixed. After 5 minutes, 1 ml of NaOH (1 M) was added and mixed well, and the absorbance was measured against the reagent blank at 510 nm. The standard curve for total flavonoids was prepared using

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quercetin standard solution (0–100 mg/l). The total flavonoids were expressed as milligrams (mg) of quercetin equivalents per ml of sample.

Statistical analysis

Statistical analysis was performed for the data obtained using the IBM Statistical software for Social Science version 23 to determine the mean and standard deviation, and One-way ANOVA was done to determine their significant differences.

Flow Chart

Plant Collection and Identification

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Drying and Pulverization of Seeds

↓

Extraction (Aqueous Maceration → Filtration → Concentration)

↓

Preparation of Extract Doses

↓

Animal Procurement and Acclimatization (14 days)

↓

Experimental Grouping and Treatment (14 days)

↓

Body Weight Measurement

↓

Animal Sacrifice and Sample Collection (Blood + Epididymis)

↓

Semen Analysis (Count, Motility, Morphology)

↓

Biochemical Analysis:

- Lipid Profile (TC, HDL, TG, LDL)
- Antioxidant Assays (DPPH, FRAP, Phenolics, Flavonoids)

↓

Statistical Analysis (Mean ± SD, ANOVA)

↓

Interpretation of Results

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RESULTS

This section presents the findings of the study on the effects of *Brachystegia eurycoma* seed extract on male reproductive parameters, lipid profile, and antioxidant status in Wistar albino rats. Data obtained from the experimental groups are expressed as mean ± standard deviation and are presented in tables and figures where appropriate. Comparative analysis between control and treated groups was performed to evaluate dosedependent responses and statistical significance of the observed effects.

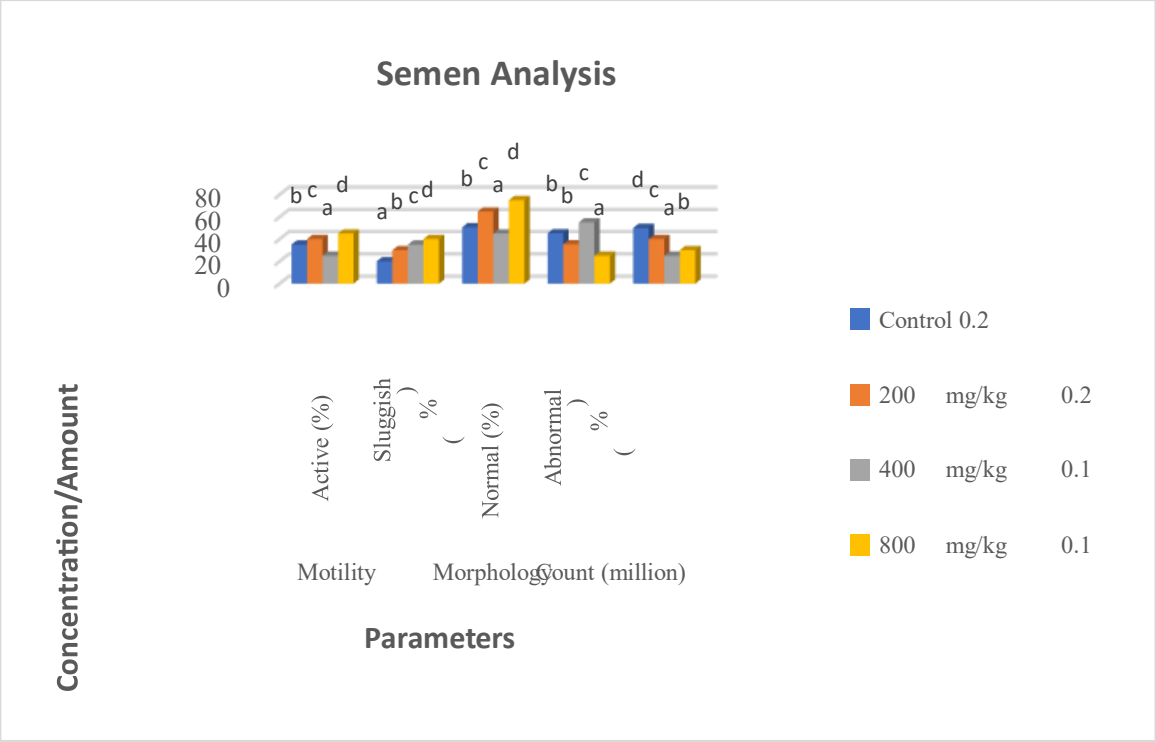
Effect of Brachystegia eurycoma extract on semen parameters of male Wistar rats

