

REPRODUCTIVE-ENHANCING POTENTIAL OF METHANOL EXTRACT OF *SPHENOSTYLIS STENOCARPA* SEED ON MALE WISTAR RATS

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ABSTRACT

A study was undertaken on the reproductive-enhancing potential of Sphenostylis stenocarpa seed methanol extract on male Wistar rats. A total of 144 adult male rats were used for the experiment, divided into four groups (A – D), and replicated thrice. Group A served as the standard control, while Groups B, C, and D received three graded doses (800 mg/kg, 1200 mg/kg, and 1600 mg/kg) of the extract by oral intubation. The rats' gonad characteristics, sperm parameters, and hormonal analyses were determined weekly using standard procedures from Week 0 (day 1) to Week 12. Data obtained were analyzed using analysis of variance (ANOVA). Means with differences in p-values (< 0.05) were considered significant. The results showed overall dose and significant time-dependent differences in the mean weekly gonad characteristics of the male rats in the treatment groups compared to the control. The body weights of the male rats significantly reduced ($p < 0.05$), whereas the testes weights, gonad somatic index, sperm count, and sperm motility of the rats had a significant increase ($p < 0.05$). The hormone testosterone responded to the plant extract. The testosterone levels of all the treated rats had substantial gains. In conclusion, the methanol seed extract of Sphenostylis stenocarpa demonstrated an overall potency to enhance reproduction in male Wistar rats.

Keywords: *Sphenostylis stenocarpa*, reproductive indices, gonad somatic index, testosterone.

1. INTRODUCTION

Medicinal plant extracts in folkloric medicine are still prevalent in developing countries, attributable partly to poverty and illiteracy, which militate against the availability and accessibility of conventional medical services (Raji et al., 2006). According to Burns, 30% of all modern drugs are derived from plants (Burns, 2000). The African yam bean (*Sphenostylis stenocarpa*) is one such plant that has attracted much research interest for its nutritional and medicinal applications.

However, despite its potential, the plant remains under-exploited (Nyananyo and Nyingifa., 2011) because there is still a

shortage of literature regarding its nutritional and therapeutic value. It is cultivated mainly in tropical African countries like Nigeria, Ghana, and Cameroon. In Nigeria, it is mainly cultivated at a subsistence level in the southern part for its edible seeds. In the region, it is called "Okpochundu," "ijiriji," and "Azama" (Ekpo, 2006). The seeds can be roasted and eaten with palm kernels as snacks (Elegbede, 1998; Ezueh, 1984). The seed is a highly-priced food legume in southern Nigeria due to its high crude protein content (Asoiro and Ani, 2011).

Legumes are a rich source of proteins and essential amino acids. They are superior to

animal sources and provide an alternative and cost-effective source of protein to people, particularly in low-income countries. The amino acid content of the African yam bean (AYB) is reported to be higher than in most popular legumes like cowpea, pigeon pea, and Bambara groundnut. In one such study, amino acid profiling was reported. The amino acid profile (g/100g) of AYB showed values of 9.12g, 6.47g, 6.12g, and 5.05g for aspartic acid, arginine, lysine, and phenylalanine, respectively. These values were comparatively higher than those obtained in a similar study from Soybean and equally comparable with those obtained from a whole chicken egg (Omeire, 2012).

Several studies on the *Sphenostylis stenocarpa* plant have proven it to be of outstanding medical importance (Ubaka and Ukwe, 2010; Okonkwo et al., 2013; Onyeke and Ugwuoke, 2011; Uchegbu and Amulu, 2015). The methanolic seed extract of *S.stenocarpa* significantly reduced the blood glucose level but not the hypoglycemic level (Ubaka and Ukwe, 2010). The methanolic extract of *S. stenocarpa* seed showed anti-anaemic activity, therefore, has lent credence to the use of the plant seeds in the treatment and management of anaemia (Okonkwo et al., 2013). The plant extract's antifungal (Onyeke and Ugwuoke, 2011) and antioxidant activities (Uchegbu and Amulu, 2015) have also been reported.

Despite all these vast arrays of medicinal values exhibited by *S. stenocarpa*, no information exists regarding its reproductive potential. Hence, the present research aims to investigate the effect of the methanol seed extract of *Sphenostylis stenocarpa* on the reproductive indices of male Wistar rats.

