Original Research Article

Androgenic properties and subchronic toxicity of the aqueous extract of *Pycnanthus angolensis* (Welw.) Warb. wood (Myristicaceae)

ABSTRACT

Aims:*Pycnanthus angolensis (Welw.) Warb.*also known as "*Etengué*" in the *Baka* language, isa medicinal plant used by the Baka Pygmies of Cameroon to treat erectile dysfunction. This study aimed to evaluate the androgenic activity and subchronic toxicity of the aqueous extract of *Pycnanthus angolensis* wood.
Study design: Experimental design.
Place and Duration of Study: Laboratory of Biochemistry, Faculty of Medicine and Biomedical Sciences, University of Yaoundé I, between November 2022 and October 2023.
Methodology: Twenty-five male *Wistar* strain rats were divided into 5 groups of 5 rats each including a positive control group that received testosterone enanthate (5 mg/kg) intramuscularly once a week; a normal control group that received distilled water (10 ml/kg) and three test groups which received 134 and 267 and 533 mg/kg of the aqueous extract of *Pycnanthus angolensis* daily *per os*. On the 29th day, the animals were killed, and several biochemical parameters were assessed for androgenic properties.
Results: At a dose of 134 mg/kg, the extract significantly increased ($p<0.05$) the levels of testicular cholesterol**,** testosterone and serum prostatic acid phosphatase activity compared to those of the normal control. A toxicity study showed that at doses of 134 and 267 mg/kg, the extract did not induce any significant variation ($p>0.05$) in the creatinine level, but a significant reduction ($p<0.05$) in of aspartate and alanine aminotransferase activities was recorded. Histopathology revealed destruction of testicular spermatozoa at a dose of 533 mg/kg compared to that of the controls.
Conclusion: These results showed that at a dose of 134 mg/kg, the aqueous extract of *Pycnanthus angolensis (Welw.) Warb.* wood is capable of stimulating the production of androgens and is tolerated by the rat's body. Our results justified the traditional use of this plant for thetreatment of male infertility and erectile dysfunction.

Keywords: Pycnanthus angolensis; androgenic properties; subchronic toxicity; male rat.

1. INTRODUCTION

Androgens or male sex hormones, such as testosterone, play vital roles in the development and function of the male reproductive system[1]. These individuals are responsible for male sex characteristics, sexual health, muscle growth, bone density and cognitive function in men. Androgens exert their effects by binding to specific receptors in target cells, there by activating signaling pathways that regulate many biological processes[2,3].Dysfunctions in the production, regulation or action of androgens can lead to disorders of the male reproductive system such as erectile dysfunction, decreased libido and impaired fertility. The incidence of male sexual dysfunction resulting from hormonal imbalance is

estimated to be 20-25% and this imbalance can significantly impact the quality of life and overall health of the individuals involved[4].
Thus, to try to overcome these dysfunctions, the use of medicinal plants is a common and safe practice in many cultures[5]. Medicinal plants provide a rich natural source of bioactive compounds that can modulate androgen activity. Its use dates back thousands of years and many cultures have developed valuable knowledge about plants used effectively to improve male health, boost libido and treat male sexual dysfunction. The study of medicinal plants as potential agents for regulating androgen release and improving male health is therefore highly important. Indeed, several plants such as *SpathodeaCampanulata* and *Ficus asperifolia* are used for their androgenic properties[4,6]. However, despite their multiple benefits, previous studies have reported that plants are the cause of3-5% of reported poisonings in previous studies, 17% of which were associated with fatal events[7]. Therefore, evaluating the toxicity of any plant extract intended for use in humans or animals is important[8]. Thus, *Pycnanthus angolensis (Welw.) Warb. (P. angolensis)* is a medicinal plant traditionally used in Cameroon by the Baka'a Pygmies to manage erectile dysfunction in men. Previous studies have confirmed the effectiveness of the *in vitro* androgenic activity of *P. angolensis* wood through the increasing of testosterone levels[9] and penile nitric oxide production in rats[10].An acute toxicity study of *P. angolensis* woodalsodemonstratedthat this plant is practically nontoxic with a LD50 greater than 5000 mg/kg of body weight[11]. However, there is a lack of information on the *in vivo* androgenic activity and long-term toxicity of the aqueous extract of *P. angolensis* wood. Thus our research aimed to evaluate the androgenic activity and determine the subchronic toxicity of the aqueous extract of the wood of *P. angolensis* in male rats. The results of this study will contribute to expanding the knowledge on the use of this plant in traditional medicine and will provide crucial information regarding its therapeutic potential and its safety for long-term use.