Table 1: Effect of Treatments on Semen Parameters

Parameter		**Treatment**			
		Control	**200 mg/kg**	**400 mg/kg**	**800 mg/kg**
Motility	Active (%)	35.0 ± 2.0^{b}	40.0 ± 2.0^{c}	25.0 ± 1.0^{a}	45.0 ± 1.0^{d}
	Sluggish (%)	20.0 ± 0.0^{a}	30.0 ± 0.0^{b}	35.0 ± 1.0^{c}	40.0 ± 0.0^{d}
Morphology	Normal (%)	50.7 ± 1.0^{b}	64.7 ± 1.5^{c}	45.0 ± 1.0^{a}	75.0 ± 1.0^{d}
	Abnormal (%)	45.2 ± 0.0^{b}	35.3 ± 1.5^{b}	55.0 ± 1.0^{c}	25.0 ± 1.0^{a}
Count (million)		50 ± 0.0^{d}	40.0 ± 2.0^{c}	25.0 ± 1.0^{a}	30.0 ± 1.0^{b}

Values are expressed as Mean ± SD. Values with different superscript letters in the same row differ significantly ($p < 0.05$).

Fig. 1. Bar chart showing the effect of treatments on the semen parameters of the experimental animals



The aqueous seed extract of Brachystegia eurycoma demonstrated a dose-dependent improvement in male fertility parameters. Sperm motility increased from 20% in the control group to 40% at a dose of 400 mg/kg, while the percentage of normally shaped sperm rose from 50.7% in controls to 75% at 800 mg/kg, suggesting that the extract positively influenced sperm quality.

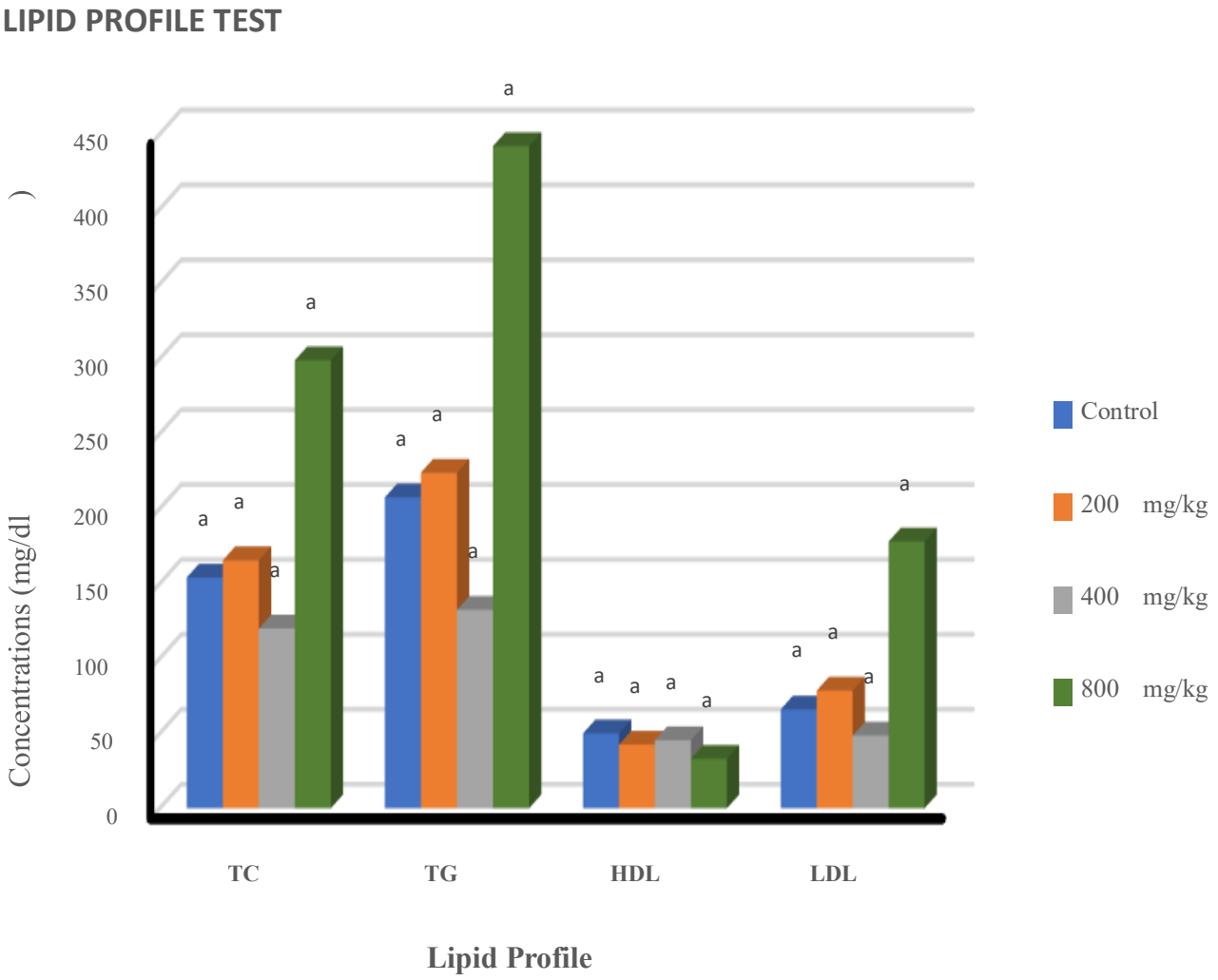
Effect of Brachystegia eurycoma extract on the lipid profile of male Wistar rats

