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EVALUATION OF RODENTICIDAL POTENTIAL OF YELLOW OLEANDER (*Thevetia neriifolia* Tuss.) SEED POWDER IN WILD FEMALE AFRICAN GIANT RAT (*Cricetomys gambianus* W.)

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ABSTRACT

Cricetomys gambianus is a wild rodent species attacking crops. Conventional rodent control is via the use of synthetic rodenticides with health and environmental concerns. *Thevetia neriifolia* seed reportedly has rodenticidal potential in the laboratory rodents. However, rodenticidal activity of *T. neriifolia* seed has not been evaluated in wild rodent species. Therefore, this study attempted to evaluate the rodenticidal potential of yellow oleander seed powder in wild *C. gambianus*. Twenty wild-caught female *C. gambianus* were acclimatized for 4 weeks in Animal house and randomly allocated into 4 groups including control. The *T. neriifolia* seed was dried under the shade, blended, and mixed with a commercial feed with 4 different concentrations of the seed powder (0%, 20%, 40%, and 80%) and then turned to pellet. The acclimatized animals were fed with appropriate concentrations of the formulated feed for 8 weeks. Body-Weight and Feed-Intake of the animals were taken. Their behavior before and after introduction of the feed was also recorded. At termination, tissue samples (Kidney, Liver, and Heart) were collected for histopathology analysis. Data were analyzed in SPSS using ANOVA and means separated using DMRT at $\alpha_{0.05}$. The results showed no significant effect on the animals' mean Body-Weight and Feed-Intake ($p>0.05$). No mortality and behavioral changes were observed in the animals at all the concentrations. No visible lesions in the kidney, liver, and heart across the treatment groups compared to control. In conclusion, *T. neriifolia* seed powder did not have rodenticidal effect on the wild female *C. gambianus*. Therefore, it could not be used as a rodenticide in the management of wild rodent species population.

Keywords: African Giant Rat, Economic Damage, Yellow Oleander, Rodenticide.

INTRODUCTION

Vertebrate species such as rodent species play ecological roles in the ecosystem which include but not limited to aiding soil mixing and aeration, serving as dispersal agents, enhancing organic matter composition, and serving as prey for predatory vertebrates (Witmer, 2018; Badmus *et al.*, 2024). Rodent species that have been implicated as pests reportedly accounted for about 5 -10% of all the species (Kingdon *et al.*, 2013). The rodent pest species are recognized as the most destructive species in agriculture both in the field and in the store (Akande, 1986; Badmus, 2021; Badmus *et al.*, 2024). They nibble food and contaminate it by their droppings thereby reducing their market value. They cause fire outbreaks by gnawing of electrical cables while some are carriers of zoonotic diseases (Kingdon *et al.*, 2013; Buckle and Smith, 2015).

All over the world, rodents cause about 30% annual crop damage (Singleton, 2001). They are known to be successful in their damaging activities because of their high prolific rate of reproduction and well developed sense of smell, hearing, and taste. One of such species of rodent pests is African giant rats (*Cricetomys gambianus*). The African giant rat is basically fossorial in nature but it is highly adaptable to different lifestyles such as terrestrial and arboreal

Across the world, especially in the developing countries, use of synthetic rodenticides has been the conventional method of rodent control (Badmus and Ala, 2020; Badmus, 2021; Badmus *et al.*, 2024). There are different types of synthetic rodenticides for the indoor and outdoor use in the store and the field respectively, and they are categorized as non-anticoagulant (acute) rodenticides e.g. zinc phosphide; and anticoagulant (chronic) rodenticides e.g. brodifacoum, bromadiolone, difenacoum, difethialone *etc* (Endepols *et al.*, 2015). All synthetic rodenticides have been reported to have one form of attendant shortcoming or the other such as the problem of resistance, bait shyness, secondary poisoning, and effect on non-target species (Buckle and Smith, 2015). So, there is need

for alternative environment-friendly rodenticides that are sustainable and more economical. To this end, efforts have been made continually in the screening of botanical products for rodenticidal properties in laboratory rodent. For example, toxicity of *Thevetia neriifolia* seed, bark and leaves were assessed by Fayinminnu *et al.* (2022) as a rodenticide in Wistar rats. It was observed by Fayinminnu *et al.* (2017) that phytochemicals were present in all the parts of the plant (*Thevetia neriifolia*). But this active phytochemicals were mostly abundant in leaf, stem and bark, and that the leaf contain the greatest toxicity and also the bark and this make them possess very effective rodenticidal effect. The stem bark of *T. neriifolia* was reported have toxic effects on rat.

