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Effects of *Zingiber Officinale* on Reproductive Functions in the Male Rat

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ABSTRACT

To investigate the effects of *Zingiber Officinale* on male reproductive functions and study the mechanisms underlying these effects, aqueous extract of *Zingiber Officinale* were administered orally to two groups of male rats at 500mg/kg b.w. and 1000mg/kg b.w. A third group served as control and received the treatment vehicle, distilled water. Treatment lasted for 14 and 28 days before sacrifice. Organ weight, epididymal sperm counts, motility, viability and morphology, seminal fructose, testicular malonhydiyaldehyde, and serum testosterone were determined. The treatment caused a significant increase ($P<0.05$) in the weight of the testis and epididymis. There were dose and duration dependent increases in sperm count and motility ($P<0.05$). There was also a significant increase ($P<0.05$) in serum testosterone level. Malonhydiyaldehyde levels were significantly reduced ($P<0.05$). Our results indicated that extract of *Zingiber Officinale* possesses pro-fertility properties in male rats which might be a product of both its potent antioxidant properties and androgenic activities.

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Key Words: *Zingiber Officinale*, testosterone, androgenic, malonhydiyaldehyde and sperm.

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INTRODUCTION

Zingiber Officinale commonly called ginger belongs to the family Zingiberaceae. The plant is a knotted, thick, beige underground stem (rhizome) that has been used in traditional medicine to aid digestion and treat stomach upset, diarrhea, nausea, and arthritis for centuries. In addition to these medicinal uses, ginger continues to be valued around the world as an important cooking spice and is believed to help the common cold, flu-like symptoms, headaches, and even painful menstrual periods. Today, ginger root is widely used as a digestive aid for mild stomach upset and is commonly recommended by health care professionals to help prevent or treat nausea and vomiting associated with motion sickness, pregnancy, and cancer chemotherapy (Bone et al, 1990; Grontved et al, 1988; Sripramote et al, 2003). Ginger is used as support in inflammatory conditions such as arthritis (Altman et al, 2001), and may even be used in heart disease (Bhandari et al, 1998) or cancer (Katiyar et al, 1996). The important active components of the ginger root are thought to be volatile oils and pungent phenol compounds such as gingerols, shogaols, zingerone, and gingerols (Sekiwa et al, 2000; Zancar et al, 2002). Although the beneficial effect of ginger has been exploited, little research has been conducted on its activity on male reproductive functions except a study that reported that *Z. Officinale* possess androgenic property (Kamtchouing et al, 2002). This work was therefore carried out in view of the paucity of literature on the action of *Z. Officinale* extract on reproductive functions and antioxidant activities in male rats.

MATERIALS AND METHODS

Animals: Adults male rats weighing between 140-160g were maintained in a well ventilated animal house under standard condition of humidity, temperature and a constant 12 hour light:12 hour dark lighting schedule. The animals were housed in clear polypropylene cages lined with wood chip beddings. They were fed with standard pellet diet (Livestock Feeds, Ikeja, Lagos) and water was

made available at all times. The health and reproductive status of the animals were assessed and only healthy animals were selected for the experiment.

Extraction of plant material

The *Z. Officinale* rhizome was purchased from local commercial sources and shade dried at room temperature before being pulverized with an electric grinder. The extracts were then obtained by maceration method with distilled water for 48hours to obtain a final aqueous concentration of 1g/ml.

Experimental Design

Thirty six adult male rats were randomly divided into 3 groups of 12 animals each. Group 1 (control group) was administered with the vehicle (distilled water) while groups 2 and 3 were given an aqueous suspension of *Z. Officinale* at 500mg/kg and 1000mg/kg. Six rats from each experimental group were randomly sacrificed after 14 days of extract administration while the remaining rats were allowed up to 28 days before sacrifice. Treatment was done daily using oral dosing needle and twenty four hours after the last dose, blood was collected and the animals were sacrificed.

Sperm Function Analysis: The rats were sacrificed after the last day of administration and weighed for the essential reproductive organs, such as testis, caudal epididymis, seminal vesicle and prostate glands. The spermatozoa were obtained by making a small incision (1ml) in the caudal epididymis and evaluated for sperm count, motility, viability and morphology. The sperm count was determined with an improved Neubauer haemocytometer and viability was assessed by eosin-nigrosin dye exclusion test. Epididymal sperm motility was assessed by calculating motile spermatozoa per unit area and was expressed as percent motility. Sperm morphology was done using the Wall and Ewas stain and examined under the microscope as described earlier in literatures.