Table 2. Effect of treatments on the lipid profile of the experimental animals

Groups	Doses	LIPID PROFILE (mg/dl)			
		TC	TG	HDL	LDL
1	Control	154.00 ± 59.00[a]	207.50 ± 105.56[a]	50.00 ± 8.00[a]	66.00 ± 27.00[a]
2	200 mg/kg	165.50 ± 90.59[a]	224.00 ± 168.15[a]	42.50 ± 11.64[a]	78.50 ± 45.61[a]
3	400 mg/kg	120.00 ± 51.24[a]	132.50 ± 55.86[a]	45.50 ± 16.57[a]	48.50 ± 7.57[a]
4	800 mg/kg	299.00 ± 254.00[a]	442.00 ± 387.07[a]	33.00 ± 20.00[a]	178.00 ± 159.00[a]

Values are expressed as Mean ± SD (n = 3). Means in the same column with the same superscript letter are not significantly different at $p > 0.05$.

Fig. 2. Bar chart showing the effect of treatments on the lipid profile of the experimental animals



The bar chart illustrates variations in total cholesterol (TC), triglycerides (TG), high-density lipoprotein (HDL), and low-density lipoprotein (LDL) levels across the experimental groups, with TC and TG showing higher values and HDL lower at 800 mg/kg. Nevertheless, as indicated in the table, these differences were not statistically significant ($p > 0.05$). The overlapping error bars in the chart, along with consistent superscripts in the table, indicate that the extract did not produce significant changes in lipid profile parameters, although dose-dependent trends were evident.

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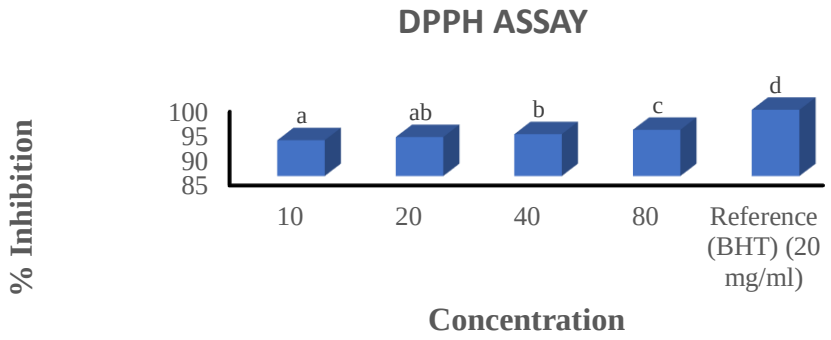
DPPH radical scavenging activity of Brachystegia eurycoma extract compared to BHT

Table 3. DPPH scavenging assay

Concentration (mg/ml)	% Inhibition
10	92.296 ± 0.146^{a}
20	92.942 ± 0.329^{ab}
40	93.537 ± 0.146^{b}
80	94.416 ± 0.219^{c}
Reference (BHT) (20 mg/ml)	98.501 ± 0.0^{d}

Values are expressed as mean ± standard deviation. Values with the same letter are not significantly ($p < 0.05$) different. Values with different letters are significantly ($p < 0.05$) different.

Fig. 3 Bar chart showing the DPPH scavenging activity in comparison to the standard (BHT)



The bar chart shows a clear concentration-dependent increase in radical scavenging activity for the ethanol extract of *Brachystegia eurycoma*. At the lowest concentration (10 mg/mL), inhibition was already high at 92.30%, and this rose steadily to 94.42% at 80 mg/mL. Although the differences between 10–40 mg/mL appear small, statistical analysis shows significant variation ($p < 0.05$), with the highest inhibition recorded at 80 mg/mL. When compared to the synthetic antioxidant standard, butylated hydroxytoluene (BHT), which produced 98.50% inhibition at 20 mg/mL, the extract demonstrated slightly lower but still appreciable scavenging activity.