2. MATERIALS AND METHODS

2.1 Procurement of *Sphenostylis stenocarpa*

The seeds of *Sphenostylis stenocarpa* were purchased from Nkwo Ibagwa Market in Nsukka. Identity was authenticated at the Taxonomy Unit, Department of Plant Science and Biotechnology, University of Nigeria, Nsukka, where a voucher specimen (PSBH/SSs/2019/008) was deposited in the departmental herbarium. The seeds were washed, air-dried for two weeks, and stored in an air-tight container until required.

2.2 Experimental Animals

A total of one hundred and forty-four (144) male rats aged between 3 – 4 months and weighing 30 – 40g were used. The entire animal models were purchased from the Genetics and Animal Breeding Unit, Department of Zoology and Environmental Biology, University of Nigeria, Nsukka. They were fed with feed (Vital Feeds® with 18.0 % crude protein and 2800 kcal/kg metabolizable energy) and water ad libitum. Weekly weights were recorded accordingly. All processes and procedures in handling the rats complied with the guidelines of the National Research Council (N.R.C., 2010). The rats were allowed to acclimatize for a week under standard photoperiodic conditions in clean cages in the Animal Breeding Unit, Department of Biochemistry, University of Nigeria, Nsukka.

2.3 Preparation of Methanol Extract

Two kilograms (2kg) of the dry seed of *Sphenostylis stenocarpa* was pulverized with a commercial blower. One thousand five hundred grams (1500g) of the powdered product was put into a conical flask to which 1500 ml of methanol was added. The mixture was allowed to stand for 24 hours and filtered using Whatman No one filter paper. The

percentage yield was calculated by dividing the weight of concentrated extract by the weight of dried-grinded seed and multiplying by 140. The extract was then concentrated using a rotary evaporator at a low temperature (30°C - 40°C). The concentrated extract was then used to prepare a stock solution of 1,600 mg/kg with Tween 80. After that, graded doses to be used for the experiment were calculated based on the body weight of the rat. This was kept in a refrigerator for phytochemical analysis and bioassay. Experimentation was conducted after determining the methanol extract's median lethal dose (LD₅₀) according to Lorke's method (Lorke, 1983).

2.4 Toxicity

The method of Lorke was used for the acute toxicity test (Lorke, 1983). Thirty Wistar rats were utilized in this study. The test involved two phases. In the first phase, the rats were grouped into three different groups of five rats each. They were administered 10, 100, and 1000 mg/kg body weight, respectively, and in the second stage, 1600, 2900, and 5000 mg/kg body weight of the extract were administered to the rats. The extract was administered by oral intubation. The methanol extract of *S. stenocarpa* seeds was not toxic at above 5000 mg/kg.

2.5 Experimental Design

The experiment lasted for 91 days. The 144 rats were broadly divided into a completely randomized design of four treatment groups (A – D), each replicated thrice. Group A was the normal control and received a standard grower's mash diet and distilled water. Groups B, C, and D received diet, distilled water, and three graded doses (800, 1200, and 1600 mg/kg) of methanol seed extracts of *S. stenocarpa* by oral intubation for 12 weeks. The three treatment doses were established after the LD₅₀ determination as a

6.25 times reduction of the LD₅₀ and subsequently increased by the addition of 400 mg/kg per body weight of rat. The animals were fed and watered *ad libitum*, and the cages were regularly cleaned. After acclimatization of the experimental rats, treatment with graded doses of extracts commenced. Reproductive parameters studied in male rats include body weight, testes weight, gonadosomatic index, sperm morphology, sperm count, and sperm motility (Idris et al., 2018). These parameters were determined before the commencement of treatment (week 0) and subsequently weekly (7 days intervals) by harvesting the testes for gonad characteristics and measuring the body weight. Similarly, three blood samples were obtained from the orbital sinus of each rat for the hormonal analyses before the commencement of treatment (week 0) and subsequently every week. The blood samples were used to ascertain the serum levels of testosterone (TL) using the method of Heywood (Heywood, 1980).

2.5 Statistical Analysis

Data collected were analyzed using analysis of variance (ANOVA). Preliminary data explorations were made using Kolmogorov-Smirnov to decide on suitable analytical approaches (parametric or non-parametric). Turkey HSD was used for the posthoc test. The level of significance for all tests was set at $p < 0.05$. Significant means were accepted at $p < 0.05$ and presented as mean \pm standard error of the mean. All analyses were done using Statistical Packages for Social Sciences (SPSS) version 23.0 (IBM Corporation, Armonk, USA).

