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ORIGINAL ARTICLE

Effects of Tahitian Noni dietary supplement on caffeine-induced testicular histo-pathological alterations in adult Sprague-Dawley rats

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Abstract Objective: To investigate the possible ameliorating effect of Tahitian Noni dietary supplement on caffeine-induced testicular histopathological alterations in Sprague-Dawley rats.

Design: This is an experimental animal study.

Methods: Thirty adult male Sprague-Dawley (SD) rats, weighing between 105 and 200 g were acclimatized and grouped into six of five rats per group. Group 1 was the control. Group 2 received 200 mg/kg of caffeine for 8 weeks, Group 3 received 200 mg/kg of caffeine for 4 weeks and 5 ml/kg of Noni for another 4 weeks, Group 4 received both 200 mg/kg of caffeine and 5 ml/kg of Noni for 8 weeks, Group 5 received 5 ml/kg of Noni for 8 weeks, Group 6 received 5 ml/kg of Noni for 4 weeks and 200 mg/kg of caffeine for another 4 weeks.

Results: Tahitian Noni caused a statistically significant increase in the mean body weight of the SD rats, opposed to the groups treated with caffeine. There was also a statistically significant increase in the testicular weight, sperm count and motility in the SD rats treated with Noni compared to those treated with caffeine. Caffeine negatively affected the histo-architecture of the seminiferous tubules

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with massive loss of spermatogenic cells while the groups exposed to Noni tended towards normal when compared with the control.

Conclusions: Administration of Tahitian Noni dietary supplement ameliorates the testicular toxicities caused by a high dose of caffeine.

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1. Introduction

Caffeine is the world's most widely consumed psychoactive substance, but unlike most other psychoactive substances, it is legal and unregulated in nearly all jurisdictions (1). An estimated 80% of the world's population consumes a caffeine-containing substance daily (2).

Caffeine is considered a psychoactive substance since it stimulates the central nervous system and alters mood and behavior. Heroin, cocaine, marijuana, nicotine and alcohol are also examples of psychoactive drugs. Physiological effects may be seen in adults after as little as one cup of coffee or two cans of cola (3).

The use and abuse of caffeine are a major public "habit" and may be as important a factor as heredity and environment in the etiology of physiological and psychological disorders (4).

Infertility affects more than 80 million people around the globe. It is a ubiquitous phenomenon that transcends race and nationality (5). Male factor and female factor infertility each accounts for about 40% of cases of infertility, the remaining 20% is as a combination of male and female (6).

Tahitian Noni or Indian mulberry originated in Tropical Asia and Polynesia (7,8). It is one of the most common plants used in herbal remedies. The fruit juice is in high demand in alternative medicine for different kinds of illnesses, such as arthritis, diabetes, high blood pressure, muscle aches, menstrual difficulties, headaches, heart disease, Human Immuno Virus/Acquired Immune Deficiency Syndrome, cancer, gastric ulcers, sprains, mental depression, senility, poor digestion, atherosclerosis, blood vessel problems and drug addiction (7).

There have been assumptions on the fertility-enhancing effect of *Morinda citrifolia* but no work has been done on its effect on the possible testicular toxicities that can be caused by a lot of substances we ingest, of which caffeine is one of the foremost.

2. Materials and methods

2.1. Collection of materials

The regular bottled commercial form of Tahitian Noni dietary supplement produced by Morinda Inc., United States of America was obtained from a registered Tahitian Noni distributor. The producing company's bottle cap was observed intact before commencement of use. A 200 g tin of Nescafe, a brand of coffee made from Robusta beans by Nestle Nigeria Plc was used for this experiment.

2.2. Animals

Thirty adult male Sprague-Dawley rats, 16–20 weeks old and weighing between 105 and 200 g were obtained from the

Department of Biochemistry, College of medicine, University of Lagos. The rats were housed in well-ventilated metal cages under standard conditions of temperature ($25 \pm 5^\circ\text{C}$) in the Department of Anatomy, College of Medicine of University of Lagos. Animals were exposed to a photo period of 12 h light, alternating with 12 h darkness. They were allowed access to standard laboratory food and water *ad libitum* throughout the experiment. The animals were kept for at least 2 weeks to acclimatize to the laboratory conditions before experimentation.

2.3. Experimental protocol

The 30 male rats were divided randomly into six groups of five rats each. The experimental groups received daily oral doses of the drugs as follows: Group 1, the control received daily oral dose of distilled water. Group 2 received 200 mg/kg of caffeine for 8 weeks, Group 3 received 200 mg/kg of caffeine for 4 weeks and 5 ml/kg of Noni for another 4 weeks, Group 4 received both 200 mg/kg of caffeine and 5 ml/kg of Noni for 8 weeks, Group 5 received 5 ml/kg of Noni for 8 weeks, Group 6 received 5 ml/kg of Noni for 4 weeks and 200 mg/kg of caffeine for another 4 weeks. Body weight was recorded weekly for every group. All procedures involving animals were performed in accordance with the guidelines guiding the use and care of laboratory animals and approved by the Departmental Committee on the use and care of animals.

2.4. Sample collection

Animals were anaesthetized by intra-peritoneal injection of ketamine (Rotex Medica, Trittau, Germany) (50 mg/kg) (9) at the end of the experiment.

2.5. Tissue processing for histological work

The organs were processed for histological work as follows: one testis from each animal was fixed in 10% formol saline. The fixed tissues were transferred to a graded series of ethanol and then cleared in xylene. Once cleared, the tissues were infiltrated in molten paraffin wax in the oven at 58°C . Serial sections of $5\ \mu\text{m}$ thickness were obtained from a solid block of tissue, cleared, fixed in clean slides, stained with Haematoxylin and Eosin stains and examined with the light microscope (Figs. 1–6).

2.6. Testicular weight (morphometry)

The testicular weight was estimated using electronic balance.

2.7. Sperm parameters

The cauda epididymis was dissected out; several incisions of about 1 mm were made and were suspended in 1 ml of Ham-

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