Malonhyaldehyde Estimation

This was measured as an indicator of lipid peroxidation and by extension reactive oxygen

species. Testes homogenate prepared was placed in micro-centrifuge tube and incubated with thiobarbituric acid (TBA). Following the incubation, samples were centrifuged (2 000 rpm, 10 min) and the absorbance of the pink clear supernatant was measured at $\lambda = 532$ nm in duplicate samples. Malonhydaldehyde bis (dimethyl acetal) was used as the external standard.

Fructose Measurement

Seminal fructose was determined according to according to a WHO manual protocol. Briefly, seminal plasma is deproteinized with zinc sulfate and barium hydroxide. The deproteinized seminal plasma is then reacted with acidic resorcinol to form a colored product. The absorbance is read at 410nm.

Testosterone Assay

Blood samples were spun at 2500rpm for 10 minutes in a table top centrifuge. The serum samples obtained were analyzed to determine the concentration of testosterone. The analysis was carried via the tube-based enzyme immunoassay (EIA) method. The protocol used for the hormone was according to the method described for the kit (Immunometrics Limited UK) and meet the WHO standards in research programme for human reproduction.

Statistical Analysis

Data are expressed as mean \pm SEM and analysed using the Student's *t*-test and ANOVA where necessary. $P < 0.05$ was accepted as significant.

RESULTS

Effects of *Z. Officinale* on body and organ weight – Administration of *Z. Officinale* caused no changes in the weight of the seminal vesicle and prostate gland. However, testicular weight was increased significantly ($P < 0.05$) after 14 and 28 days treatment in a dose and duration dependent manner when compared with the controls. In exception to the epididymal weights of other groups, the 1000mg/kg group of the 28 days

treatment showed a significant weight increase ($P < 0.05$) compared with the control (Table 1).

Table 1: Effects of *Z. Officinale* on weight of the testis, epididymis, prostate and seminal vesicle after 14 days and 28 days treatment.

Groups	Testis	Epidid.	Prostate	Seminal Vesicle
14 Days				
Control	1.47 ± 0.06	0.43 ± 0.02	0.27 ± 0.05	0.52 ± 0.04
500mg/kg	1.58 $\pm 0.02^*$	0.43 ± 0.03	0.27 ± 0.02	0.51 ± 0.01
1000mg/kg	1.63 $\pm 0.03^*$	0.44 ± 0.03	0.29 ± 0.03	0.52 ± 0.02
28 Days				
Control	1.73 ± 0.05	0.48 ± 0.02	0.28 ± 0.03	0.55 ± 0.04
500mg/kg	1.85 $\pm 0.06^*$	0.49 ± 0.03	0.30 ± 0.01	0.55 ± 0.03
1000mg/kg	1.88 $\pm 0.03^*$	0.53 $\pm 0.02^*$	0.29 0.03	0.56 ± 0.04

* $P < 0.05$, values expressed as mean \pm sem, $n = 6$, weight unit in grams. Epidyd = epididymis

Table 2: Effects of *Z. Officinale* on sperm count, motility, viability and abnormal morphology after 14 days and 28 days treatment.

Groups	Count (10^6 /ml)	Mot (%)	Viab (%)	Abnormal Morphology
14 Days				
Control	8.10 ± 0.14	85.40 ± 1.07	96.00 ± 1.52	2.18 ± 0.38
500mg/kg	8.93 ± 0.72	89.50 $\pm 2.14^*$	95.20 ± 1.77	2.23 ± 0.24
1000mg/kg	9.08 $\pm 0.17^*$	91.70 $\pm 1.68^*$	95.70 ± 1.45	2.17 ± 0.47
28 Days				
Control	8.17 ± 0.25	84.60 ± 1.31	93.80 ± 1.72	2.63 ± 0.41
500mg/kg	9.26 $\pm 0.28^*$	89.40 $\pm 2.23^*$	95.30 ± 2.11	2.59 ± 0.36
1000mg/kg	9.53 $\pm 0.42^*$	92.50 $\pm 1.84^*$	95.50 ± 1.87	2.67 ± 0.33

* $P < 0.05$, values expressed as mean \pm sem, $n = 6$, abnormal morphology in %. Mot = motility; viab = viability

Effects of *Z. Officinale* on sperm function – Caudal epididymal sperm count and motility were significantly increased ($P < 0.05$) after 14 and 28 days treatment in a dose and duration dependent manner compared with the controls. There were no

differences in other sperm parameters of viability and morphology compared with controls (Table 2).