3. RESULTS

3.1 Effects of methanol seed extract of *Sphenostylis stenocarpa* on the body weight of male Wistar rats

The effects of methanol seed extract of *S. stenocarpa* on the body weights (BWs) of

the male Wistar rats indicated an overall dose-dependent significant difference ($p < 0.05$) in the mean weekly BWs of the treated rats when compared to the control group (Table 1). The dose-dependent analysis showed that in week 0, the BWs of the treatment groups were significantly higher ($p < 0.05$) than the BWs of the control. In weeks 1, 4, 8, 9, 10, and 12, the BWs of the rats in the treatment groups were significantly lower ($p < 0.05$) compared to the control. Whereas, in weeks 2, 3, 5, 6, and 11, the BWs of the rats administered 1200 and 1600 mg/kg were significantly lower than the control. Similarly, minimal fluctuations were observed in the mean weekly BW of the rats in the treatment groups regarding the treatment duration compared with week 0.

3.2 Effects of methanol seed extract of *Sphenostylis stenocarpa* on the testes weight of male Wistar rats

As indicated in table 2, the dose-dependent analysis showed that the testes weight (TWs) of the rats in the treated group were relatively higher ($p < 0.05$) in weeks 2, 7, 8, 9, 10, and 12 when compared to the control. In the rats administered 1200 and 1600 mg/kg of the extract, there was a significant increase in the TWs of the rats in all weeks except in weeks 3, 4, 5, and 6, with a decrease compared to week 0. The time-dependent analysis showed minimal fluctuations in the mean weekly TWs of the rats in the treatment group.

3.3 Effects of methanol seed extract of *Sphenostylis stenocarpa* on the gonadosomatic index of male Wistar rats

The results presented in table 3 showed the effect of methanol seed extract of *Sphenostylis stenocarpa* on the gonadosomatic index (GSI) of male Wistar rats. The dose-dependent analysis showed that the GSI of rats in the treatment groups

was significantly higher ($p < 0.05$) than the GSI of the rats in control. In rats treated with 1200 and 1600 mg/kg of the extract, there were significant increases in the GSI at all weeks except weeks 3, 5 and 6, where the GSI of the rats significantly decreased compared to week 0.

3.4 Effects of methanol seed extract of *Sphenostylis stenocarpa* on the sperm motility of male Wistar rats

As presented in table 4, the dose-dependent analysis showed that the sperm motility (SM) of the male Wistar rats increased significantly in all weeks compared to week 0.

3.5 Effects of methanol seed extract of *Sphenostylis stenocarpa* on the sperm count of male Wistar rats

The results presented in Table 5 showed that in all the weeks, the sperm count (SC) of the rats in the treatment groups was significantly higher ($p < 0.05$) when compared to the control group, except in week 2, where there was no difference in the SC of rats administered 800 mg/kg of extract.

3.6 Effects of methanol seed extract of *Sphenostylis stenocarpa* on the testosterone level of male Wistar rats

The testosterone levels (TLs) of the male Wistar rats treated with methanol extract of *S.stenocarpa* were significantly higher ($p < 0.05$) in all weeks when compared with the control.

4. DISCUSSION

Plants contain many active compounds probably responsible for their different therapeutic properties exploited by folk medicine. However, these naturally occurring compounds may also exert a toxic effect on the development or normal functioning of the reproductive system

(Abdel-Magied et al., 2001). The present study shows that the methanolic leaf extract of *S. stenocarpa* enhanced the fertility of male Wistar rats.

In the present study, the extract was associated with a decline in the weight of the rats in a dose-dependent and duration-dependent manner. This was more obvious in the first and last months of the study. Our findings agree with the work of Gupta et al., where methanol stem extract of *Dendrophthoe falcate* decreased the body weights of male albino rats (Gupta et al., 2009).