Ferric reducing antioxidant power (FRAP) of Brachystegia eurycoma extract compared to gallic acid

Table 4. FRAP scavenging activity (%) of *Brachystegia eurycoma* extract in comparison to Gallic acid

Concentration (mg/ml)	% Scavenging	
	Brachystegia eurycoma	**Gallic acid**
10	77.478 ± 0.00^{a}	76.950 ± 0.07^{a}
20	73.478 ± 0.05^{b}	93.617 ± 0.23^{c}
40	69.624 ± 1.02^{b}	95.278 ± 0.00^{c}
80	87.999 ± 0.32^{d}	96.178 ± 2.30^{c}

Values are expressed as mean ± SD (n = 3). Different superscript letters within the same row indicate significant differences between extract and standard at the same concentration ($p < 0.05$).

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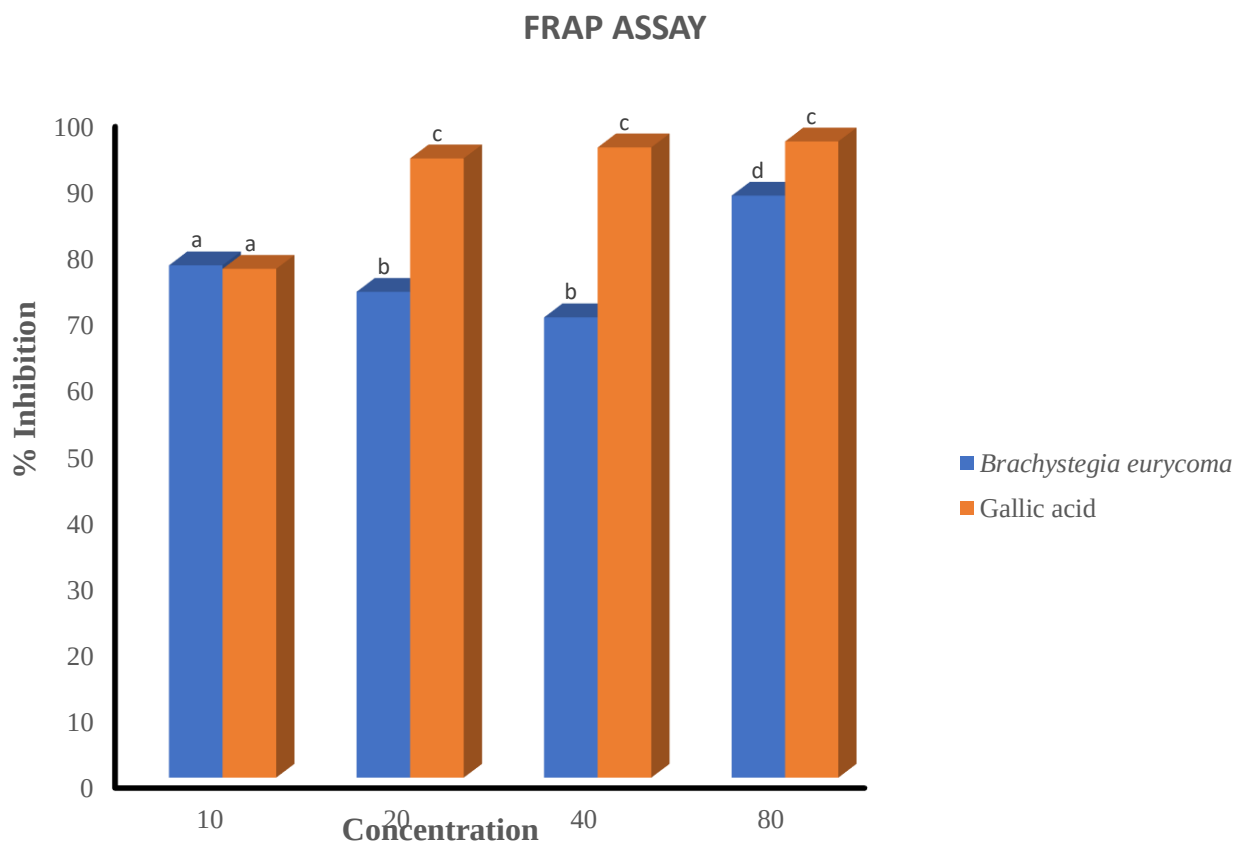
Fig. 4. Bar chart showing the FRAP scavenging activity in comparison to the standard (gallic acid)



The aqueous extract of *Brachystegia eurycoma* exhibited dose-dependent DPPH scavenging activity, with the highest activity observed at 80 mg/ml (~ 88%). While Gallic acid showed consistently higher scavenging activity at most concentrations, the extract demonstrated substantial antioxidant potential, comparable to the standard at the lowest concentration. The varying superscripts indicate significant differences between concentrations within each treatment group.