Olayioye *et al.* (2014) also investigated effect of powdered castor seed on the internal organs of Nile rats. Badmus and Alarape (2022) reported a formulated plant-based diet as a promising alternative to conventional rodent control as it promises to be relatively humane and economical. The rodenticidal effectiveness of yellow oleander seed has not been evaluated in wild-caught small mammal species such as African Giant Rat. Hence, the present study attempts to evaluate efficacy of seed powder of *Thevetia neriifolia* in female African giant rat (*Cricetomys gambianus*) caught from the wild.

MATERIALS AND METHODS

Study Location

The present study was conducted in Ecology Laboratory, Department of Crop Protection and Environmental Biology, Faculty of Agriculture. University of Ibadan, Ibadan, Nigeria.

Experimental Procedures

Collection of ethical approval

Ethical approval (number UI/19/0136) was obtained from Animal Care and Use Research Ethics Committee (ACUREC), Department of Veterinary Pathology, Faculty of Veterinary Medicine, University of Ibadan, Ibadan, Oyo State.

Collection and accclimatisation of experimental animals

A total of 20 sexually mature female *C. gambianus* were captured within the University premises and housed in well sanitized metal cages in central animal house of the College of Medicine, University of Ibadan. The animals were randomly allocated into four groups with five animals per group. Feeders and drinkers were fixed in the cages for easy access of the animals to food and water. The animals were acclimatized and stabilized for 4 weeks with 12 hours light :12 hours dark cycle and relative humidity of 80-90%. Each rat was placed individually in separate closet of the cage. Feed and water were supplied to the animals *ad libitum*.

Collection and identification of Yellow Oleander seeds

The seeds of yellow oleander (Plate 1) were obtained about 7:00a.m. within the University premises and identified at the Department of Botany Herbarium with voucher number UIH – 23176. The seeds were dried under open shade for 10 days and then milled into powder.

Formulation and administration of experimental feed

Four concentrations of experimental feed were formulated by mixing the commercial rat feed with yellow oleander seed powder (w/w) as stated below:

100% commercial rat feed + 0% *T. neriifolia* seed powder

80% commercial rat feed + 20% *T. neriifolia* seed powder

60% commercial rat feed + 40% *T. neriifolia* seed powder

20% commercial rat feed + 80% *T. neriifolia* seed powder

Each experimental feed formulation was converted into feed pellets (Plate 2) and labelled accordingly.

Experimental feed (40 g) was provided appropriately for each animal every morning, between 7 and 8 a.m. and water provided *ad libitum*. They were fed for the period of 8 weeks.

Plate 1: Yellow Oleander (*Thevetia neriifolia*): (A) The plant (B) The fruits (C) The dried seeds

Plate 2: Pelletised experimental feeds, Commercial rat feed with *T. neriifolia* seed powder, at (A) 100: 0 (B) 80:20 (C) 60:40 (D) 20:80

Data Collection

Feed intake, body weight and behavioural changes

During the period of experimental feed administration, body weight of each rat was taken weekly for four weeks using digital weighing scale. This was done to assess gain or loss in weight. Daily percent feed intake of each rat was determined during the experimental period using the formula below:

$$Daily\ feed\ intake\ (\%) = \frac{weight\ of\ the\ feed\ offered\ (g) - weight\ of\ the\ leftover\ feed\ (g)}{weight\ of\ the\ feed\ offered\ (g)} \times 100$$

Fayinminnu *et al.* (2017)

Observations on treatment toxicity were made on behavioral patterns of the rats during feeding. These include loss of appetite, and loss or gain of weight.

Tissue/organ harvest for histological analysis

The animals were sacrificed using cervical dislocation to collect tissue samples from kidney, liver, and heart. They were dissected and their tissues/organs removed. They were then preserved in 10% buffered formalin and taken to the Department of Veterinary Pathology, Faculty of Veterinary Medicine, University of Ibadan, for histological analysis.

Photography and illustration

Necessary photography was done during gross morphological and histological investigations for better illustration of the result. The gross anatomical pictures were taken directly from the organs by using digital camera and the histological pictures were taken from light microscope (x 40 objective) (Bhakta *et al.*, 2019). This was carried out after the animals have been sacrificed and the pictures of the organs were taken from two animals per group.