It has been reported that sperm number and normal testicular histology are indices of fertility (Al-Sa'aidi et al., 2009; Etuk and Muhammad, 2009). In this study, the testes and gonad characteristics, namely sperm count, testes weight, gonad somatic index, and sperm motility, were affected depending on the extract concentration and duration of treatment. Each one of the parameters was significantly affected by extract concentration and course of treatment. The extract caused increases in testicular weights, sperm count, and sperm motility. Successful male fertility requires an adequate sperm count and adequate sperm motility. The present study corroborates the work of Ekere et al., where methanol extract of *Dracaena Arborea* in albino rats caused a dose-dependent significant increase in the testes' weights, gonad somatic index, sperm count, and sperm motility (Ekere et al., 2013). The gradual increase in the mean testicular weight of the treated rats is probably due to the increased activity in their testes. This may include increased testosterone secretions

(O'Keane et al., 1986). Mylchreest et al. reported that an increase in epididymal sperm number implicated elevated serum levels of testosterone in rats (Mylchreest et al., 2002). Meanwhile, the impact of food with high protein content in enhancing semen quality was reported by Oyeyemi and Okediran and recently confirmed by Mutwedu et al. (Oyeyemi and Okediran, 2007; Mutwedu et al., 2019). The ability of methanol extract of *S. stenocarpa* to increase sperm motility and counts, as observed in this study, is of great interest given that these parameters are determinants of the fertilizing capacity of sperm cells.

The testosterone hormones responded to the extract in male Wistar rats administered the *S. stenocarpa* extract. There were significant increases in the testosterone levels of all the treated rats. This study's findings agree with the report of Gamal et al., where ethanol extract of *Emex spinosa*, *Leptadenia pyrotechnica*, *Haloxylon salicornicum*, and *Ochradenus baccatus* significantly elevated the serum levels of testosterone in treated rats (Gamal et al., 2012).

5. CONCLUSION

The findings in this study prove that the methanol seed extract of *S. stenocarpa* orally administered to male Wistar rats increased the gonad characteristics, sperm parameters, and hormonal indices and has lent credence to the fact that it has fertility-enhancing ability in a dose-dependent manner. *S. stenocarpa* may yield similar or related results of reproductive enhancement in humans.

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Table 1: Weekly effect of different concentrations of methanol seed extract of *S. stenocarpa* on the body weight (g) of male albino rats

Conc. (mg/kg)	Duration (Weeks)												
	0	1	2	3	4	5	6	7	8	9	10	11	12
0.00	184.5± 1.45 ^{d2}	194.0± 1.11 ^{a9}	181.1± 1.66 ^{b1}	193.3± 2.74 ^{b8}	198.3± 1.92 ^{a13}	190.9± 1.01 ^{b4}	193.0± 1.15 ^{b7}	196.0± 0.05 ^{b10}	197.9± 1.75 ^{a11}	190.8± 0.10 ^{a3}	198.2± 1.00 ^{a12}	191.0± 1.50 ^{b5}	192.4± 5.10 ^{a6}
800	187.3± 2.56 ^{b3}	186.03± 2.56 ^{b1}	187.33± 2.56 ^{a3}	197.46± 2.56 ^{a11}	192.56± 2.56 ^{b8}	191.06± 2.56 ^{a6}	198.16± 2.56 ^{a12}	196.86± 2.56 ^{a10}	190.10± 2.56 ^{b5}	189.30± 2.56 ^{b4}	186.80± 2.56 ^{b2}	192.65± 2.56 ^{a9}	191.25± 2.56 ^{b7}
1200	186.21± 2.56 ^{c11}	139.33± 2.56 ^{d3}	129.55± 2.56 ^{d1}	178.33± 2.56 ^{c10}	159.16± 2.56 ^{c6}	155.56± 2.56 ^{c4}	187.80± 2.56 ^{d13}	186.43± 2.56 ^{c12}	169.85± 2.56 ^{d9}	156.00± 2.56 ^{c5}	160.43± 2.56 ^{c7}	168.55± 2.56 ^{c8}	132.70± 2.56 ^{c2}
1600	191.96± 2.56 ^{a13}	148.26± 2.56 ^{c7}	131.67± 2.56 ^{c2}	173.33± 2.56 ^{d9}	136.96± 2.56 ^{d5}	155.16± 2.56 ^{d8}	187.96± 2.56 ^{c12}	186.00± 2.56 ^{d11}	179.00± 2.56 ^{c10}	138.05± 2.56 ^{d6}	133.30± 2.56 ^{d4}	132.80± 2.56 ^{d3}	130.65± 2.56 ^{d1}

Values as mean ± standard error. Values with different small letter alphabet superscripts down a column for each solvent significantly differed between concentrations (p<0.05). In contrast, different numeric superscripts across a row significantly differed for durations (p<0.05).

