HISTOLOGICAL ASSESSMENT OF EFFECTS OF ALCOHOLIC EXTRACT OF BENISEED ON REPRODUCTIVE ORGANS OF MALE SPRAGUE-DAWLEY RATS

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To the Editor:

It has been established that antioxidants such as Vitamin C and E are active enhancers of gonadal functions1-3. This is due to the fact that they scavenge free radicals (oxidants) in tissues and blood vessels neutralizing their damaging effects. Antioxidants increase gonadal functions by enhancing the hormonal secretions necessary for the proliferation of spermatogenic cells3.

Beniseed oil has been implicated to regulate blood pressure to normal in adult hypertensive patients. Upon withdrawal, the blood pressures were elevated4. This might be due to the fact that it contains antioxidants such as sesamin (0.34 to 1.13%), sesamolin (0.13 to 0.58%) and sesamol, Sudesh & Vickii reasoned that antioxidants may achieve antihypertensive effects through one of three ways: reducing aldehyde conjugate AGE formation and oxidative stress, normalizing calcium channels and peripheral vascular resistance, or by improving insulin resistance and endothelial function. In the presence of oxidants, constricttion of the blood vessels and subsequent hypertension may result. In the same study, antioxidants (Vitamin C & E, b-carotene and reduced glutathione) were increased. These types of effects are pro-fertility in nature. We earlier argued that Benisede enhances fertility by production of substances necessary for the proper function of the reproductive organs.

Among all reactive oxygen species, hydrogen peroxide is the most toxic to testicular functions. Treatment with superoxide dismutase and catalase reduced the degree of testicular damage in experimental acute torsion by scavenging the superoxide radicals and converting them into hydrogen peroxide and water and the catalase then converts the hydrogen peroxide produced into water and oxygen10. These reactive oxygen species are regularly formed during the process of normal respiration. However, the production is kept at physiologically low levels by intracellular free radical scavengers11.

The seed contain 7-11% palmitic acid, 2-6% stearic acid, 39-56% Linoleic acid, 32-54% Oleic and other fatty acids in lesser than 1%. The plant seeds also contain two lignans which are oil-soluble: sesamin and sesamolin as well as lignan glucosides such as sesaminol di-glucoside and tri-glucosides, and pinoresinol mono-, di- and triglucosides12.

The present study aim at evaluating the effects of alcoholic extract of Beniseed seed on the architecture of selected reproductive organs.

Twenty (20) adult male rats of Sprague-Dawley strain obtained from the faculty of Basic medical sciences' animal house, Ladokun feeds, Ibadan and water made available ad libitum. They were allowed to acclimatize for two weeks before the experiments.

Benisede was obtained at a local market in Ogbomoso, Oyo State, Nigeria authenticated by the Botany Department, University Lagos. The seeds were peeled and sundried. They were finely powdered. The powdered form was allowed to stand in 70% alcohol for 24 hours and then filtered. The filtrate was concentrated to a semi-solid form using rotary evaporator. The Lethal Dose (LD50) estimation of benisede was done at Pharmacognosy Department, College of Medicine University of Lagos and was found to be 5000mg/kg. A stock solution of the extract was prepared at a concentration of 300mg/ml, the solution was kept in the refrigerator to prevent fermentation. A known volume was dispensed daily for administration using an oropharyngeal canula.

The rats were divided into four (4) groups of 5 rats each. The groups were designated as control (C) and Treatments (T1, T2, T3).

Each of the rats in the treatment groups (T1, T2, T3) received a single dosage of 3000mg/kg body weight of extract solution, orally, daily for 8 weeks. T3 also received 2.5mg of Vitamin C (Tuyil Pharmaceutical Company Ilorin, Kwara State) in addition to benisede extract. The animals were weighed once every week. After the initial period of 8 weeks, rats in T2 were withdrawn from benisede extract for a period of 4 weeks. The animals were sacrificed by cervical decapitation as follows: C, T1 and T3 at the end of 8th week and T2 at the end of 12th week.
The testes, epididymides and seminal vesicles were removed by midline laparotomy. The epididymides were dissected free of the testes while the testes, epididymides and seminal vesicles were weighed in electrical weighing balance before fixing in 10% formol saline. The organs were histologically processed according to a routine method used and described by Oluyemi et al.\textsuperscript{8}.