Data Analysis

The data collected were analysed in Statistical Package for Social Science (SPSS) software. Mean feed intake and body weight were analysed using ANOVA. Means were separated using Tukey HSD test.

RESULTS

Effect of *Thevetia neriifolia* seed powder on feed intake and body weight and of female African giant rat

The results of the present study show variation in daily feed intake (%) of female African giant rat per group. However, there is no significant

difference in the average feed intake across groups, including control (F= 1.97, df=19, p > 0.05)

The result of the present study shows the variation in the weekly average weight (kg) of the female African giant rat per group. At week one of the experimental period, the group that received the 80:20 concentration of the experimental feed had the highest mean weight (1.65±1.69) values with the same alphabet are not significantly different while the group that received the mid concentration of experimental feed had the lowest mean weight and in comparison with control at week one (Table 4.1). However, there is no significant difference in the average weight across the treatment groups including control (F = 1.920, df =19, p > 0.05).

At week three, the group that received the lowest (80:20) concentration of the experimental feed recorded the highest mean weight (1.86±0.39) kg while the group that received the mid (60:40) and lowest (80:20) concentrations of experimental feed recorded the lowest mean weights (kg) - (1.62±0.81) and (1.62±0.08) – respectively (Table 4.1). Meanwhile, there was no statistical significant difference in the average weight across the treatment groups including control (F = 4.409, df = 19, p > 0.05).

At week four, group 2 fed with the mid concentration of experimental feed record the highest mean weight (1.85±0.04) kg though it was statistically similar to group 1; while the group that received the highest concentration of the experimental feed had the lowest mean weight (1.56±0.07) kg (Table 4.1). There was statistical significant difference across the experimental groups and in comparison with the control group (F = 1.331, df = 19, p > 0.05).

At week five, the group that received the highest concentration of the experimental feed had the lowest mean weight (1.57±0.07) kg while the group that received the mid and lowest concentration of experimental feed had the same and highest mean weight (1.83±0.04) kg (Table 1). However, the difference in weight recorded across the groups in comparison with the control was statistically significant (F = 9.119, df =19, p < 0.05).

Table 1. Daily Feed Intake (%) and Body Weight (kg) of female African giant rat in each group per week during the experimental period

GROUP	MEAN DAILY FEED INTAKE (%)	MEAN BODY WEIGHT (KG)				
		WEEK1	WEEK2	WEEK3	WEEK4	WEEK5
1	99.67±0.32a	1.65±1.69a	1.80±0.03a	1.86±0.39a	1.84±0.39bc	1.83±0.04b
2	99.15±0.80a	1.44±0.04a	1.64±0.04a	1.62±0.08a	1.85±0.04ab	1.83±0.04ab
3	99.14±0.40a	1.46±0.76a	1.61±0.07a	1.62±0.81a	1.56±0.07a	1.57±0.07a
4	100.00±0.00a	1.52±0.04a	1.67±0.30a	1.74±0.40a	1.74±0.35c	1.74±0.04b

Means with the same alphabet within a column are not significantly different (p > 0.05)

Group 1 (80% of Commercial rat feed + 20% of *T. neriifolia* seed powder); Group 2 (60% of Commercial rat feed + 40% of *T. neriifolia* seed powder); Group 3 (20% of Commercial rat feed + 80% of *T. neriifolia* seed powder); Group 4 (Control: Only Commercial rat feed

Effect of *T. neriifolia* seed powder on kidney, heart, and liver of the female African giant rats

The histomicrographs of the kidneys of the female African giant rats from each the treatment group showed that there was no observable lesion

in the groups that received the lowest and mid concentrations of the experimental feeds with the control (Plate 3). However, the histomicrograph obtained from the kidney of the group fed with the highest concentration of the feed shows random tubular epithelial coagulation necrosis (Plate 3).

The histomicrographs of the liver of the female African giant rats from each of the treatment groups show no observable lesion in all the groups except in the group fed with mid concentration of the experimental feed which shows moderate centrilobular hepatocellular degeneration and Kupffer cell hyperplasia (Plate 4).