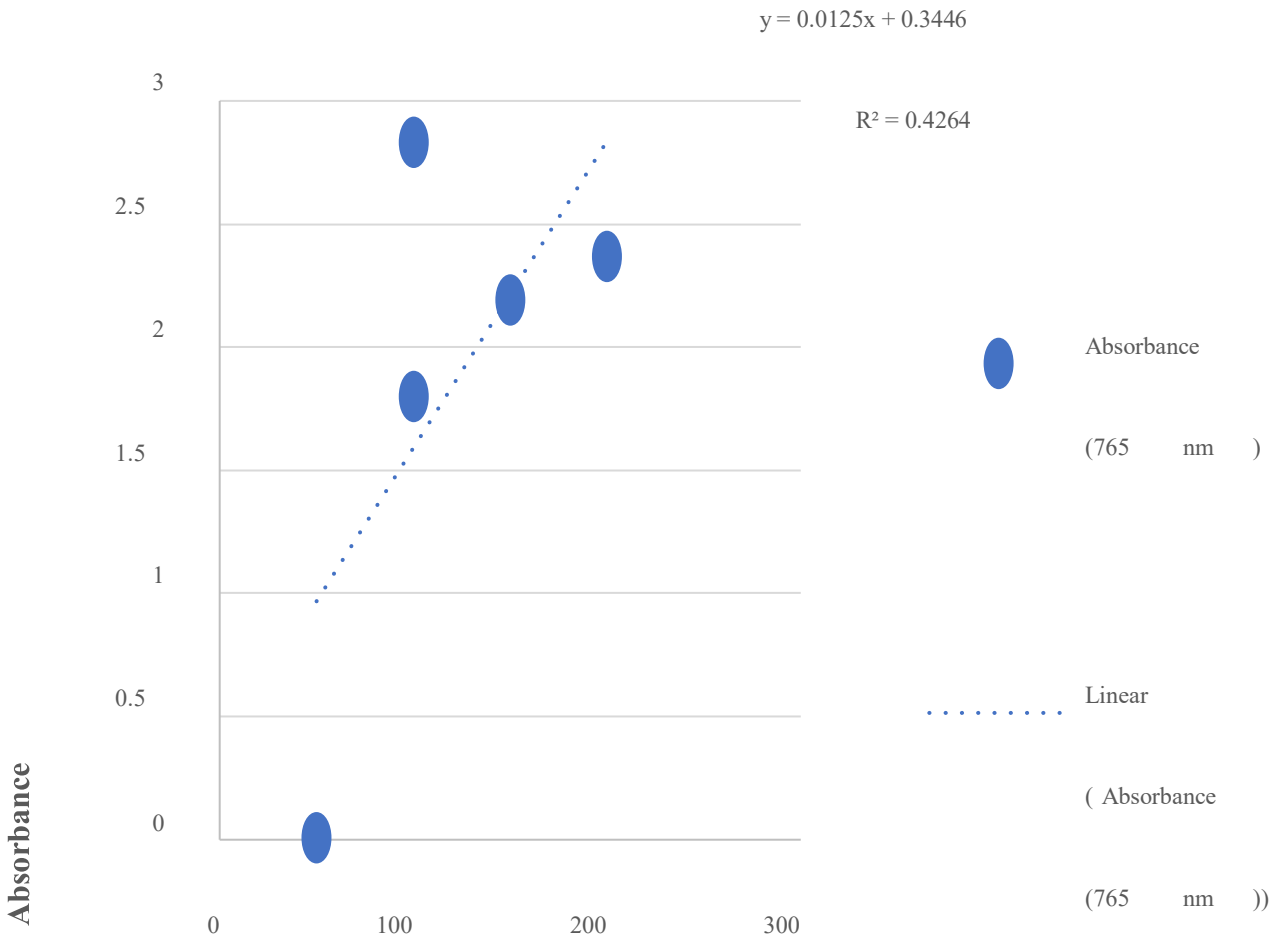
Total phenolic content of Brachystegia eurycoma extract

Table 5. Estimation of total phenolic content in ethanol extract of *Brachystegia eurycoma* seeds

Sample	**Concentration (µg/ml)**	**Absorbance (765 nm)**	**TPC (mg GAE/g extract)**
Standard (Gallic acid)	50	0.01	
	100	1.80	
	150	2.19	
	200	2.37	
Brachystegia eurycoma	100	2.83	1802.5

The absorbance of *Brachystegia eurycoma* extract (2.83 at 765 nm) was higher than that of the gallic acid standard across all tested concentrations, indicating a very high phenolic content. The calculated total phenolic content (TPC) of 1802.5 mg GAE/g extract suggests that the extract is exceptionally rich in phenolic compounds, which are known for their strong antioxidant potential.