2. MATERIAL AND METHODS

2.1 Plant collection and extract preparation

The wood of *Pycnanthus angolensis* (Welw.) Warb was harvested on November 2022in the Centreregion of Cameroon. A sample was identified in the National Herbarium of Cameroon with specimen number HNC31369. The plant name has been checked with http://www.worldfloraonline.org (accessed 16 January 2024).The wood was dried at room temperature and powdered in order to prepare the 10% aqueous extract by decoction for 30 min in distilled water as recommended by the traditional healer. The obtained aqueous extract was then filtered, evaporated and kept in a sealed glass container. The extract was reconstituted in distilled water in order to obtain the three animal equivalent doses:134, 267 and 533 mg/kg of body weight. The extract yield was 4.4%.

2.2 Reagents

Testosterone enanthate (TE), Androtardyl 250 mg, was used during the experiment.

2.3 Animals

The animals used for this study were 3-months-old male *Wistar* albino rats of, with a body weight between 140 and 200 g. These animals were obtained from the animal house of the Department of Animal Biology of the Faculty of Sciences of the University of Yaoundé I. All the rats had freeaccess to a standard diet and received food and tap water *ad libitum*. The study design was approved by the “Animal Ethics Committee”, the Join Institutional Review Board of Animal and Human Bioethics in Cameroon under the following reference number: BTC-JIRB2022-039.

2.4 Treatment of animals

During this experiment, 25 mature male rats were randomly divided into 5 groups of 5 rats each and treated for 28 days [12]as follows:

- The normal control group (NO): normal rats that received oral administration of distilled water (10 ml/kg, body weight (BW));
- The positive control group (TE): rats that received 5 mg/kg of testosterone enanthate per week via intramuscular injection[6];
- The first test group (D1) included rats that received 134, mg/kg, BW of *P.angolensis* aqueous extract;
- The second test group (D2): rats that received 267 mg/kg, BW of *P.angolensis* aqueous extract;
- The third test group (D3): rats that received 533 mg/kg, BW of *P.angolensis* aqueous extract.

Animals received daily an oral administration of distilled water or plant extract. After 28 days of treatment, the animals were fasted for 12 hours, anaesthetized with ether and killed by decapitation. Blood samples were collected in dry tubes and were centrifuged at 3000 rpm for 15 min. The collected sera were stored at -20°C. Androgen-dependent organs such as the testis, epididymis, seminal vesicles and other general metabolic organs (kidneys and liver) were carefully removed. Each organ was weighed with a sensitive electronic balance and preserved in Bouin’s solution for histological sectioning.

2.5 Assessment of biochemical markers involved in androgenic activity

Total testicular protein levels were determined by the method of Gornal *etal.*[13]. Testicular total cholesterol was measured by the enzymatic method described by Allain *et al*.[14] using the kit *BIOLABO* (Ref: 80106). Serum testosterone levels were determined by the enzyme-linked immunosorbent method using the *MONCENT* kit (Ref: EL1-1263) and prostatic acid phosphatase activity was determined by the Hillmann method using the *CHRONOLAB* kit (Ref: 101-0311).

2.6 Determination of biochemical parameters for subacute toxicity

Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST) activities were determined by the colorimetric method of Reitman and Frankel[15]. Serum total cholesterol and high-density lipoprotein cholesterol (HDL) were measured by the colorimetric method of Warnick *et al.*[16]. Serum triglyceride levels were determined by the method of Fossati and Prencipe[17], and serum LDL levels were determined by the method of Friedewald*etal*.[18]. Creatinine levels were evaluated as described by Bartels *etal.*[19] and the urea levels were determined via the Berthelot colorimetric method.

2.7 Histopathological study

Organs such as the testis, epididymis, seminal vesicles, kidneys and liver were previously fixed in Bouin solution for two weeks. Histological studies were carried out according to the method of Ahmadi *et al.*[20].

2.8 Statistical analysis

The results obtained are expressed as the means ± standard deviations and were analyzed using GraphPad Prism software (version 8.0.1) by the Kruskal-Wallis test followed by the Dunn's post hoc test. Differences were considered significant at $P<0.05$.

