

Effect of aqueous root extract of *Fadogia andersonii* on sperm count and motility in adult male Wistar rats

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ABSTRACT

The effect of administration of aqueous root extract of *Fadogia andersonii*(FA) was evaluated on sperm count, motility and epididymal weight of the adult male Wistar rats. Twenty rats were randomly divided into four groups (Group A, B, C and D) of 5 rats each. Group A served as the control and was administered 0.9% normal saline, group B, C and D served as the treatment groups and received oral dose of 100 mg/kg, 200 mg/kg and 400mg/kg of the extract, respectively for 20 days. Sperm counts, sperm motility and epididymal weight were evaluated. Results of the experiment revealed a dose dependent significant increase ($P < 0.05$) in the sperm count and sperm motility. Thus, the plant had a spermatotrophic action demonstrated by the increase in sperm count and sperm motility.

Keywords: *Fadogia andersonii*, sperm count, sperm motility, epididymal weight.

INTRODUCTION

Many plants are used for medicinal purposes. Medicinal herbs and plants extracts are now generally considered as effective medicines that play a major role in modern pharmacy [1]. In 1985, the world health organization estimated that about 80% of the world's population relies on medicinal plant for their primary health care needs. Although herbal medicine has existed since the dawn of time, our knowledge of how plants actually affect physiological function remains largely unexplored [2]. The traditional Hausa people believe that the consumption of *F.andersonii*, natively called *Gagayin* Hausa language can enhance fertility in males. They also believe that taking *Fadogia andersonii* can stimulate erection in males and enhance sexual performance of males that are impotent. However, this has not been scientifically proven and the mechanism by which this plant works remain unknown. *F.andersonii* is a plant that belongs to the genus family of Rubiaceae. There are many species in this family with evergreen leaves; they grow in a humid climatic environment. *F. andersonii* is a popular African medicinal plant, which has long been used in Africa in treatment of diseases including amoebic dysentery. Members of this family have been studied for the treatment of burns, inflammation, wound and stomach ulcer etc., [3].

MATERIALS AND METHODS

1.1 Experimental design and treatment of animals

1.1.1 Acute toxicity studies of *Fadogia andersonii*

Acute toxicity (LD₅₀) of *F. andersonii* was determined using the method of Lorke [4]. The study of *Fadogia andersonii* on Wistar rat toxicity was divided into two phases. Nine Wistar rats were used in the first phase in 3 divided groups of 3 each named group A, B and C. Group A received extract doses of 10 mg/kg body weight orally, while group B and C received extract doses of 100 and 1000 mg/kg body weight by oral route respectively. The treatment animals were observed for 24 hours for sign of discomfort or death. In the second phase, four Wistar rats were divided into four groups, named group D, E, F and G. Group D received the extract at a dose of 600 mg/kg body weight orally, group E, F and G received the extract at a dose of 1000, 1600 and 2900 mg/kg body weight respectively. The animals were also observed for 24 hours for sign of discomfort or death. No sign of discomfort or death was observed during the period of observation. Therefore, the extract was considered relatively non-toxic even at doses higher than 5000 mg/kg [4].

1.1.2 Experimental animals

Twenty mature male albino Wistar rats weighing between 160 – 190 g raised in the Animal House of the Department of Human Physiology, Ahmadu Bello University, Zaria, Nigeria were used for the study. They were housed in well-ventilated plastic cages and maintained at normal room temperature of 21 ± 2 °C. They were provided with food and water *ad libitum*.

1.1.3 Identification of plant and extract preparation

The *Fadogia andersonii* plant was authenticated and given herbarium/voucher number 588. The roots were then shaded-dried and pounded into powder and extraction done using the method of cold maceration [5]. The powder was poured into a conical flask and one litre of distilled water was added to it and was shaken for 1 hour, and then allowed to stand overnight. It was then filtered using a filter paper and the filtrate was poured into an evaporating dish, and concentrated with water bath (45-50°C) to granules. The granules/extract was then scrape off and preserved in a sample bottle.

1.2 experimental procedure

A total of 20 Wistar rats were used for the experiment. They were divided into four groups of five animals each. Group A served as control and given 5 ml/kg of 0.9% saline, while groups B, C and D were treated with *F. andersonii* extract at oral doses of 100, 200, 400 mg/kg/day for 20 days, respectively. After 24 hours from the last treatment, the animals were used for the experiment. Each rat was euthanized in a chloroform chamber. After euthanasia the rat was dissected and the epididymis was exposed by scrotal incisions and transferred into petri dish. The weight of the epididymis was recorded for each rat.

