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# THE EFFECTS OF INCREASING AQUEOUS ROOT EXTRACT OF *MORINGA OLEIFERA* ON SPERM PRODUCTION OF ALBINO RATS

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## ABSTRACT

*The phytochemical effects of aqueous extract of root on sperm production of White albino rats were investigated. The rats were divided into five treatment dose levels; 0 (5.0 ml saline); 5.0; 10.0; 15.0 and 20.0 mls consisting five (5) animals per treatment and administered orally for 10 days using aqueous extract of Moringa oleifera roots. The extract tested positive to alkaloid, flavonoids, saponin and cyanogenic glycosides as the active ingredient and showed negative to tannin and oxalates. The results also showed increased ($p<0.05$) mass activity, progressive motility, sperm concentration and semen output in the treated rats. The histological examination revealed no lesion on the testis. The findings of this study indicated that the active ingredient from Moringa oleifera could significantly increase sperm production.*

Keywords: active ingredient, aqueous root extract, *Moringa oleifera,* phytochemical, semen output

## INTRODUCTION

*Moringa oleifera*, Lam (*M. oleifera*), also known as *Moringa pterygosperma* Gaertn, Drumstick tree (Engl.), Arunggai (Pang.), is a member of the Moringaceae family of perennial angiosperm plants, which includes 12 other species (Olson, 2002). It is a native of the Indian subcontinent, where its various parts have been utilized throughout history as food and medicine. The therapeutic use of *M. oleifera* parts in the Indian subcontinent dates back to antiquity (Mbikay, 2012). It is now cultivated in all tropical and sub-tropical regions of the world. *Moringa oleifera* is an edible plant with a wide variety of nutritional and medicinal values which have been attributed to its roots, bark, leaves, flowers, fruits, and seeds (Ramachandran *et al.,* 1980; Anwar *et al*., 2007; Kumar *et al*., 2010). Since application of artificial insemination in animals, there has been a growing interest and necessity for more knowledge concerning semen technology improvement. However, the repeated use of compounds such as progestagens, testosterone and THC (tetrahydrocannabinol) have been reported to reduce fertility over a time period (*Sell et al*., 2000), the condition being attributed to immunity against the compounds (Atterwill and Flack, 1992; Wildeus, 1995; Considine, 2001). Phytochemical analyses have shown that moringa leaves are particularly rich in potassium, calcium, phosphorous, iron, vitamins A and D, essential amino

acids, as well as such known antioxidants such as â-carotene, vitamin C, and flavonoids (Bennett *et al*., 2003; Aslam *et al.,* 2005; Manguro and Lemmen, 2007; Amaglo *et al*., 2010; Gowrishankar et al., 2010). There is no scientific report on the use of moringa extract in white albino rats as aphrodisiac substances as reported for sugarcane (Sas, 1990). The experiment was therefore aimed at determining the effects of root extract on the reproductive performance of White albino rats.

## MATERIALS AND METHODS

### Plant Materials and Extraction

Roots of *Moringa oleifera* Linn were collected from Ologoji Farm Settlement, Ijan- Ekiti, Ekiti State. The roots were washed, shade-dried and crushed, 250g of powdered material was soaked using 1 litre of distilled water for 48 hours in a similar manner reported by Stout and Tolman (1941) and Lovett *et al.* (1981). The extracts were recovered using Watman filter paper.

### Animal and Management

Twenty- five (25) adult albino rats with body weight ranging from 200g to 250g were obtained from Biological Sciences Department, Ondo State University of Science and Technology, Okitipupa, Ondo State, Nigeria. The rats were housed (5 rats per cage) in metallic boxes (cages) in the Department of Biological Sciences, Ondo State University of Science and Technology, Okitipupa, Ondo State, Nigeria. The rats were allowed free access to normal rats feed and drinking water. They were allowed to acclimatize for 2 weeks, prior to the commencement of the experiment.

### Experimental Design

The rats were divided into five (5) groups of five (5) rats each. Group I served as control, Group II, Group III, Group IV and Group V were study groups. Rats in group II, III, IV and V were administered orally with 5.0, 10.0, 15.0 and 20.0 ml of aqueous extract of *Moringa oleifera* roots respectively. Rats in group I were given 5.0ml saline instead of the extract. The administration of the aqueous extract was done at 9.00 am in the morning for ten (10) consecutive days. On the twenty- eighth day, the rats were sacrificed by decapitation, the testes dissected out, the content dispelled and the spermatogenic analysis were carried out according to Bearden and Fuquay (1999). The surrounding tissues were removed, testis were blotted on filter paper and weighed quickly on a sensitive balance and fix in Bouin's fluid for 24hours. The paraffin- embedded tissues were cut at 6ìm and stained with haematoxylin-eosin solution for histological observation.

### Phytochemical Screening

To determine the presence of alkaloids, glycosides, flavonoid, tannins, oxalates, saponin, a preliminary phytochemical study (colour reactions) with root extracts was performed according to the procedures of (Wall *et al* (1952); Harborne (1973); Tracey *et al* (1980); Trease and Evans (1985); Fansworth (1999).

### Statistical analysis

Data generated were analyzed using the Completely Randomized Design (CRD) and means tested (Duncan, 1955) in a COSTAT program.

## RESULTS

The phytochemical screening of aqueous of *Moringa oleifera* root extract revealed the presence of active ingredient as shown in Table 1.