Fig. 5. Calibration curve of Gallic acid in the estimation of total phenolic content



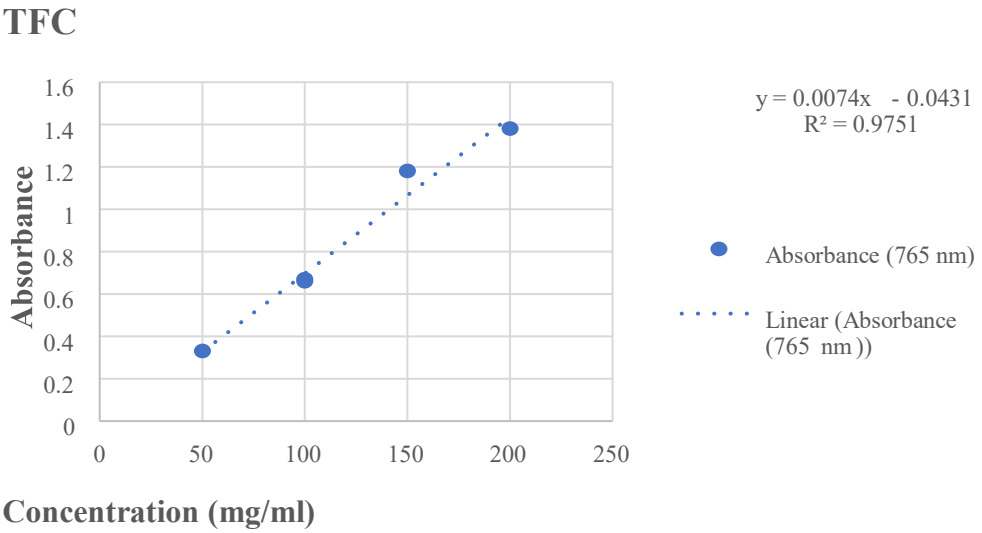
Total flavonoid content of Brachystegia eurycoma extract

Table 6. Estimation of total flavonoid content in ethanol extract of *Brachystegia eurycoma* seeds

Sample	Concentration (mg/ml)	Absorbance (765 nm)	TFC (mg QE/g extract)
Standard (Gallic acid)	50	0.33	
	100	0.66	
	150	1.18	
	200	1.38	
Brachystegia eurycoma	100	0.67	957.0

The absorbance value of *Brachystegia eurycoma* extract (0.67 at 765 nm) closely aligns with that of the quercetin standard at 100 mg/ml, indicating a considerable presence of flavonoid compounds. The calculated total flavonoid content (TFC) of 957.0 mg QE/g extract suggests that the extract is rich in flavonoids, which contribute significantly to its antioxidant, anti-inflammatory, and therapeutic properties.

Fig. 6. Calibration curve of Quercetin in the estimation of total flavonoid content



Comparison of present findings with previous studies on plant-based male fertility and antioxidant effects

TABLE 7: Comparison Of Present Findings With Related Studies On Plant-Based Male Fertility And Antioxidant Effects

Parameter	**Present Study (*Brachystegia eurycoma*)**	**Previous Studies**	**Key Observation**
Sperm Motility (%)	Increased (35% → 45% at 800 mg/kg)	Moringa oleifera improved motility in rats (Panova et al., 2025); Zingiber officinale enhanced motility (Gholami-Ahangaran *et al.*, 2021)	Confirms plant extracts enhance sperm motility
Sperm Morphology (%)	Increased normal forms (50.7% → 75%)	Nigella sativa improved morphology (Pandey *et al.*, 2024)	Suggests structural sperm improvement
Sperm Count (million)	Decrease at higher doses (50 → 30)	Carica papaya showed reduced sperm count at high doses (Lohiya *et al.*, 1994; Ansah *et al.*, 2016)	Indicates possible dose-dependent toxicity
Lipid Profile	No significant change ($p > 0.05$)	Allium sativum reduced TC and LDL (Irshad *et al.*, 2017)	Suggests weak hypolipidemic effect
DPPH Activity (%)	High (92–94%)	Vernonia amygdalina showed strong scavenging activity (Hussen & Endalew, 2023)	Comparable antioxidant activity
FRAP Activity (%)	Moderate to high (~88%)	Psidium guajava demonstrated strong reducing power (Venkatachalam *et al.*, 2012)	Confirms reducing potential
Total Phenolics (mg GAE/g)	Very high (1802.5)	Camellia sinensis rich in phenolics (de Moura *et al.*, 2022)	Supports antioxidant richness
Total Flavonoids (mg QE/g)	High (957.0)	Garcinia kola high flavonoids (Doh *et al.*, 2023)	Indicates bioactive potential

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DISCUSSION

This section interprets the findings of the study in relation to existing literature and underlying biochemical mechanisms. The observed changes in sperm parameters, lipid profile, and antioxidant status following administration of *Brachystegia eurycoma* extract are critically analyzed to elucidate its potential role in improving male reproductive function. Emphasis is placed on explaining the dose-dependent effects, possible mechanisms of action, and how the present findings align with or differ from previous studies, thereby highlighting the contribution of this study to current knowledge.

