

The Histological and Hormonal Effect of Aqueous Extract of the Bark of *Boswellia dalzielii* on the Testis of Male Wistar Rat

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ABSTRACT

This study evaluated the effect of aqueous extract of stem bark of Boswellia dalzielii on the testis male Wistar rats.

Methods and Material: *Twelve (12) adult male Wistar rats were randomly divided into three groups; A-C (n=4). Group A was given distilled water, groups B and C were administered 100mg/kg and 300mg/kg of aqueous extract of Boswellia dalzielii respectively. All administrations were done orally for 21 days. Animals were sacrificed; histological studies and hormonal assays were carried out.*

Results: *Administration of Boswellia dalzielii resulted in a significant (p<0.05) increased testosterone level in the study groups compared to the control group. Testicular sections of the study groups showed increase in number and volume of germinal epithelium, which was more pronounced in group B.*

Conclusion: *The results of this study suggest that Boswellia dalzielii possesses aphrodisiac and fertility potentials.*

Key words: *Boswellia dalzielii, Male fertility, Testis, Testosterone, Phytotherapy.*

INTRODUCTION

Background of Study

Phytotherapy is the oldest and most widely used therapy, plant materials are used improve health. These plant materials are less expensive and provide safer means of treatment than conventional medications. Africa is endowed with abundant plant materials that serve as important sources of

nourishment and are often used to treat of different diseases^{1,2}.

Boswellia dalzielii commonly known as frankincense tree growing up to 13m high and is found mainly in the Savannah region of West Africa. It is abundant in northwestern Nigeria, where the Hausa speaking people refer to it as “Ararrabi”, “Basamu” or “Hanu”³.

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The tree has a characteristic pale papery ragged bark that peels. The stem bark secretes a fragrant white gum that is burnt to fumigate cloth and to drive out flies and mosquitoes from rooms.

Boswellia dalzielii is consumed locally and is traditionally considered to have medicinal properties; the leaf extract is used to treat diarrhea, the root decoction of *B.dalzielii* administered enhance wound healing and the bark is eaten to induce vomiting and relieve symptoms of giddiness and palpitation. Oil from the leaves of *B.dalzielii* has been reported to show anti-ulcer activity and reduced gastrointestinal motility³.

The biological active compounds in *B.dalzielii* include: cholesterol, flavanoid, tannis, glycosides, alkaloids, anthracene and saponin^{4,5}.

The decoction of the bark of the plant is consumed among the Hausas of northern Nigeria for its aphrodisiac and fertility potentials³. It is important to; evaluate the effect of aqueous extract of stem bark of *Boswellia dalzielii* on the testis male Wistar rats.

MATERIALS AND METHODS

PLANT COLLECTION

The stem bark of *Boswellia dalzielii* was harvested fresh from its natural habitat in Kano state and authenticated at the Department of Botany, University of Nigeria, Nsukka. Voucher number UNH252.

PREPARATION OF EXTRACT

The stem bark of *Boswellia dalzielii* was air dried and pulverized using a mortar and pestle and sieved to a fine powder. The powdered stem bark was soaked in water for 24 hours and filtered using a white muslin cloth to remove debris. The resultant filtrate was allowed to evaporate to dryness which was stored in a sealed plastic container until required⁵.

PHYTOCHEMICAL ANALYSIS

Phytochemical analysis of the stem bark was done at Project Development Institute (PRODA), ENUGU to ascertain the active biochemical component of the stem bark. The following tests were carried out: Mayer's test,

Borntrager's test, Test for Saponins, Alkaline reagent test, Libermann-Burchard's test⁷.

EXPERIMENTAL ANIMALS

12 male adult Wistar rats were procured from the animal house of the Department of Pharmacology and Toxicology, Faculty of Pharmaceutical Sciences, University Of Nigeria, Nsukka. The rats were housed in cages in the animal house of Anatomy Department Faculty of Basic Medical Sciences University of Nigeria, Enugu campus. The rats were allowed acclimatize for two weeks and maintained under standard environmental conditions. The animals had free access to standard livestock pellets and water.