Table1: Summaries of Results of the Phytochemical Tests

| Constituents | Test | Inference |
|---|---|---|
| Alkaloid | Dragendorff's reagent | + |
| Flavonoids | Shibata's reac tion | + |
| Tannin | Ferric chloride test | - |
| Saponin | Frothing test | + |
| Oxalates | Anion analysis | - |
| Cyanogenic glycosides | Hydrogen cyanides test | + |

Key (+) = presence, (-) = absence

No mortality and slight behavioural changes were observed in the treated groups. The present study revealed that the aqueous extract of *Moringa oleifera* root produces androgenic effect. Treatment of rats with aqueous extract for 10 days exhibited sexual activity ($p<0.05$) as presented on Table 2. The semen colour of control (5.0 ml saline) and low dose (5.0 ml) of the extract produced white colour, whereas, higher doses (10.0 ml, 15.0 ml and 20.0 ml) of the extract produced milky colour. The treated rats had higher ($p<0.05$) mass activity, mass motility, sperm concentration, total sperm cell and liveability than the control. These trends showed that the moringa extract enhanced the sperm viability and quality. No significant cytoarchitectural changes were observed in the testes and also there was no abnormal morphological modifications of the spermatozoa in both the control and treated groups. The sperm count was also normal in the samples taken from the caudal epididymis. The histological observation of the seminiferous tubules was also normal. Therefore the extract did not have a detectable change in the cytoarchitecture of rats in the treated groups.

Table 2: Mean spermiogramic characteristics of semen of white albino rats influenced by the extract

| Parameters | TREATMENT (ml) | | | | | |
|---|---|---|---|---|---|---|
| | 5.0ml saline | 5.0ml | 10.0ml | 15.0ml | 20.0ml | S.E |
| Ejaculate colour | White | White | Milky | Milky | Milky | - |
| Semen volume (ml) | 9.47 $^{c}$ | 11.13 $^{b}$ | 12.23 $^{ab}$ | 12.53 $^{a}$ | 13.17 $^{a}$ | 1.303 |
| Mass activity | 2.49 $^{b}$ | 2.76 $^{ab}$ | 2.87 $^{a}$ | 3.12 $^{a}$ | 3.36 $^{a}$ | 0.637 |
| Mass motility (%) | 58.34 $^{c}$ | 61.25 $^{c}$ | 67.47 $^{b}$ | 71.48 $^{ab}$ | 74.37 $^{a}$ | 2.372 |
| Sperm concentration (x10 $^{6}$/ml) | 7.54 $^{c}$ | 9.46 $^{b}$ | 11.23 $^{ab}$ | 11.46 $^{a}$ | 12.36 $^{a}$ | 0.051 |
| Total sperm cell (x10 $^{7}$/ml) | 7.14 $^{d}$ | 10.53 $^{c}$ | 13.73 $^{bc}$ | 14.36 $^{ab}$ | 16.28 $^{a}$ | 1.104 |
| Liveability (%) | 64 $^{bc}$ | 66 $^{bc}$ | 70 $^{ab}$ | 73 $^{a}$ | 75 $^{a}$ | 2.423 |
| Unripe sperm cells (%) | 40 | 40 | 38 | 40 | 42 | 0.125 |

a b, c,d : values along the same row with different superscripts are significant ($p<0.05$).

S.E = Standard error

## DISCUSSION

The result of the phytochemical studies showed that the aqueous extract contains alkaloid, flavonoids, saponin and cyanogenic glycosides as the active ingredient and absence of others in the aqueous extract (Table 1). The presences of these active ingredients are in line with other workers who reported that it contains active principles. The phytochemical composition of *M. oleifera* parts have been shown to vary significantly among regions and seasons (Iqbal and Bhanger, 2006; Juliani *et al.* 2009). The ability of the animal to withstand the toxic effect of the extract has been demonstrated since the animals could tolerate the aqueous extract. This result justifies the use of this plant root in herbal medicine in India and South Africa. The increased in mass activity was in line with the reports of Aslam *et al*. (2005), Amaglo *et al*. (2010) and Gowrishankar, *et al.* (2010) who reported the correlation between the flavonoids and the sperm production and that moringa inhibit 6-beta-hydroxylation of testosterone thereby produces adrogenic effect by enhancing sexual drive through increased serum and testicular testosterone levels

(Cajuday and Pocsidio, 2010). The semen volume observed in this study was in agreement with the values reported by Strader, *et al* (1999) for rats. However, the increased in the semen volume could be linked with the ability of the extract to induce sperm production (Adaikan and Ng, 2000). The higher mass activity, mass motility and sperm concentration of spermatozoan has been reported to have correlation (Oyeyemi *et al.* 2001). Also, the higher mass activity and motility of semen observed in this study is in line with the report of Rege, *et al.* (2000) that mass activity is usually high when higher concentration of spermatozoa are produced resulting in a higher wave motion. Hafez, *et al* (1980) also reported that mass activity depends on the viability of the spermatozoa, so high mass activity in the aqueous extract treated groups was an indicator of viability of the spermatozoa. Although, the total sperm output observed in this study agreed with the value reported by Strader *et al.* (1999) as 100  140 millions, however, the result indicated that the extract enhanced the sperm production in the animal. The histological examination revealed that the extract did not cause any detrimental effect on the androgenic process of the sperm cells.

## CONCLUSION

The results of the present study indicated that the aqueous extract of *Moringa oleifera* root extract significant enhanced spermatogenesis and sperm quality. The root of this plant could be used to induce sexual activity and extract from this plant could be further explored for conception use.

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