The present study evaluated the antioxidant activity and effects of aqueous extract of *Brachystegia eurycoma* seeds on lipid profile and sperm fertility parameters in male Wistar albino rats. The findings provide insights into the potential role of this plant extract in modulating reproductive health and oxidative status. Traditional medicine, as ancient as humanity itself, often relies on plants, a practice known as herbalism or botanical medicine, outside conventional Western medicine. Approximately two-thirds of the global population, particularly in developing countries, depend on it for primary healthcare. Plants contain bioactive compounds such as anthraquinones, flavonoids, glycosides, saponins, and tannins, which serve as the basis for treating various diseases. Many modern pharmaceuticals are derived from these phytochemicals, with each plant offering unique active principles responsible for their therapeutic effects (Anderson *et al.*, 2023). Herbal remedies are increasingly recognised as natural agents capable of enhancing male fertility. These plants exert their effects through multiple biological pathways that support sperm health and overall reproductive function. Key mechanisms include the stimulation of sperm production through hormonal modulation, where phytochemicals such as ginsenosides and diosgenin act on the hypothalamic–pituitary–gonadal axis (HPGA) to elevate testosterone levels and promote spermatogenesis. They also help improve reproductive blood flow by supporting hormonal balance within the HPGA. Additionally, many herbal agents provide anti-inflammatory and antioxidant benefits, further protecting sperm cells and enhancing fertility potential (Shakeri *et al.*, 2024).

Sperm morphology analysis delivers indispensable insights into a man's fertility potential and reproductive vitality (Shin & Kathrins, 2026). Abnormal forms often stem from genetic roots or reversible stressors – physiological, psychological, or environmental – that demand attention. While testes rebound from isolated stresses, chronic or repeated assaults inflict lasting damage to normal sperm morphology (Fig 1). Thus, prioritising sperm morphology evaluation is imperative for diagnosing male infertility and orchestrating effective clinical interventions (Menkveld *et al.*, 2010). Normal sperm morphology was also greatest in the 800 mg/kg group (75%), whereas the lowest value was recorded at 400 mg/kg (45%) as seen in Table 1. Administration of *B. eurycoma* extract significantly improved sperm motility, morphology, and count. The highest dose delivered unparalleled enhancements, showcasing markedly elevated progressive motility and the supreme share of morphologically normal spermatozoa compelling evidence of superior sperm performance driven by fortified antioxidant activity (Noh *et al.*, 2020; Monageng *et al.*, 2023) and reduced lipid peroxidation (Fatah *et al.*, 2023) in the epididymal milieu. By contrast, the 400 mg/kg dose yielded dismal semen quality, with rock-bottom active motility and peak abnormal morphology.

Oxidative stress in sperm arises from both internal and external sources. Mitochondria are a primary internal source of ROS, where excessive production impairs ATP generation and damages mitochondrial DNA, reducing motility and compromising genetic integrity. Externally, seminal leukocytes generate ROS, particularly during infections or inflammation, leading to localised oxidative damage. While this immune response targets pathogens, elevated ROS near sperm can impair function. Conditions like leukocytospermia, marked by increased seminal leukocytes, are associated with higher ROS levels and greater sperm DNA fragmentation (Wang *et al.*, 2025). Oxidative stress reduces sperm quality by lowering vitality, motility, and fertilising ability while increasing DNA and membrane damage. Excess ROS can cause structural abnormalities, greater membrane permeability, and reduced fertility (Sengupta *et al.*, 2024). Excessive ROS induces lipid peroxidation (Akhigbe, 2025; Potiris *et al.*, 2025), leading to membrane disruption and mitochondrial dysfunction in male reproductive cells and tissues (Akhigbe, 2025), and because sperm membranes contain high levels of unsaturated fatty acids, they are highly prone to lipid peroxidation. Damage to membrane integrity directly impairs motility. Lipid peroxidation can begin in mitochondrial membranes due to elevated ROS production, spreading to the plasma membrane. Mitochondrial dysfunction and impaired energy generation that follow are major contributors to sperm immobility and overall functional decline (Fatah *et al.*, 2023).

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The antioxidant assessment powerfully demonstrated the B. eurycoma extract's superior free radical scavenging capabilities across diverse assays. In the DPPH assay as seen in Table 3, inhibition rates surged impressively with concentration, achieving a remarkable 94.42% at 80 mg/ml, nearly rivalling the gold-standard BHT and highlighting its exceptional electron-donating and radical-neutralising prowess. FRAP outcomes further affirmed this potency, with reducing power escalating from 77.48% at 10 mg/ml to 87.99% at 80 mg/ml; while Gallic acid edged ahead overall, the extract's formidable ferric-reducing strength underscored its outstanding antioxidant profile (Fig 3; Fig 4). Reinforcing these results, phytochemical analysis unveiled extraordinarily elevated total phenolic content alongside substantial flavonoids, hallmarks of elite antioxidant and anti-inflammatory plants. These polyphenolic powerhouses unequivocally drove the enhanced sperm function by vanquishing oxidative stress, the chief assailant on membrane integrity, DNA stability, and motility, supporting the report by Bouhadana *et al.* (2025), that antioxidant supplements improve sperm quality by reducing oxidative stress, which could help prevent the disruption of sperm DNA integrity.