Effects of *Z. Officinale* on malonhydiyaldehyde estimation – Treatment with *Z. Officinale* significantly decreases ($P<0.05$) the levels of malonhydiyaldehyde in the experimental rats in a dose and duration dependent manner as compared with the control. This reduction was highest for the group 3 (1000mg/kg) in the 28 days and lowest for the group 2 (500mg/kg) 14 days treatment regimen (Fig. 1).

Effects of *Z. Officinale* on fructose levels – *Z. Officinale* caused no significant changes in the fructose level in the experimental rats after 14

days and 28 days administration (Table 3).

Effects of *Z. Officinale* on serum testosterone levels – There were dose and duration dependent increases in the serum testosterone levels in the *Z. Officinale* administered rats compared with the controls (Fig. 2).

Table 3:
Effects of *Z. Officinale* on seminal fructose level after 14 days and 28 days treatment.

Groups	Control	500mg/kg	1000mg/kg
14 Days	4.50±0.84	4.61±0.53	4.56±0.67
28 Days	4.72±0.64	4.60±0.79	4.67±0.53

* $P<0.05$, values expressed as mean±sem, n= 6, fructose level units in $\mu\text{mol/g}$.

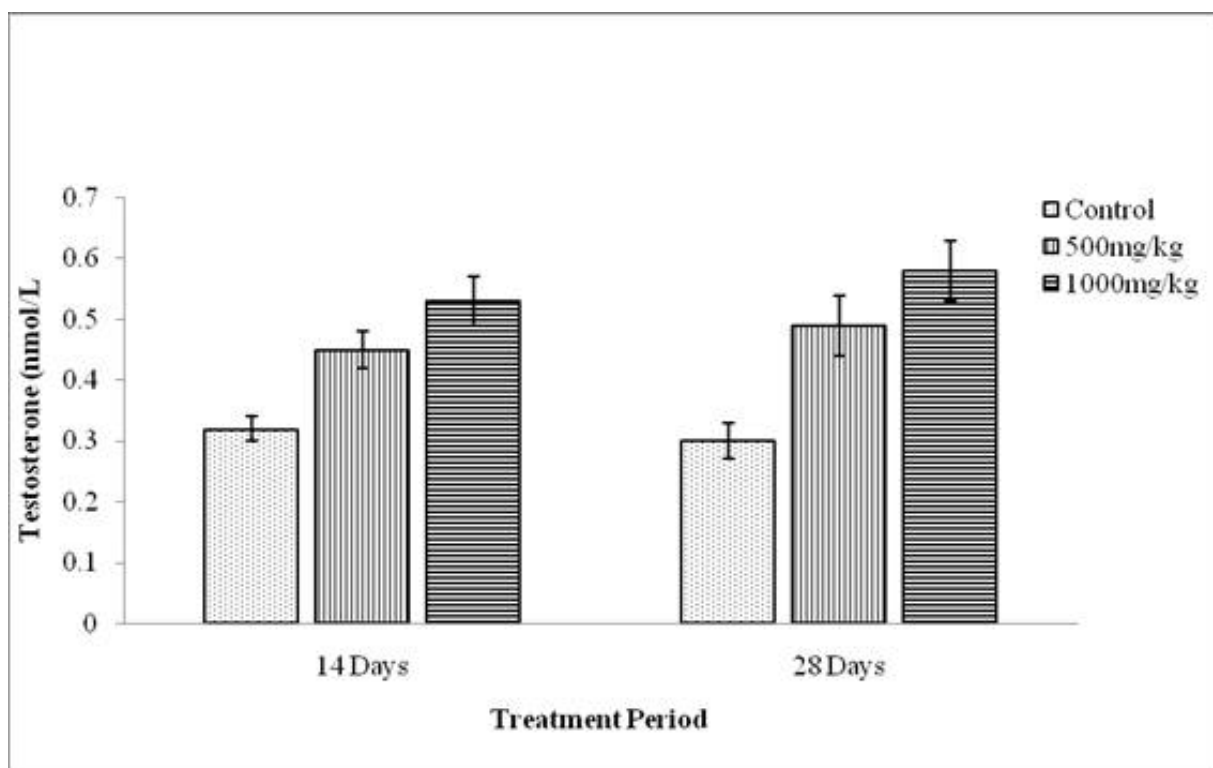


Figure 1:
Effects of *Z. Officinale* on malondialdehyde levels in rats after 14 days and 28 days treatment