Table 2: Weekly effects of different concentrations of methanol seed extract of *S. stenocarpa* on the testes weight of male albino rats

Conc. (mg/kg)	Duration (Weeks)												
	0	1	2	3	4	5	6	7	8	9	10	11	12
0.00	5.13± 0.40 ^{a12}	4.53± 0.51 ^{a10}	3.43± 0.20 ^{d2}	5.63± 0.51 ^{b13}	3.40± 0.43 ^{c1}	3.63± 0.15 ^{b5}	3.60± 0.26 ^{c4}	3.70± 0.20 ^{d6}	4.40± 0.20 ^{d8}	3.55± 0.05 ^{d3}	3.50± 0.20 ^{d9}	3.15± 0.05 ^{d7}	3.70± 0.20 ^{d11}
800	5.10± 1.30 ^{b2}	5.66± 0.30 ^{b4}	5.73± 0.40 ^{a5}	5.83± 0.15 ^{a7}	6.83± 0.30 ^{a8}	6.63± 0.35 ^{a6}	5.36± 0.15 ^{a3}	7.63± 0.15 ^{a12}	7.20± 0.10 ^{a9}	7.75± 1.15 ^{a13}	4.85± 0.05 ^{c1}	7.25± 0.15 ^{b10}	7.60± 0.10 ^{a11}
1200	4.90± 0.60 ^{d5}	5.80± 0.20 ^{d9}	5.03± 0.66 ^{c7}	4.08± 0.55 ^{c4}	4.03± 0.37 ^{b3}	2.87± 0.35 ^{d1}	3.63± 0.15 ^{b2}	5.03± 0.51 ^{c6}	6.10± 0.30 ^{b11}	5.90± 0.10 ^{b10}	5.43± 0.20 ^{a8}	7.20± 0.20 ^{c12}	7.25± 0.15 ^{c13}
1600	5.03± 0.30 ^{c5}	5.10± 0.43 ^{c7}	5.63± 0.45 ^{b11}	4.86± 0.55 ^{d4}	2.86± 0.45 ^{d1}	2.96± 0.30 ^{c2}	3.47± 0.65 ^{d3}	5.06± 0.35 ^{b6}	5.40± 0.20 ^{c8}	5.50± 0.10 ^{c10}	5.40± 0.40 ^{b9}	8.10± 0.10 ^{a13}	7.45± 0.15 ^{b12}

Values as mean ± standard error. Values with different small letter alphabet superscripts down a column for each solvent significantly differed between concentrations (p<0.05). In contrast, different numeric superscripts across a row significantly differed for durations (p<0.05).

Table 3: Weekly effects of different concentrations of methanol seed extract of *S. stenocarpa* on the gonad somatic index of male albino rats

Conc. (mg/kg)	Duration (Weeks)												
	0	1	2	3	4	5	6	7	8	9	10	11	12
0.00	5.52± 0.14 ^{a13}	4.78± 0.06 ^{d8}	4.64± 0.09 ^{d6}	5.31± 0.13 ^{b12}	4.19± 0.16 ^{d1}	4.58± 0.10 ^{d5}	4.40± 0.20 ^{d2}	4.44± 0.10 ^{d3}	4.75± 0.06 ^{d7}	4.48± 0.02 ^{d4}	4.79± 0.75 ^{d10}	4.79± 0.04 ^{d9}	5.06± 0.10 ^{d11}
800	5.43± 0.45 ^{c3}	5.66± 0.03 ^{c4}	5.84± 0.27 ^{c6}	6.05± 0.12 ^{a8}	6.16± 0.13 ^{a9}	6.05± 0.09 ^{a7}	5.18± 0.05 ^{a1}	6.37± 0.15 ^{a11}	6.41± 0.03 ^{a12}	5.91± 0.36 ^{c5}	5.27± 0.03 ^{c2}	6.36± 0.09 ^{c10}	6.59± 0.03 ^{c13}
1200	5.46± 0.44 ^{b4}	7.68± 0.24 ^{a12}	7.58± 0.31 ^{b11}	5.14± 0.27 ^{c3}	5.71± 0.19 ^{c6}	5.08± 0.38 ^{c2}	4.51± 0.17 ^{c1}	5.52± 0.04 ^{c5}	6.54± 0.18 ^{b8}	6.99± 0.30 ^{b9}	6.49± 0.10 ^{b7}	7.26± 0.39 ^{b10}	9.25± 0.38 ^{b13}
1600	5.27± 0.10 ^{d4}	6.85± 0.23 ^{b8}	7.90± 0.24 ^{a10}	5.12± 0.23 ^{d2}	5.77± 0.48 ^{b6}	5.25± 0.07 ^{b3}	4.53± 0.27 ^{b1}	5.60± 0.27 ^{b5}	5.81± 0.16 ^{c7}	7.63± 0.49 ^{a9}	7.81± 0.44 ^{a11}	9.87± 0.28 ^{a13}	9.57± 0.61 ^{a12}