Hyperlipidemia, defined by abnormally high serum levels of total cholesterol, low-density lipoprotein, verylow-density lipoprotein, or triglycerides, or critically low high-density lipoprotein, severely undermines male fertility. Compelling experimental evidence from rats fed high-cholesterol diets, inducing hypercholesterolemia and hypertriglyceridemia, demonstrates sharp declines in sperm motility alongside surges in abnormal sperm morphology. In stark contrast, elevated HDL-C levels powerfully correlate with viable, morphologically normal sperm (Zubi & Alfarisi, 2021). Male reproductive health hinges on meticulous cholesterol homeostasis, with cholesterol acting as the indispensable precursor for steroid hormone synthesis vital to spermatogenesis (Sèdes *et al.*, 2018). The regulation of lipid metabolism is essential for normal sperm maturation, motility, capacitation, and acrosome function. Evidence from previous studies by Singh *et al.* (2024) indicates a strong association between lipid profile parameters and semen quality. Sperm concentration has been shown to correlate positively with triglycerides and very low-density lipoprotein (adjusted $p = 0.001$ and $p = 0.005$, respectively). Similarly, total and progressive motility increase with higher levels of low-density lipoprotein and cholesterol (adjusted $p = 0.008$ and $p < 0.001$, respectively). Conversely, elevated serum total cholesterol, free cholesterol, and phospholipids are linked to a reduced proportion of sperm with intact acrosomes and smaller sperm head dimensions (Singh *et al.*, 2024).

Administration of *Brachystegia eurycoma* (achi) seed extract resulted in a substantial rise in total cholesterol and triglycerides at 800 mg/kg (TC: 299 mg/dl; TG: 442 mg/dl), whereas the 400 mg/kg dose produced notable reductions in both indices compared with the control. At 800 mg/kg, HDL reached its lowest value (33 mg/dl), while LDL peaked at 178 mg/dl (Fig 2). The pronounced increase in TC and TG at the highest dose may indicate elevated lipid availability for steroid hormone synthesis, potentially contributing to improved sperm output and motility observed at this level. Conversely, the reductions in lipid components at 400 mg/kg may have limited steroidogenesis or disrupted membrane lipid balance, contrary to the findings of Sèdes *et al*. (2018), thereby contributing to the observed decline in sperm quality.

CONCLUSION

Although spermatozoa possess intrinsic antioxidant defences, these are constrained by their limited cytoplasmic volume, high polyunsaturated fatty acid content, and mitochondrial ROS production. Strategies such as supplementation, dietary interventions, and lifestyle modifications can enhance antioxidant capacity, mitigating oxidative damage and improving male fertility. *Brachystegia eurycoma* seed extract demonstrated potent antioxidant activity and effectively improved key sperm parameters, particularly at higher doses. While its impact on lipid profile was not statistically significant, the extract’s rich phenolic and flavonoid content indicates substantial potential to support reproductive function. Further investigations into hormonal regulation and oxidative biomarkers are warranted to clarify the underlying mechanisms.

Limitations of the Study

This study has some limitations that should be acknowledged. The sample size was relatively small, which may limit generalizability. Only short-term (14-day) administration of the extract was assessed, leaving long-term effects unexplored. Key reproductive hormones such as testosterone, LH, and FSH were not measured, and only aqueous extraction was performed, potentially limiting identification of specific bioactive compounds. In vivo oxidative stress markers in reproductive tissues were not evaluated, and variability in doses and methods across

previous studies limits direct comparison. Future research incorporating hormonal profiling, multiple extraction methods, long-term administration, and tissue-level oxidative stress analysis would provide a more comprehensive understanding of *Brachystegia eurycoma*'s reproductive and antioxidant effects.

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Author Contributions

NIE and ORN conceptualised and designed the study, with EUO and AO providing supervision. EUO, NIE, and AO established the research methodology. AO and ORN conducted the study, while EUO and NIE performed validation. The original manuscript draft was prepared by ORN, with AO and EUO responsible for reviewing and editing the draft. All authors reviewed and approved the final version of the manuscript for submission.

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Data Availability

The datasets generated during and/or analysed during the study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

All ethical guidelines have been adhered. There are no studies by any of the authors in this article that used human subjects.

Consent for publication

All authors have reviewed and approved the final version of this manuscript and consent to its publication in Discover Chemistry.

Competing interests

The authors declare no competing interests.

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