3. RESULTS

3.1 Effect of the extract on the relative weight (RW) of the testis and epididymis

Table 1 summarizes the effect of the aqueous extract of *P. angolensis* on the relative weight of the testis, epididymis and seminal vesicles of rats. At a dose of 134 mg/kg, the extract significantly increased ($P<0.05$) the RW of the testis and seminal vesicles of the rats compared to those of the normal control.

Table 1.Effect of the extract on the relative weights of androgen-dependent organs

	Groups				
Organs	**NO**	**TE**	**D1**	**D2**	**D3**
Testicles	1.02 ± 0.14	0.98 ± 0.10	1.43 ± 0.43 [ab]	1.21 ± 0.22	1.05 ± 0.37
Epididymis	0.36 ± 0.07	0.34 ± 0.11	0.33 ± 0.10	0.42 ± 0.04	0.37 ± 0.05
Seminal vesicles	0.16 ± 0.01	0.24 ± 0.05	0.41 ± 0.02 [a]	0.31 ± 0.16	0.18 ± 0.02

The values are expressed as the means ± SDs (n=5). NO: normal control group; TE: positive control group; D1: test group at a dose of 134 mg/kg body weight; D2: test group at a dose of 267 mg/kg body weight; D3: test group at a dose of 533 mg/kg body weight. [a]: P< 0.05 significant difference compared to the normal control group; [b]: P< 0.05 significant difference compared to the positive control group.

3.2 Effect of the extract on biochemical markers involved in androgenic activity

Table 2 shows the effects of the aqueous extract on the testicular total protein concentration, concentration of testicular total cholesterol, serum testosterone concentration and the activity of prostatic acid phosphatase (PACP). The administration of the extract at 134 mg/kg body weight (D1) significantly increased ($P<0.05$) the testicular protein level compared to that of the positive control. A significant increase ($P<0.05$) was observed in testicular cholesterol levels in animals in the D1 group compared to those in the control groups. There was a significant increase ($P<0.05$) in testosterone levels in the D1 group compared to the normal control but there was no significant difference ($P>0.05$) compared to the positive control group. Administration of the extract induced a significant increase ($P<0.05$) in PACP activity in the D1 group compared to that in the normal control group. The extract significantly ($P<0.05$) increased the nitric oxide concentration in the D1 group compared to the normal control.

Table 2. Effect of the extract on biochemical markers involved in androgenic activity

Biochemical markers	Groups				
	NO	TE	D1	D2	D3
Testicular total protein concentration (g/L)	7.50 ± 0.57	5.80 ± 0.48	12.00 ± 1.1 [bc]	7.50 ± 1.60	3.70 ± 1.00 [a]
Total testicular cholesterol concentration (mg/dL)	9.30 ± 0.49	9.80 ± 0.44	14 .00± 0.73 [ab]	12.00 ± 1.20	10.00 ± 0.61
Serum testosterone concentration (ng/mL)	10.00 ± 0.14	38.00 ± 3.80	32.00 ± 4.20 [ac]	24.00 ± 2.60	18 .00± 1.30 [b]
Activity of prostatic acid phosphatase (U/L)	14.00 ± 0.67	53.00 ± 1.80	36.00 ± 4.90 [ac]	22.00 ± 0.71	20.00 ± 0.56 [b]
Serum nitric oxide concentration (mg/dl)	2.70 ± 0.33	4.30 ± 0.90	4.80 ± 0.49 [ac]	3.60 ± 0.43	3.00 ± 0.49

The values are expressed as the means ± SDs (n=5). NO: normal control group; TE: positive control group; D1: test group at a dose of 134 mg/kg body weight; D2: test group at a dose of 267 mg/kg body weight; D3: test group at a dose of 533 mg/kg body weight. [a]: P< 0.05 significant difference compared to the normal control group; [b]: P< 0.05 significant difference compared to the positive control group, [c]: P< 0.05 significant difference compared to D3 group.

3.3 Effect of *P. angolensis* aqueous extract on testicular histology

An illustration of the effect of *P. angolensis* on the microarchitecture of the testis is given in Figure 1. Compared to those in the normal control group (NO), the histopathological changes and destruction of testicular spermatozoa were observed only in the D3 group. Like in to the positive control group, improvements effects in the proliferation of testicular spermatozoa, were observed in the D1 and D2 groups.