Values as mean ± standard error. Values with different small letter alphabet superscripts down a column for each solvent were significantly different between concentrations (p<0.05), while different numeric superscripts across a row were significantly different for durations (p<0.05).

Table 4: Weekly effects of different concentrations of methanol seed extract of *S. stenocarpa* on the sperm motility of male albino rats

Conc. (mg/kg)	Duration (Weeks)												
	0	1	2	3	4	5	6	7	8	9	10	11	12
0.00	23.67± 2.51 ^{d12}	21.67± 2.51 ^{d7}	20.00± 1.00 ^{d3}	21.00± 3.60 ^{d5}	22.67± 2.30 ^{d10}	21.33± 1.52 ^{d6}	22.00± 2.00 ^{d9}	21.67± 3.21 ^{d8}	33.00± 2.00 ^{d13}	20.00± 6.55 ^{d4}	17.33± 2.51 ^{d1}	18.33± 2.88 ^{d2}	23.67± 2.30 ^{d11}
800	24.33± 1.52 ^{b1}	59.00± 1.00 ^{a3}	55.00± 1.00 ^{a2}	72.67± 1.52 ^{a5}	73.00± 2.64 ^{a6}	68.33± 2.08 ^{a4}	82.67± 1.52 ^{a9}	83.00± 2.64 ^{a10}	81.33± 2.08 ^{a7}	85.00± 2.64 ^{a11}	81.33± 3.05 ^{a8}	83.00± 2.64 ^{a10}	85.33± 2.08 ^{a12}
1200	24.00± 4.58 ^{c1}	42.67± 8.08 ^{c4}	44.33± 6.02 ^{c6}	43.33± 1.63 ^{b5}	35.00± 2.64 ^{b2}	42.33± 8.62 ^{b3}	54.67± 1.52 ^{b7}	57.33± 5.50 ^{b8}	57.67± 3.51 ^{b9}	73.67± 6.11 ^{b10}	74.67± 4.93 ^{b11}	74.67± 5.13 ^{b12}	80.33± 1.52 ^{c13}
1600	28.00± 3.60 ^{a1}	51.67± 1.52 ^{b5}	51.67± 1.52 ^{b5}	35.00± 2.64 ^{c3}	30.00± 3.00 ^{c2}	40.33± 5.50 ^{c4}	52.67± 1.52 ^{c6}	54.00± 3.60 ^{c7}	56.00± 5.56 ^{c8}	62.00± 2.64 ^{c9}	73.00± 2.64 ^{c10}	84.00± 2.64 ^{c12}	81.33± 3.78 ^{b11}

Values as mean ± standard error. Values with different small letter alphabet superscript down a column for each solvent were significantly different between concentrations (p<0.05), while different numeric superscripts across a row were significantly different for durations (p<0.05).

Table 5: Weekly effects of different concentrations of methanol seed extract of *S. stenocarpa* on the sperm count of male albino rats