EXPERIMENTAL DESIGN

12 male Wistar rats for this study were randomly divided into 3 groups of 4 rats each. The administration period spanned for 21 days:

Group A (positive control group) was given 5ml of normal saline only for the period of 21 days. Group B (Extract group) was given 100mg/kg body weight of aqueous extract of *Boswelliadalzielii* for 21 days. Group C (Extract group II) were given 300mg/kg body weight of aqueous extract of *Boswellia dalzielii* per day for 21 days. All administrations were done orally.

SAMPLE COLLECTION

At the end of the treatment, each rat was anaesthetised with 25% urethane at a dose of 0.6 ml/100g. The blood was collected for hormonal assay and the testes were rapidly dissected for processing and light microscopic study.

DETERMINATION OF HORMONAL LEVELS

Blood was collected from each rat via the left ventricular cardiac puncture and kept in non-heparin vacuum container which was span at 2500rpm for 10min using a bio-centrifuge (MSE, O-5122A, Germany). The level of free serum testosterone, luteinizing hormone (LH) and follicle stimulating hormone (FSH) was measured with ECOBAS-6000 hormone analyzing machine^{1,7}.

LIGHT MICROSCOPIC STUDY

The harvested testes were fixed in modified Davidson's fluid (MDF) for 24 hours^{1, 8}. Standard protocol was followed in processing the tissue for microscopic examination^{1, 9, 10}. Paraffin sections were cut at 3µm thick^{1, 8} and stained with hematoxylin and eosin (H & E).

STATISTICAL ANALYSIS

Data collected was analysis using Statistical Package for Social Science (SPSS) version 20.0. Data were expressed as mean ± standard deviation. One-way analysis of variance (ANOVA) and student t-test were used to

study variations between groups. Statistical significance was considered at the level of p-value <0.05.

RESULTS

PHYTOCHEMICAL ANALYSIS

The result of the phytochemical analysis (table 1) revealed the presence of flavonoid, tannin and saponin in the aqueous extract of stem bark of *Boswellia dalzielii*. Flavonoid occurred at a higher percentage compared to tannin and saponin. Alkaloid, steroid and glycoside were absent.

Table 1: Phytochemical composition of aqueous bark extract of *Boswellia dalzielii*

S/N	PHYTOCHEMICAL CONSTITUENT	AMOUNT PRESENT
1	ALKALOID	-
2	TANNIN	++
3	SAPONIN	++
4	CARDIAC GLYCOSIDE	-
5	FLAVONOID	+++
6	STEROIDS	-

Where: +++ is very high concentration,

++ is high concentration, - is absence

BODY WEIGHT

The result in table in Table 2 showed increase in weight of control and group B, there was however, decrease in the weight of group C,

the decreased in weight was significant ($p < 0.05$) when compared to the control and group B.

Table 2: Showing weight change of experimental animals

GROUP	INITIAL WEIGHT	FINAL WEIGHT	WEIGHT CHANGE
A	239.88	254.28	14.40 ± 8.40 ^a
B	250.88	258.10	7.33 ± 7.10 ^A
C	202.75	192.93	-10.63 ± 26.0 ^{Aa}

HORMONAL ASSAY

The result of the hormonal assay (table 3) showed significantly higher ($p < 0.05$) serum testosterone level group B and C compared to the control (group A). Group B had the highest serum testosterone level. The serum FSH and

LH levels were significantly lower ($p < 0.05$) in group C compared to control. In group B the FSH level decreased and the LH level increased compared to control, the changes were however, not significant ($p < 0.05$).

