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Protective effects of Vitex doniana (Black plum) against ischemic testes torsion injury: Histological and morphometric features

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ABSTRACT

We evaluated the effects of Vitex doniana leaves on testes histology and morphometry in Wistar rats following testicular torsion. Thirty male Wistar rats (120–160 g) were randomly assigned into six groups (n=5). Surgical procedures were performed under ketamine anaesthesia (5 mg/kg i.m). Sham surgery was performed on group A rats and given distilled water for 7 days. Animals in other groups had their left testes twisted at 720° for one hour, sutured, and treated for 7 days. Group B rats were left untreated, groups C, D and E received 50 mg/kg, 100 mg/kg and 200 mg/kg of extract respectively while group F received 100 mg/kg of vitamin C. Animals were sacrificed using ketamine (100 mg/kg), testes excised, fixed in Bouin's fluid and processed for histology and histomorphometry. Data were analyzed using Oneway ANOVA and Student Newman- Keuls post- hoc test for multiple comparisons. Seminiferous tubule diameter reduced significantly in group B when compared with control group while the luminal size increased significantly in group B when compared with control group and the treated groups. Group B also showed significant reduction of germinal epithelium when compared with groups A, E and F. Histology and histochemical results revealed atrophic seminiferous tubules, widened interstitial spaces and accumulation of glycogen in the basement membrane in the testes of untreated rats. The study concluded that aqueous extract of Vitex doniana leaves had ameliorative effect on testicular morphology and histology in the torsioned rat model.

ished fertility [3–6].

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1. Introduction

Testicular torsion is described as the twisting of the spermatic cord, resulting in acute pain and ischemia [1], particularly of the testis. The twisting of the spermatic cord results in a compromised blood flow to the testis, the extent of which depends on the degree of arterial compression [2] and the rotation of the testis. The cause of testicular torsion is unknown, but certain anatomical conditions such as clapper bell deformity, horizontal lie and maldescent of the testis are traditionally referred to as the risk factors.

Testicular torsion, or the torsion of the spermatic cord, is a surgical emergency in humans that causes testicular injury and subfertility. It is generally accepted that unilateral testicular torsion accepted. Vitex doniana, is a tree of 10-15 m (32-49 feet) in height, commonly called Fon, or Ewe oyi by traditional healers and plants sellers in Bénin. The leaves consist of 5 sepals. Flowers are white and sometimes stained with purple. The fruit are black, edible, sweet and mealy. It is frequently eaten as a snack and sold in local markets. The fruits are approximately 3 cm long and contain one to four

seeds. The fruits are collected from April to June. Fresh fruits cannot

causes contralateral testicular deterioration and results in dimin-

Nagler and de Vere White [7] demonstrated that unilateral tor-

sion caused contralateral histological damage. Consentino et al. [8]

also bred rats after unilateral torsion and demonstrated diminished

fertility. The exact mechanism of contralateral testicular damage

still remains unclear; immunological responses, subclinical attacks

of contralateral testicular torsion, underlying congenital defects,

release of acrosomal enzymes, and alterations in the blood flow

have all been proposed [6,9–11], but none has been universally







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be stored for long time periods [12]. In Nigeria, from information available from the indigenous traditional healers, a decoction of the chopped stem bark part of *V. doniana* is prepared and taken orally for treatment of gastroenteritis. It is administered for ailments including diarrhea and dysentery. It is also taken to improve fertility and the juice may be squeezed into the eyes to treat eye troubles. It is also used in the treatment of liver disease. *V. doniana* is used by traditional healers alone or in a combination with stem bark of *Adansonia digitata* to treat diarrhea, leprosy and dysentery [13]. The leaves are used as antiseptic and anti- diabetic [14]. The aerial parts are used in Mali as diuretic, tonifiant, aphrodisiac and bactericide [15,16]. They can be used as a febrifuge, anti-dysenteric and in the treatment of smallpox and measles [17,18]. The bark of the trunk is used in the treatment of malaria and also used to bath babies to encourage a smooth skin [13,19].