Conc. (mg/kg)	Duration (Weeks)												
	0	1	2	3	4	5	6	7	8	9	10	11	12
0.00	30.67± 3.05 ^{d12}	22.67± 2.51 ^{d2}	30.00± 1.00 ^{a11}	26.33± 7.02 ^{d5}	28.67± 4.16 ^{d9}	29.33± 1.52 ^{d10}	28.67± 1.52 ^{d8}	25.00± 4.00 ^{d4}	28.00± 1.00 ^{d7}	31.00± 7.00 ^{d13}	23.00± 1.00 ^{d3}	22.00± 3.00 ^{d1}	27.00± 1.00 ^{d6}
800	37.00± 4.35 ^{a1}	68.00± 1.00 ^{a3}	65.00± 1.00 ^{a2}	82.67± 1.52 ^{a5}	83.67± 1.52 ^{a6}	77.67± 2.51 ^{a4}	92.00± 1.00 ^{a18}	91.67± 1.52 ^{a7}	92.00± 1.00 ^{a8}	96.00± 2.00 ^{a12}	92.33± 1.52 ^{a9}	93.00± 1.00 ^{a10}	94.33± 3.21 ^{a11}
1200	37.33± 1.52 ^{b1}	48.00± 2.64 ^{b5}	52.67± 3.21 ^{c6}	47.67± 2.08 ^{b3}	45.67± 1.52 ^{b2}	47.67± 2.51 ^{c4}	55.67± 5.50 ^{c7}	61.33± 6.42 ^{b8}	68.00± 3.60 ^{b9}	83.00± 6.00 ^{b11}	82.00± 5.29 ^{b10}	84.00± 5.00 ^{c12}	90.67± 2.08 ^{c13}
1600	33.00± 1.00 ^{c1}	39.67± 4.16 ^{c2}	53.00± 2.64 ^{b6}	46.33± 2.08 ^{c4}	42.00± 1.73 ^{c3}	52.00± 2.64 ^{b5}	56.67± 7.50 ^{b7}	58.00± 2.64 ^{c8}	60.00± 3.60 ^{c9}	69.00± 1.00 ^{c10}	79.00± 3.00 ^{c11}	92.00± 2.00 ^{b13}	91.60± 3.60 ^{b12}

Values as mean ± standard error. Values with different small letter alphabet superscripts down a column for each solvent were significantly different between concentrations (p<0.05), while different numeric superscripts across a row were significantly different for durations (p<0.05).

Table 6: Weekly effects of different concentrations of methanol seed extract of *S. stenocarpa* on the testosterone of male albino rats

Conc. (mg/kg)	Duration (Weeks)												
	0	1	2	3	4	5	6	7	8	9	10	11	12
0.00	0.57± 0.01 ^{b11}	0.60± 0.41 ^{d12}	0.43± 0.05 ^{d6}	0.46± 0.01 ^{d8}	0.57± 0.06 ^{d10}	0.46± 0.04 ^{d9}	0.38± 0.05 ^{d4}	0.44± 0.05 ^{d7}	0.39± 0.03 ^{d5}	0.35± 0.03 ^{d2}	0.38± 0.04 ^{d3}	0.38± 0.04 ^{d3}	0.35± 0.02 ^{d1}
800	0.55± 0.03 ^{d1}	0.70± 0.02 ^{c3}	0.64± 0.05 ^{c2}	0.74± 0.02 ^{b6}	0.84± 0.05 ^{c11}	0.76± 0.01 ^{b7}	0.83± 0.05 ^{a10}	0.78± 0.02 ^{b8}	0.73± 0.01 ^{c4}	0.73± 0.01 ^{c4}	0.74± 0.06 ^{c5}	0.83± 0.01 ^{c9}	0.87± 0.10 ^{c12}
1200	0.60± 0.08 ^{a2}	2.57± 0.25 ^{a10}	2.21± 0.28 ^{b9}	0.74± 0.08 ^{a4}	0.97± 0.11 ^{b5}	1.06± 0.15 ^{a6}	0.51± 0.02 ^{c1}	0.65± 0.05 ^{c3}	1.19± 0.21 ^{b7}	3.05± 0.05 ^{a11}	1.65± 0.15 ^{b8}	3.15± 0.45 ^{b12}	3.81± 0.31 ^{b13}
1600	0.56± 0.05 ^{c1}	2.34± 0.16 ^{b9}	2.58± 0.16 ^{a11}	0.73± 0.04 ^{c4}	1.09± 0.14 ^{a6}	0.70± 0.10 ^{c3}	0.63± 0.02 ^{b2}	0.87± 0.25 ^{a5}	1.40± 0.20 ^{a8}	1.15± 0.35 ^{b7}	2.46± 0.25 ^{a10}	3.90± 0.30 ^{a12}	9.15± 0.75 ^{a13}

Values as mean ± standard error. Values with different small letter alphabet superscripts down a column for each solvent were significantly different between concentrations (p<0.05), while different numeric superscripts across a row were significantly different for durations (p<0.05).