The results are presented in Figures 1 to 3

![Figure 1](http://biomed.uninet.edu/2009/n3/ashamu.html)
FIGURE 2: Photomicrograph of (a) sections through the epididymis of the control group show round tubes in cross sections lined by pseudostratified columnar epithelium with stereocilia projection from their surface. Single layered muscle cells surround these. Within the lumina are dense long eosinophilic processes of sperm tail.

(b) Sections through the epididymis of the T1 show round tubes in cross sections lined by stratified epithelium with abundant stereocilia projection from their surface. These are surrounded by a thick muscle layer. Within the lumina are dense long eosinophilic processes of sperm tail.

(c) Sections through the epididymis of the T2 show round tubes in cross sections lined by pseudostratified columnar epithelium with stereocilia projection from their surface. These are surrounded by a thick muscle layer. Within the lumina are abundant long eosinophilic processes of sperm tail seen clumping together.

(d) Sections through the epididymis of the T3 show round tubes in cross sections lined by pseudostratified columnar epithelium with stereocilia projection from their surface. These are surrounded by single layered muscle cells. The lumina are virtually empty of sperm cells.

FIGURE 3: Photomicrograph of (a) sections through the seminal vesicle of the control show glands arranged in a honeycomb fashion. They are lined by tall columnar epithelium. They secrete abundant eosinophilic material into a large lumen.

(b) Sections through the seminal vesicle of the T1 show glands glandular hyperplasia with more pronounced honeycomb arrangement. They are lined by tall columnar epithelium. The eosinophilic secretions within the lumen are reduced.

(c) Sections through the seminal vesicle of the T2 show glands arranged in a honeycomb fashion. They are lined by tall columnar epithelium. There is increased intraglandular secretion in addition to eosinophilic material within large lumen.

(d) Sections through the seminal vesicle of the T3 show glands glandular hyperplasia with labyrinthine projections. They are lined by stratified epithelium. There is increased secretion of eosinophilic material into a large lumen.

The photomicrographs of the testis of the experimental animals have proved that alcoholic extract of Beniseed seed has a profertility effects enhancing the production of spermatozoa and buffering the integrity of cells of spermatogenic series as well of the testicular cyto-architecture. These findings are in agreement with the findings of Shittu et al.13. In the presence of steroid-containing substances, spermatogenesis tends to become enhanced leading to the maturation and proliferation of spermatogenic cells. As can be seen in Fig. 1c and 1d, there were massive accumulations of spermatozoa in the lumen of the seminiferous tubules. This is an indication that beniseed extract is a potent fertility-enhancing plant. Earlier, we reported a dose-dependent increase in sperm counts in wistar rats6. There were observed increase in the thickness of seminiferous epithelia: a strong fact pointing to the proliferative effects of the plant. Beniseed oil induces developmental abnormalities in the male reproductive system of juvenile rats but these were restored at puberty14 as a result of disruption in the steroidogenesis, which is kept at base during childhood but shoots up at puberty.

The integrity of the epididymal cytoarchitecture were preserved in our experiment (Fig. 2a-2d) suggesting that Beniseed does not exert any adverse effects on the epididymis and the developing luminal spermatozoa as the latter further develop to acquire motility. This agrees with the work of Shittu et al.13.

In experiment 3, the vitamin C was administered as adjuvant to increase the anti-oxidative function of Beniseed extract. In this class of experiment it was discovered that the presence of vitamin C does not alter significantly the antioxidant properties. In the overrall, the results show a progressive increase in the integrity of cytoarchitectures of the reproductive organs of the experimental animals.

These results thus confirm the traditional consumption of Beniseed for the purpose of fertility treatment especially in males.

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Received October 30, 2009.
Published, December 18, 2009