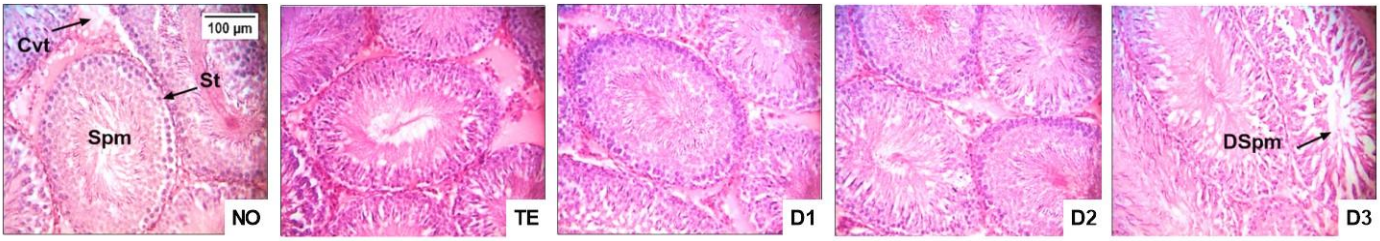

Fig. 1. Photomicrographs of the testes (100X) and haematoxylin staining
St= Seminiferous tube; Cvt= Connective-vascular tissue; Spm= Sperm; Dspm= Destruction of sperm

3.4 Effects of the extract on subchronic toxicity parameters

3.4.1 Effects of the extract on the body weight (BW) of the rats

Table 3 shows the effect of the extract on the BW of the rats. The results showed that treatment increased the BW of rats with BW gains of 42.27; 45.22; 20.40 and 12.13 g in the TE-treated group; D1; D2 and D3 groups, respectively compared to the initial day. Only the BW of the rats in D1 group was significantly greater than that of the normal control rats.

Table 3. Effect of the extract at different doses on the body weight (g) of rats

Body weight (g)	Groups				
	NO	TE	D1	D2	D3
BW initial	174.92 ± 11.02	170.65 ± 21.13	178.55 ± 21.69	176.35 ± 9.29	169.17 ± 7.67
BW final	209.00 ± 10.39	212.92 ± 19.37	223.77 ± 25.2 [ab]	196.75 ± 10.55	181.30 ± 9.04

Gain of BW	34.07 ± 0.63	42.27 ± 1.76	45.22 ± 3.51	20.40 ± 1.26	12.13 ± 1.37

The values are expressed as the means ± SDs (n=5). NO: normal control group; TE: positive control group; D1: test group at a dose of 134 mg/kg body weight; D2: test group at a dose of 267 mg/kg body weight; D3: test group at a dose of 533 mg/kg body weight. [a]: P< 0.05 significant difference compared to the normal control; [b]: P< 0.05 significant difference compared to the positive control group.

3.4.2 Effects of the extract on the relative weights of the liver and kidneys

Table 4 summarizes the effects of the extract on the relative weights of the liver and kidneys. Administration of the extract at all doses did not significantly affect the relative weight of the liver or kidneys compared to those of the normal control or the positive control.

Table 4. Effect of the extract on the relative weights of the liver and kidneys of the animals after 28 days of treatment

	Groups				
Organs	**NO**	**TE**	**D1**	**D2**	**D3**
Liver	3.56 ± 0.47	3.56 ± 0.47	3.29 ± 0.23	3.11 ± 0.65	3.42 ± 0.39
Kidneys	0.70 ± 0.67	0.61 ± 0.03	0.65 ± 0.07	0.63 ± 0.08	0.60 ± 0.08

The values are expressed as the means ± SDs (n=5). NO: normal control group; TE: positive control group; D1: test group at a dose of 134 mg/kg body weight; D2: test group at a dose of 267 mg/kg body weight; D3: test group at a dose of 533 mg/kg body weight.

3.4.3 Effects on the activity of hepatic transaminases: ASAT and ALT

The effects of the extract on the AST and ALT activities of the rats are displayed in Figure 2. Administration of the extract at doses of 134 mg/kg and 267 mg/kg significantly decreased (P<0.05) AST activity compared to that of the positive control group. The AST activity significantly increased (P>0.05) in D3 group compared to that of the normal control. However, the treatment did not induce any significant difference in ALT activity in any of the treated groups compared to the normal control group. A significant decrease in ALT (P< 0.05) was recorded in the D1 and D2 groups compared to the positive control group.