The histomicrographs of the hearts of the female African giant rats from each of the treatment groups show that there was no observable lesion in the groups that received the lowest and mid concentrations of the experimental feeds with the control (Plate 5). Meanwhile, the histomicrograph obtained from the heart of the group fed with the highest concentration of the feed shows moderate atrophy of myofibres.

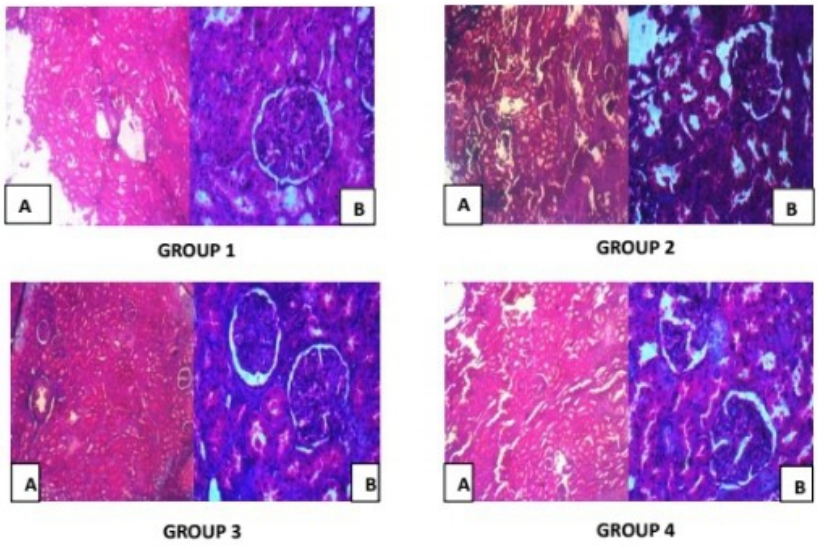


Plate 3: Histomicrograph of the Kidney of the female African giant rat fed with lowest (Group 1), mid (Group 2) and highest (Group 3) Concentration of experimental feed including control (Group 4) (A) x100 (B) x400

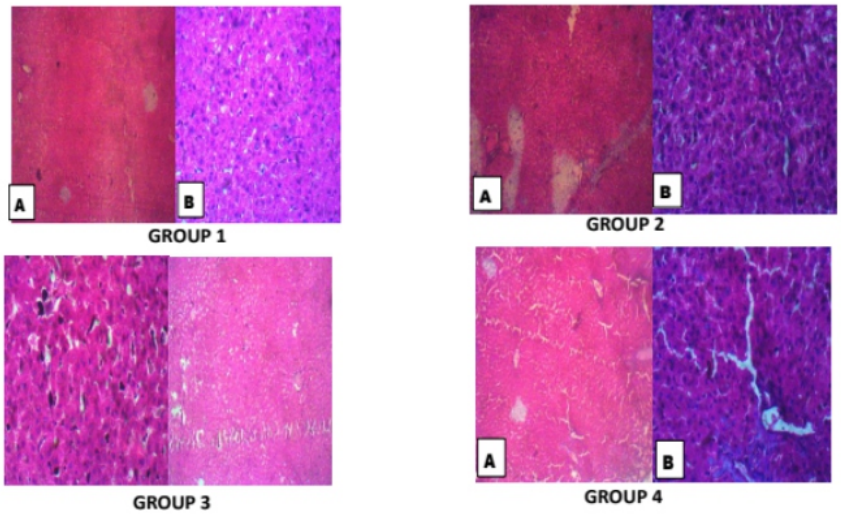
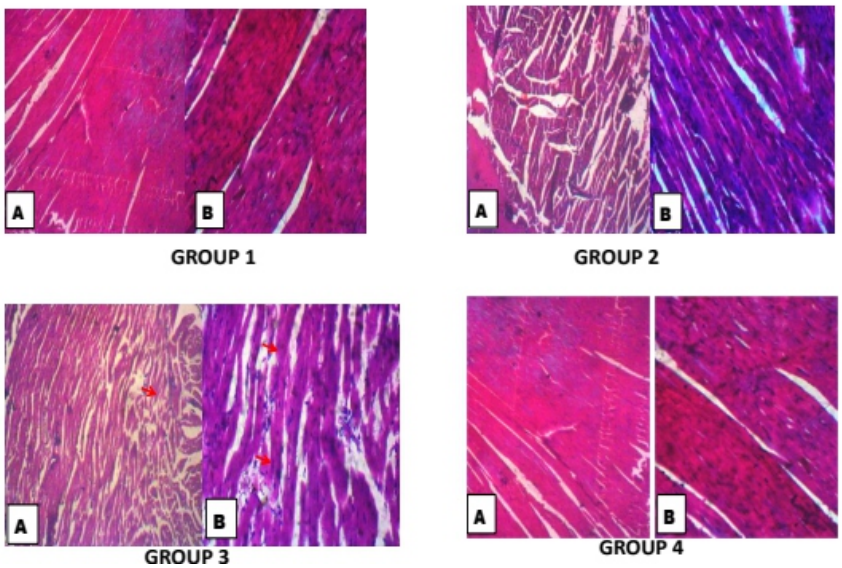