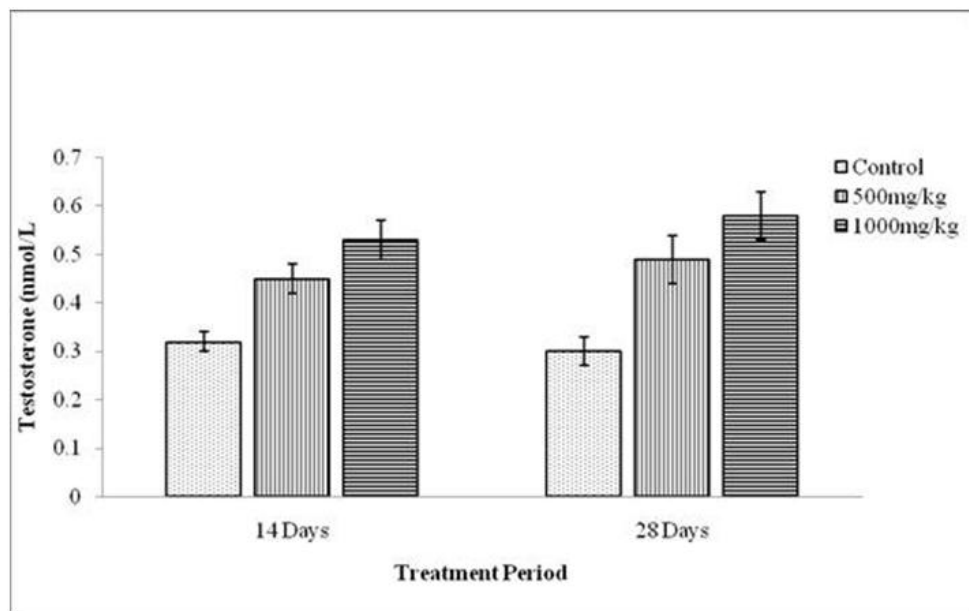


Figure 2: Effects of *Z. Officinale* on serum testosterone levels in rats after 14 days and 28 days treatment.

DISCUSSION

The results of the present study suggested that *Zingiber Officinale* have a beneficial effect on male reproductive functions in rats. These data are confirmed by our observation on the increased sperm counts, motility, testosterone, and decrease malonhydiyaldehyde levels. The significant increase in the absolute weight of the testis and epididymis could therefore be due to increased androgen biosynthesis as evidenced by a significant increase in serum testosterone levels in the experimental rats. Androgens have been shown to be necessary for the development, growth and normal functioning of the testes and male accessory reproductive glands and studies have shown that the level is positively correlated with the weight of testis, epididymis, seminal vesicle and prostate glands (Setty et al, 1997; Prins et al, 1991). This increased reproductive organ weight is equally consistent with the report of Kamtchouing et al who observed an increase in the testicular weight of rats treated with *Z. Officinale* for 8 days with a concomitant increase in testosterone level. Moreover, an effect due to testosterone changes alone should have led to an increase in the weight of all accessory organs; it is therefore plausible

that the increased weight of the testis and epididymis reflects a dual effect of increased testosterone levels and sperm contained in these organs.

The observed increase in the sperm functions in the *Z. Officinale* administered rat could be attributed to favourable and increased spermatogenic activities as results of high testosterone levels. Testosterone is known to be critically involved in the development of sperm cells and derangement results widely in leydig cell dysfunction and testicular steroidogenic disorder (Zhang et al, 2001). It is known that a major function of the epididymis is sperm maturation which leads to the acquisition of fertilizing ability and viability of spermatozoa. Therefore, improvement in the activities of the epididymis could have led to an increase in progressive motility of sperm in the experimental rats. The increased sperm count and motility thereby shows that treatment with *Z. Officinale* improves and enhances the fertilizing capacity of the Semen. These qualities were often used as a measure of sperm production, testicular function and/ or male fertility. Low sperm count and motility

and high percentage abnormal spermatozoa level each have been associated with reduced fertility (Raji et al 2003, Adeeko and Dada 1998).