2. Methods

2.1. Extraction of Vitex doniana leaves

Vitex doniana leaves were collected from the lawn behind Moremi Hall, Obafemi Awolowo University, lle-Ife and were taken to a taxonomist at the Department of Botany, Obafemi Awolowo University, lle-Ife for authentication after which a voucher specimen was deposited at Ife Herbarium for reference with a reference number IFE-17377433. The leaves were cleaned and air-dried at room temperature. The dried leaves were pulverized using an electric blender. The extraction was done using percolation method which involved continuous washing of the pulverized leaves with water at 80 °C and 30 extraction cycles for 3 h using Soxhlet extractor. The extract was concentrated *in vacuo* at 65 °C using a vacuum rotary evaporator and freeze-dried in a lyophilizer. The extract was stored in a desiccator until used.

2.2. Phytochemical constituents of aqueous extract of Vitex doniana

The phytochemical screening of aqueous extract of *Vitex doniana* according to the methods of Harbone [20] and Trease and Evans [21] has revealed the following constituents: Alkaloids, Flavoloids, saponins, tannins, anthraquinones and terpenoids.

Some of the specific, isolated components of these phytochemicals include: β -phellandrene, phytol, β -caryophyllene, α -caryophyllene, caryophyllene oxide, α -pinene and bicyclogermacrene [22]. The chemical structures of these constituents and their percentage composition are listed in Table 1 [23–26].

2.2.1. Vitamins composition of Vitex doniana

Vitamin analysis of *Vitex doniana* young leaf confirmed the presence of vitamin A, B1, B2, B3, C, D, E and K. The result shows that vitamin C has the highest of 20.80 mg/100 g and vitamin K has the lowest value of 0.017 μ g/100 mg. The recommended daily allowance (RDA) vitamins for human showed that vitamin C is most needed among all the listed vitamins, with recommended quantity of 75 mg per day. Vitamin K is the least needed among all the vitamins listed with recommended quantity of 80 μ g per day [27]

2.2.2. Minerals composition of Vitex doniana

The analysis of macro mineral contents of the *Vitex doniana* young leaf revealed that Calcium (Ca) content is very high (0.73%) while the value observed for Magnesium (Mg), Potassium (K) and Sodium (Na) are 0.41%, 0.26% and 1.87 ppm respectively and similar to content in Amaranthus hybridus [28]. The analysis of micro mineral contents of the *Vitex doniana* young leaf revealed that Iron (Fe) content is very high (24.156pm) and fall within the range earlier published for edible vegetables [29] while the value observed

Table 1

Phytochemical constituents of Vitex doniana.



for Manganese (Mn), Zinc (Zn) and Copper (Cu) are 18.36 ppm, 9.487 ppm and 6.224 ppm respectively is moderate for Vegetables [28]

2.3. Animal care and management

Thirty young male Wistar rats weighing between 120–160 g (age 10 weeks) were obtained from the Animal House of the College of Health Sciences Obafemi Awolowo University Ile-Ife, and used for this research. The rats were randomly assigned into six groups of five rats per group (Groups A, B, C, D, E and F). Animals were housed in clean plastic cages under natural light and dark cycles and at room temperature. Animals in all groups were fed on normal laboratory chow, and had access to water *ad libitum*.

2.4. Surgical procedure and experimental protocol

Animals in group A had their testes exposed for one hour without twisting the spermatic cords, sutured back (sham surgery) and then were given distilled water for 7 days. Group B had their spermatic cords twisted at 720°, left for one hour, sutured back and then received distilled water for 7 days. Group C, D and E also had their spermatic cords twisted at 720°, left for one hour, sutured back and then were orally administered with *V. doniana* extract (50 mg/kg, 100 mg/kg and 200 mg/kg per body weight) respectively for 7 days. Group F had their spermatic cords twisted at 720°, left for one hour, sutured back and then were orally administered with vitamin C (100 mg/kg per day) for 7 days. Vitamin C was given to group F to serve as a standard drug so as to compare the antioxidant effect of *V. doniana* with it. All surgical procedures were performed Download English Version:

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