1.3 Sperm analysis

The epididymis was crushed using a blunt forceps in a petri dish and 1 ml of normal saline was added to semen and mixed thoroughly using a syringe to draw and release the mixture continuously [6]. The semen mixture was then sucked into a red blood cell pipette to the 0.5 mark, then normal saline was sucked up to the 101 mark. The normal saline in the stem of the pipette was discarded and the content of the bulb of the pipette was mixed thoroughly. A drop of the mixture was placed on the counting chamber which then spreads under the cover slip by capillary action. The counting chamber was then mounted on the slide stage of the microscope and viewed under x 40 magnification. A grid system divides the counting chamber into five major squares each containing 16 smaller boxes. The count included all the sperm cells within the five major squares using the top and right or left and bottom system of counting as described by Vermaet *al.* (2002) and Zaveneid and Polakoski (1977) [6-7].

The sperm count for a rat was calculated as = $n \times 1 \times 10^{-6}$ /mL of semen.

1.4 Sperm motility

A drop of the semen mixture was placed on a glass slide using 2 ml syringe, the preparation was placed on a microscope. Sperm motility was assessed as described by Sonmez *et al.* (2007) [8]. The motility of epididymal sperm was evaluated microscopically within 2–4 min of their isolation from the caudal epididymis and data were expressed as percentages of fast motile, slow motile and non-motile spermatozoa. The percentage of motility was evaluated visually at X 40 magnification.

1. Fast movement sperm cells
2. Slow movement sperm cells
3. No movement or immotile sperm cells

1.5 Statistical analysis

The results obtained were presented as mean \pm SEM. The data were analyzed using ANOVA and Tukey's post hoc test to determine the level of significance between the control and experimental groups. Values of $P < 0.05$ were considered to be of statistical significance.

RESULTS

The result showed that mean sperm counts in the treatment groups were higher ($P < 0.05$) when compared to the control groups (Table 1). The mean epididymal weight in group C was significantly higher than the control group ($P < 0.05$).

Table 1: Effects of oral aqueous root extract of *Fadogia andersonii* on sperm count and epididymal weight in adult male Wistar rats

Parameters	Group A (Control)	Group B 100mg/kg	Group C 200mg/kg	Group D 400mg/kg
Sperm count (Million cells/mL)	30.20 \pm 0.583	40.40 \pm 1.700 ^a	55.60 \pm 2.712 ^b	68.60 \pm 2.619 ^c
Epididymal weight (g)	0.134 \pm 0.021	0.260 \pm 0.029	0.260 \pm 0.018 ^a	0.200 \pm 0.018

Data were expressed as Mean \pm SEM. $N = 5$
a, b, c = Means with different superscript letters are significantly ($P < 0.05$) different

There was significant increase in mean fast sperm motility of the treatment groups as compare to the control group ($P < 0.05$). While there was relative decrease in the slow motile sperm cell of the treatment groups (except group B) as when compared to the control group that is statistically significant ($P < 0.05$). Also there was considerable decrease in immotile sperm cells in the treatment groups ($P < 0.05$) when compared to the control group (Figure 1).