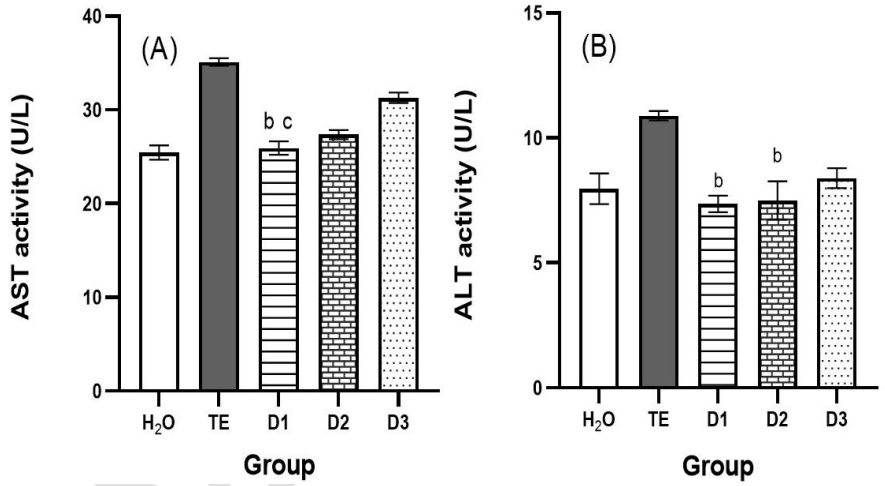

Fig. 2. Effect of the extract at different doses on the activity of AST (A) and ALT (B) in rats
The values are expressed as the means ± SDs (n=5). NO: normal control group; TE: positive control group; D1: test group at a dose of 134 mg/kg body weight; D2: test group at a dose of 267 mg/kg body weight; D3: test group at a dose of 533 mg/kg body weight. a: P< 0.05 significant difference compared to the normal control; b: P< 0.05 significant difference compared to the positive control; *: p< 0.05 significant difference compared to D3 group.

3.4.4 Effects of extract on several biochemical parameters

Figure 3 exhibits the effect of the extract on the total cholesterol concentration; serum HDL cholesterol concentration and total LDL cholesterol concentration. Administration of the extract at all doses induced a significant increase (P<0.05) in total cholesterol levels compared to those in the normal control group. The total cholesterol levels were significantly increased (P<0.05) in D2 group compared to those in the positive control group. Treatment significantly (P<0.05) reduced

HDL levels in all treated groups compared to those in the normal and positive control groups but it significantly increased (P<0.05) the LDL level in all treated groups compared to that in the normal and positive control groups.Table 5 shows the effect of the extract on creatinine and urea concentrations. There was no significant difference (P>0.05) in the serum creatinine concentration between the different groups of rats and between the normal and positive control groups. The treatment significantly (P<0.05) reduced the urea concentration in all the treatment groups compared to that in the normal control group. There was no significant difference compared to the positive control, except in the D3 group.