Another explanation for these improved testicular/sperm function in this study is the reduced level of malondialdehyde, the most abundant individual aldehyde resulting from lipid peroxidation breakdown in biological systems. Malondialdehyde is an indirect indicator of reactive oxygen species, ROS, which has potential toxic effects on sperm quality and function (Sharma and Agarwal, 1996; Sikka et al, 1995). For instance, Agarwal and co-workers (1994) reported that increased formation of ROS is correlated with the reduction of sperm motility. In the light of the above, *Z. Officinale* seems to confer a protective antioxidant defence capacity on the treated rats as evidenced by a significant reduction in the level of malondialdehyde.

In conclusion, the extract of *Zingiber Officinale* might be said to have pro-fertility properties in male rats which might be a consequence of both its potent antioxidant properties and androgenic activities.

REFERENCES

- Adeeko, A.O. and Dada, O.A. (1998). Chloroquine reduces the fertilizing capacity of epididymal sperm in rats. *Afri. J. Med. Med. Sci.* 27, 63 -68.
- Agarwal A, Ikemoto I, Loughlin KR. (1994). Relationship of sperm parameters to levels of reactive oxygen species in semen specimens. *J Urol.* 152: 107-10.
- Altman RD, Marcussen KC. (2001). Effects of a ginger extract on knee pain in patients with osteoarthritis. *Arthritis Rheum.* 44(11):2531-2538.
- Bhandari U, Sharma JN, Zafar R. (1998). The protective action of ethanolic ginger (*Zingiber Officinale*) extract in cholesterol fed rabbits. *J. Ethnopharmacol.* 61:167-171.
- Bone ME, Wilkinson DJ, Young JR, McNeil J, Charlton S. (1990). Ginger root, a new antiemetic. The effect of ginger root on postoperative nausea and vomiting after major gynaecological surgery. *Anaesthesia.* 45(8): 669-71.
- Grontved A, Brask T, Kambskard J, Hentzer E. (1988). Ginger root against seasickness: a controlled trial on the open sea. *Acta Otolaryngol.* 105: 45-49.
- Kamtchouing P, Fandio GYM, Dimo T, Jatsa HB. (2002). Evaluation of androgenic activity of *Zingiber officinale* and *pentadiplandra brazzeana* in male rats. *Asian J Androl.* 4:299-301.
- Katiyar SK, Agarwal R, Mukhtar H. (1996). Inhibition of tumor promotion in SENCAR mouse skin by ethanolextract of *Zingiber officinale* rhizome. *Cancer Res.* 56:1023-1030.
- Meistrich ML, Samuels RC. (1985). Reduction in sperm levels after testicular irradiation of the mouse: a comparison with man. *Radiat Res.* 102: 138-47.
- Prins SG, Birch L, Greene GL. (1991). Androgen receptor localization in different cell types of the adult rat prostate. *Endocrinology.* 129: 3187-99.
- Raji Y, Udoh US, Mewoyaka OO, Ononye FC, Bolarinwa AF. (2003) Implication of reproductive endocrine malfunction in male antifertility efficacy of *Azadirachta indica* extract in rats. *Afri. J. Med.Med.Sci* 32:159-165.
- Sekiwa Y, Kubota K, Kobayashi A. (2000). Isolation of novel glucosides related to gingerdiol from ginger and their antioxidative activities. *J.Agric.Food Chem.* 48:373-377.
- Setty BS, Riar SS, Kar AB. (1977). Androgenic control of epididymal function in rhesus monkey and rabbit. *Fert Steril.* 22:674-81.
- Sharma RK, Agarwal A. (1996). Role of reactive oxygen species in male infertility. *Urology.* 48: 835-50.
- Sikka SC, Rajasekaran M, Hellstrom WJG. (1995). Role of oxidative stress and antioxidants in male infertility. *J Androl.* 16: 464-8.
- Sripramote M, Lekhyananda N. (2003). A randomized comparison of ginger and vitamin B6 in the treatment of nausea and vomiting of pregnancy. *J Med Assoc Thai* 2003; 86(9): 846-853.
- Zancan KC, Marques MO, Petenate AJ, Meireles MA. (2002). Extraction of ginger (*Zingiber officinale* Roscoe) oleoresin with CO₂ and co-solvents: a study of the antioxidant action of the extracts. *J.Supercrit.Flu.* 24:57-76.
- Zhang X, Yamamoto N, Soramoto S, Takenaka I. (2001). Cisplatin-induced germ cell apoptosis in mouse testes. *Arch Androl.* 46:43-49