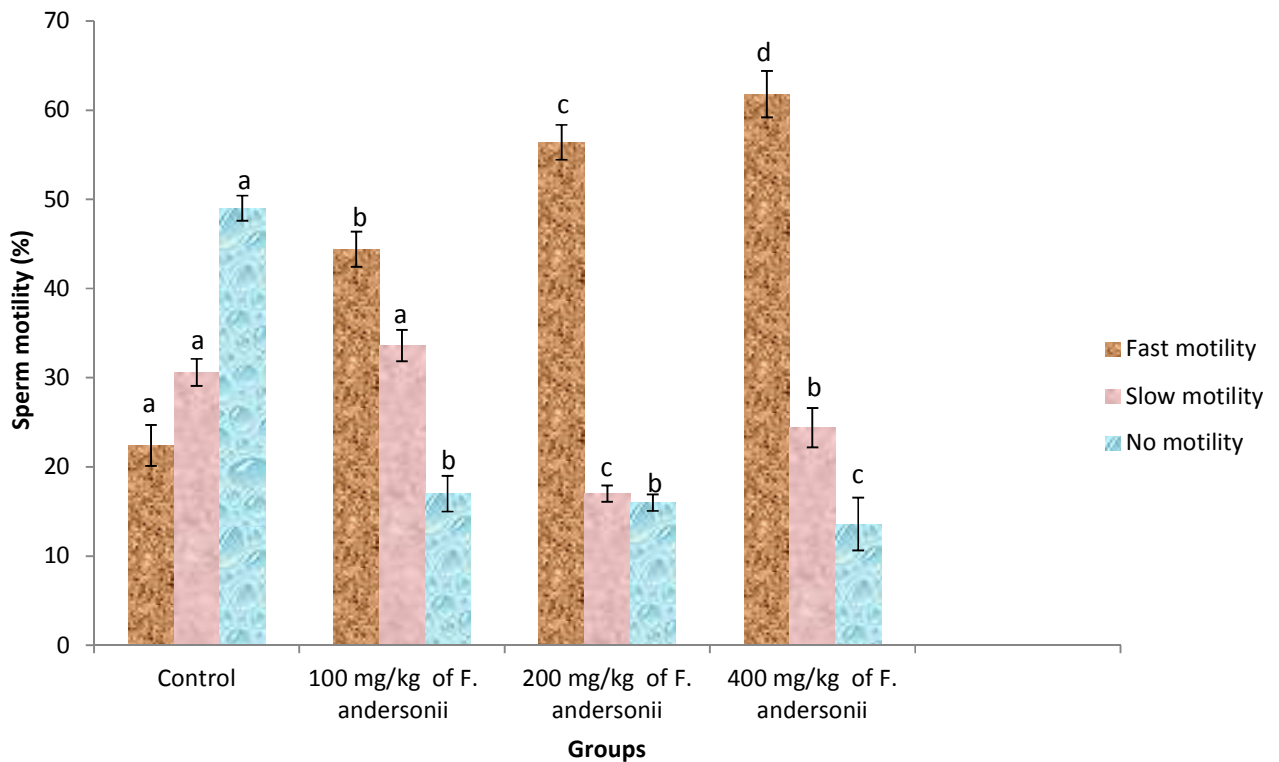


Figure 1: Effects of oral aqueous root extract of *Fadogia andersonii* on sperm count
a, b, c, d = Means with different superscript letters are significantly ($P < 0.05$) different

DISCUSSION

Oral administration of aqueous root extract of *F. andersonii* for 20 days in adult male Wistar rats caused a significant increase in sperm count, epididymal weight and fast motility, but a significant decrease in slow sperm movement and immotile sperm cell count.

The dose dependent increase in sperm count in the treatment groups when compared to the control group was probably induced by the indole alkaloid component of *F. andersonii* extract which had earlier been reported by Aly (2005) [9], who observed that indole alkaloids act on the pituitary gland to stimulate the synthesis and release of follicle stimulating hormone and luteinizing hormone. The observation made in the present study is consistent with the findings of Emilia *et al.* (2002) and Gonzales *et al.* (2006) [10][11], they observed that daily oral administration of high doses of trans-resveratrol increased sperm count and sperm motility and black *Lepidiummeyenii* increased sperm count and sperm motility in rats which could be due indole alkaloid present in them. Similarly, Unnithan and Tandon (1982) [12] reported that the indole alkaloid acts on the hypothalamo-pituitary gland, to increase secretion of gonadotropin releasing hormone and promote spermatogenesis.

Wan *et al.* (2007) [13] also reported that indole alkaloid induces spermatogenesis in rats through the activation of cAMP responsive element modulators (CREM). The significant increase in mean values of epididymal weight in the treatment group as compared to the control group may due to a possible effect of the extract component on the epididymal tissue to enhance the synthesis of adenosine triphosphate (ATP) [9], to increase the growth of gonadal tissue due to the activity of indole alkaloid.

The significant increase in sperm fast motility in the treated rats as compared the control rats may be due to the ATP generating effect of the indole alkaloid on the activities of the sperm cells. The extract of FA also contained triterpened and saponins. Saponins are spermicidal through plasma membrane disintegration and dissolution of acrosomal cap of a sperm cells previous study described by Pakrashiet *al.* (1991) [14]. It is possible that the spermicidal effect of saponins was ameliorated by the effect of indole alkaloid in this study.

CONCLUSION

Based on the results of the experiment, it was concluded that the aqueous root extract of FA increases sperm count, sperm motility and epididymal weight. This means that FA has a profertility effect on testicular function in rats.

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