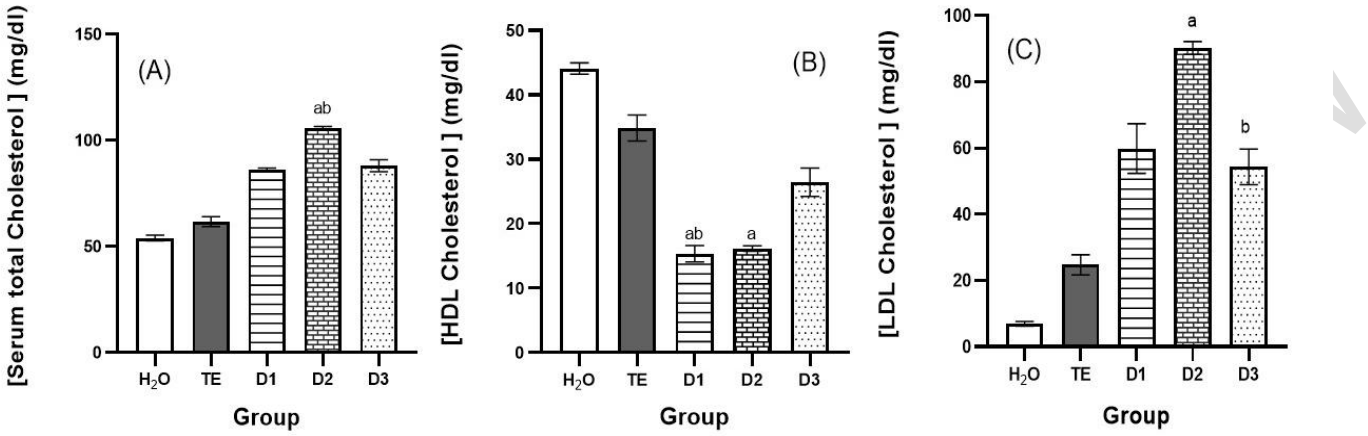

Fig. 3. Effects of the extract at different doses on the total cholesterol (A), HDL (B) and LDL (C) concentrations in the serum
The values are expressed as the means ± SDs (n=5). NO: normal control group; TE: positive control group; D1: test group at a dose of 134 mg/kg body weight; D2: test group at a dose of 267 mg/kg body weight; D3: test group at a dose of 533 mg/kg body weight. a: P< 0.05 significant difference compared to the normal control; b: P< 0.05 significant difference compared to the positive control; *: p< 0.05 significant difference compared to D2 group.

Table 5. Effect of the extract on creatinine and urea concentrations

	Groups				
Parameters	**NO**	**TE**	**D1**	**D2**	**D3**
Creatinine concentration (mg/L)	12.00 ± 0.77	12.00 ± 1,30	10.00 ± 0.80	10.00 ± 1.20	11.00 ± 0.40
Urea concentration (mg/L)	17.87 ± 0.87	1.59 ± 0.51	3.29 ± 1.43 [a]	4.07 ± 0.66 [a]	6.45 ± 0.51 [ab]

The values are expressed as the means ± SDs (n=5). NO: normal control group; TE: positive control group; D1: test group at a dose of 134 mg/kg body weight; D2: test group at a dose of 267 mg/kg body weight; D3: test group at a dose of 533 mg/kg body weight. [a]: P< 0.05 significant difference compared to the normal control; [b]: P< 0.05 significant difference compared to the positive control group.

3.4.5 Effects of *P. angolensis* aqueous extract on liver and kidney histology

The effects of the extract on the histology of the liver and kidney of rats after 28 days of treatment are illustrated in Figure 4. No notable alterations were observed in the liver or kidneys of the different groups.