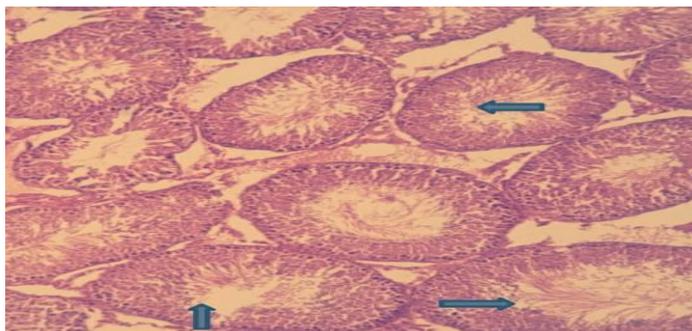
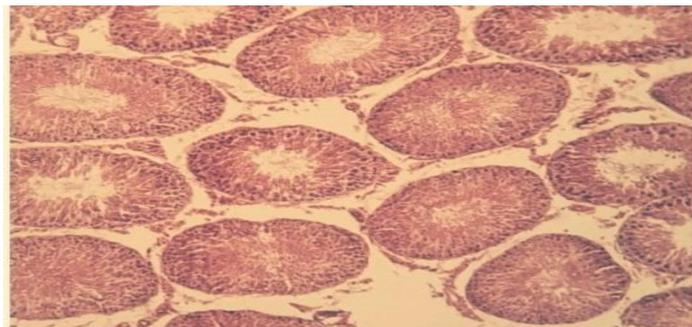
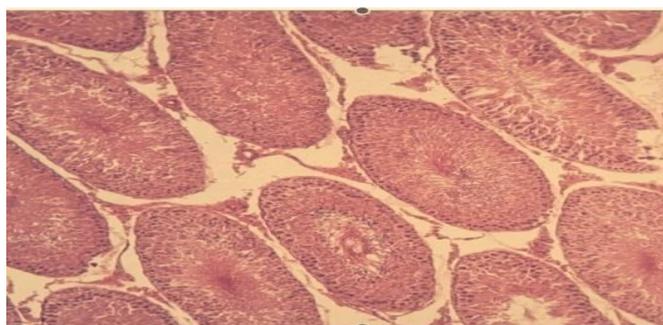
Table 3: The levels of testosterone, FSH and LH of the experimental animals

GROUPS	TESTOSTERONE	FSH	LH
A	0.10 ± 0.00 ^a	13.50 ± 0.70	10.0 ± 1.41
B	1.35 ± 0.71 ^A	13.00 ± 0.00	11.5 ± 0.71
C	0.40 ± 0.00 ^{Aa}	3.9 ± 0.28 ^{Aa}	7.5 ± 0.00 ^a

TESTICULAR HISTOLOGY

The light microscopic study showed that the testis of the control group had an apparently normal architecture and cellular composition

(plate I). Testicular sections of group B and C (plate II & III respectively) showed increase in number and volume of germinal epithelium, which was more pronounced in group B.

**Plate I****Plate II****Plate III**

Photomicrograph of testis of male Wistar rat in group A, B and C (Plate I, II & III respectively)

DISCUSSION

Phytochemical analysis of aqueous extract of the bark of *Boswellia dalzielii* revealed the presence of flavonoid, saponin and tannin in high concentrations. Our finding is in agreement with the report of Nwinye *et al*³. However, the study by Uzama *et al.*,¹¹ reported the presence of alkaloid, cardiac glycoside and steroid, this was in contrast with our finding. The difference could be as a result of differences in soil type or geographical locations.

Boswellia dalzielii at high dose was found to reduce body weight, a property that

can be used in managing obesity related diseases. This is as a result of the high presence of tannin in aqueous bark extract of *Boswellia dalzielii*. Tannin has been suggested to be responsible for decrease in feed intake, growth rate and protein digestibility in experimental animals¹². This explains the reduction in body weight following administration of high dose of the aqueous extract of *Boswellia dalzielii*.

Evidenced in this study was an increase in the number and volume of germinal epithelium, the interstitial cells as well as the sizes of the seminiferous tubules. It

is well established that morphological changes in rat testis has been associated testosterone level^{1, 13}. The result of hormonal assay showed that there was increased testosterone, LH and FSH levels following administration of low dose *Boswellia dalzielii*. These hormones enhances germ cell proliferation. LH binds to LH stimulates synthesize testosterone by the interstitial cells (of leydig) while FSH stimulates sertoli cells to synthesis and release androgen binding proteins (ABP) into the seminiferous tubules. ABP combines with testosterone and in turn increases its concentration in the seminiferous tubules which then stimulates spermatogenesis^{1, 15}. Increased libido and spermatogenesis has been associated with increased concentration testosterone^{1, 14, 16}.

CONCLUSION

The results of this study suggest that *Boswellia dalzielii* has aphrodisiac and fertility potentials.

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