Plate 4: Histomicrograph of the Liver of the female African giant rat fed with lowest (Group 1), mid (Group 2) and highest (Group 3) Concentration of experimental feed including control (Group 4) (A) x100 (B) x400



Pplate 5: Histomicrograph of the Heart of the female African giant rat fed with lowest (Group 1), mid (Group 2) and highest (Group 3) Concentration of experimental feed including control (Group 4) (A) x100 (B) x400

DISCUSSION

The feed intake of the experimental animal was not significantly affected as revealed from the result of the present study. This contradicts the findings of the previous research by Fayinminnu *et al.* (2022) who reported that the seed are unpalatable and their consumption caused slight numbling sensation which led to dryness in the throat of the experimental animals, hence the low feed intake. Several factors could result to this such as non-palatability of the feed. The number of times the rat was fed with the seed per day and the proportion of seeds to commercial feed in the diet.

The present study revealed that the experimental feed supplied did not have significant effect on body weight of female African giant rats used for the study except in weeks 4 and 5 where there was significant reduction in the body weight especially in group 3 compared with the control. This is contrary to previous findings reported by Taiwo *et al.* (2004) who stated that *T. neriifolia* seed cake even at lowest dosage had significant effect on experimental rabbits. This could be as a result of concentration of the seed cake incorporated in the experimental feed.

The study recorded no mortality or morbidity during the experimental period. This could be due to the fact that the concentration of the seed powder of *T. neriifolia* used did not have rodenticidal effect on the female African giant rats presumably because the animals were caught from the wild and would probably have more adaptive lifestyle than the laboratory rodent as reported by Fayinminnu *et al.* (2017).

The result of the present study showed that *Thevetia neriifolia* did not have rodenticidal effect on the wild species of rodent used (*Cricetomys gambianus*). This is contrary with the previous findings reported by Fayinminnu *et al.* (2017) who stated that *T. neriifolia* seed powder incorporated in the commercial rat feed had rodenticidal effects on laboratory rodent used as the experimental animal.

Several factors could be responsible for this. It could be as a result of animal used (laboratory animal), the environment where the experiment was conducted has a significant effect on the animals in the reaction to experimental feed (Olayioye *et al.*, 2014; Fayinminnu *et al.*, 2017).

Yarkwan (2013) evaluated the effect of *Thevetia neriifolia* seed oil on Wistar Albino rat. The result showed no significant effect in the liver and kidney when the rats were fed with the seed, that is, the organs were not affected but there was decrease in

body weight and feed intake when compared with the control group. There was no decrease in the liver triglycerides and only a slight decrease in the heart triglycerides.

All these observations were discovered on the African giant rat used for this study. There was no observable lesion on the heart, kidney and liver. Only a random tubular epithelial coagulation necrosis was found on the kidney of one animal used.

CONCLUSION

The study established that *Thevetia neriifolia* seed does not have rodenticidal effect on the wild female African giant rat. No mortality and behavioral changes were observed in the animals at all the concentrations. However, there was significant reduction in the mean weight of the animals when fed with the experimental feed.

T. neriifolia seed powder did not have rodenticidal effect on the wild female *C. gambianus*. Therefore, it could not be used as a rodenticide in the management of wild rodent species population.

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CONFLICT OF INTEREST

There is no conflict of interest among the authors.

AUTHORS' CONTRIBUTION

The first and corresponding author designed and supervised the project, carried out the statistical analyses as well as edited the write-up. Samples were collected and laboratory analyses were carried out by the last author. All the authors made inputs in the methods used in the study. Proofreading and editing were done by the first and second authors.

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