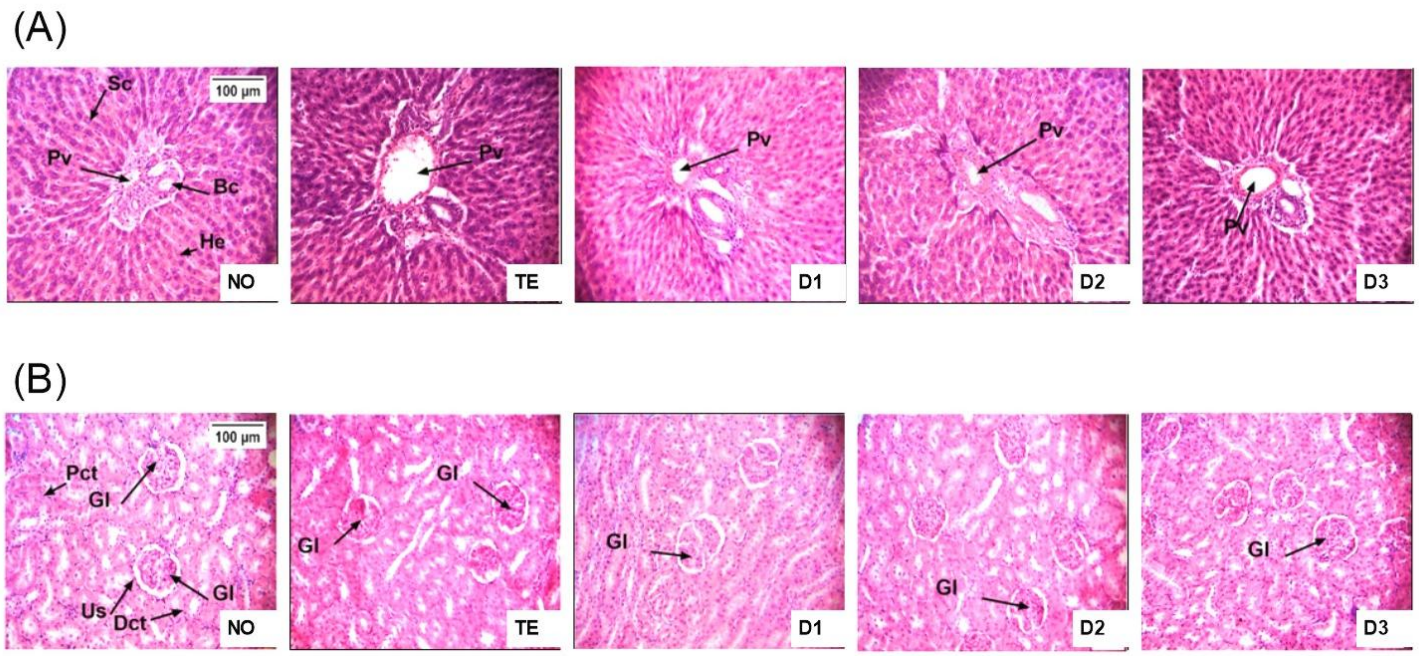

Fig. 4. Photomicrographs of the liver (A) and kidney (B) (X250); Haematoxylin-eosin staining
Pv= portal vein; He = Hepatocytes; Bc= Bile canaliculus; Sc= sinusoid capillaries; Gl= glomerulus; Us= urinary space; Dct= distal convoluted tubule; Pct= Proximal convoluted tubule

4. DISCUSSION

The aim of this work was to study the androgenic activity and subchronic toxicity of the aqueous extract of *P. angolensis* wood. The empirical use of its wood extract as an aphrodisiac combined with previous work on this plant has stimulated the evaluation of its activity as an androgenic agent.

Evaluation of androgen-dependent sexual organ weight ratios provides useful information on the androgenic activity of compounds and plant extracts. An increase in relative organ weight might indicate inflammation or an increase in the organ's secretory capacity, while a decrease would indicate cell shrinkage[21]. The dose of 134 mg/kg BW resulted in an increase in the relative weight of the sexual organs compared to that of the controls. However, no evidence of inflammation was reported by histological analysis at this dose. Thus, the increase in relative organ weight observed in the group treated with this dose could be attributed to increased secretory activity in the testes and seminal vesicles. Testicular proteins are substances that ensure steroidogenesis and spermatogenesis[22-24]. An increase in testicular protein concentration suggests enhanced testicular growth and androgenic activities[4]. Testicular proteins increased in the group treated with 134 mg/kg of BW, which could be attributed to the action of testosterone and/or its functional analogues. Such an increase in protein could improve sperm maturation[24] as indicated by histological analysis at this dose. A constant supply of cholesterol is necessary for androgen biosynthesis and its requirement for normal testicular activity is well established[4].In the groups treated with doses 134 or 267 mg/kg of BW, the testicular cholesterol level increased, possibly through increased steroidogenesis. Such stimulation of the steroidogenic pathway leads to an increased concentration of androgens. Alkaloids can increase testicular cholesterol levels[21]. Indeed, alkaloids can promote the absorption of cholesterol by testicular cells by modifying membrane fluidity[25] Therefore, the increase in testicular cholesterol recorded in this study could be due to the alkaloids present in the aqueous extract of *P. angolensis* wood. Testosterone, LH and FSH are hormonal markers of androgenicity[5]. Testosterone is the main androgen secreted by Leydig cells. It controls the growth and physiology of accessory sexual organs making it the main index of androgenicity[1].After treatment with 134 mg/kg, the extract increased the serum testosterone concentration compared to that in the normal and positive control groups. Since cholesterol is the precursor of androgen biosynthesis, the increase in serum testosterone levels could result from the increase in testicular cholesterol levels observed at this dose. Additionally, this increase could also be due to the presence of saponins which are known to increase endogenous testosterone levels probably through their steroidal nature or by increasing LH levels and binding to enzymes involved in steroidogenesis[4]. This result is even better than that of Kamtchouing*et al.*[26], who reported such an increase in the serum testosterone concentration in rats after treatment with a dose of 600 mg/kg of the aqueous extract of *Zingiber officinale* in rats. Prostatic acid phosphatase is a glycoprotein synthesized by prostate gland cells under the stimulation of testosterone[21]. According to Tagne *et al*.[27]**,** a reduction in circulating testosterone in rats leads to a reduction in prostatic acid

phosphatase activity. Consequently, the increase in prostatic acid phosphatase activity observed in the treated group at 134 mg/kg could be due to the increase in the blood concentration of testosterone or its functional analogues. Histology sections of the testes of the rats that were administered 134 and 267 mg/kg of the extract, revealed that the proliferation of testicular spermatozoa was comparable to that of the positive control group. These findings suggest that the histology of the organs improved in response to the extracts of *P. angolensis* at these two doses. Thus, the present metabolites revealed in the aqueous extract of the wood by Keumedjio*et al.* [9]might be responsible for its androgenic potential by increasing the secretory activity of androgens by sexual organs.

Subchronic toxicity allows us to highlight functional and/or anatomopathological alterations following repeated administrations of a bioactive substance [28]. The liver is the center of xenobiotic metabolism. Its damage can lead to an increase in the activity of serum transaminases, such as alanine transaminase (ALT) and aspartate transaminase (AST), proportional to the degree of the damage[29-31]. At the doses of 134 and 267 mg/kg the extract significantly reduced the activities of ALT and AST, compared to those in the positive control group but not significantly lower than those in the normal control group. Thelow activities of these enzymes could be due to the hepatoprotective effect of *P. angolensis* on rats, probably because of its antioxidant activities, as demonstrated in the aqueous extract of the plant[32] In addition, a decrease in serum triglyceride concentration was observed and could be the consequence of the protective effect of the extract on the liver since hepatotoxicity is generally associated with an increase in this parameter. This result is in line with the findings ofAchudume*et al.*[33]**,** who showed that the aqueous extract of *P. angolensis* had a hepatoprotective effect on rats. Indeed, the extract was able to treat liver damage caused by tetrachloride[33]. Elevated serum LDL and triglyceride levels are known to be associated with the risk of coronary heart disease. In our case, the doses 134 mg/kg and 267 mg/kg caused an increase in LDL compared to the normal control group while decreasing the levels of HDL and triglycerides. Serum creatinine and urea levels are excellent markers of kidney function. All the 3 doses of the extract did not change the serum creatinine level or decreased the serum urea level compared to those in the control groups. According to Amarasiri*et al.*[31], renal dysfunction leads to increased serum creatinine and urea levels. Our results revealed the proper functioning of the kidneys, particularly the glomeruli. Furthermore, it has been shown that the extract of *P. angolensis* has no harmful effects on the renal function of rats after acute oral exposure[11]. Additionally, an increase in protein synthesis can cause blood urea levels to decrease. The observed decrease in urea levels could also explain the anabolic effects of *P. angolensis* and could exclude its toxic effect on renal function at all doses. The hepatoprotective and nephroprotective effects of the extract were confirmed by histopathological analysis of the liver and kidneys, which showed no lesions in the rats treated with the 3 doses of the extract.

5. CONCLUSION

This study assessed the androgenic properties and subchronic toxicity of the aqueous extract of *P. angolensis* wood. The results showed that the extract at a dose of 134 mg/kg could stimulate androgen production but was not harmful to the rats.Overall, these results justify the traditional use of this plant by the *Baka* Pygmies for the treatment of male infertility and erectile dysfunction. Further studies should be performed to investigate the effects of this plant on sperm analysis to better understand the underlying mechanism of action.

ETHICAL APPROVAL

All authors hereby declare that "Principles of laboratory animal care" (NIH publication No. 85-23, revised 1985) were followed, as well as specific national laws where applicable. All experiments have been examined and approved by the appropriate ethics committee", the Join Institutional Review Board of Animal and Human Bioethics in Cameroon under the following reference number: BTC-JIRB2022-039.

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ABBREVIATIONS

AST: Aspartate Aminotransferase
ALT: Alanine Aminotransferase
BW: Body weight
ELISA: Enzyme-Linked Immuno-Sorbent Assay
HDL: High-density Lipoprotein
LD 50: Lethal dose 50
LDL: Low-density lipoprotein
LH: Luteinizing hormone
NO: Nitric oxide
OECD: Organization for Economic Cooperation and Development
PA: *Pycnanthus angolensis*
PACP: Prostatic Acid Phosphatase
RW: Relative weight
TE: Testosterone Enanthate