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Research Article

Evaluation of Ichthyotoxicity Activity of *Raphia farinifera* (Gaertn.)Hyl. (Arecaceae) Fruits Extract

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Abstract

Harvesting of fish in Nigeria by local artisans is done in various ways. One of these ways is the use of plant extract as poison to kill fish in various fresh water habitats. This research was conducted in order to evaluate in detail fish poisoning potentials (Ichthyotoxicity) of *Raphia farinifera* aqueous fruits extract. Ripped fruits of R. *farinifera* were harvested from raffia palm tree, and soaked in water for 7 days to enable them decay properly so that the brown-reddish imbricate scales covering the ovoid glossy fruits can be removed from the fruits, thus exposing the yellowish fruits. After this, the fruits were pounded into coarse form using local mortar in water, and covered with polythene for 48 hrs. The yellowish solution from the polythene was collected into a beaker, evaporated into dryness and tested for phytochemical constituents, while the coarsely pounded fruits were tightly covered for use. Preliminary phytochemical screening of the extract solution showed that it contained saponins, alkaloids, tannins, flavonoids and anthraquinones. Coarsely pounded fruits sprayed into various basins containing different species of fresh water fish produced 100 % mortality rate in dose dependent fashion in all the fish families. Surface dwelling fish like *Oreochromis niloticus* (Cichlidae), *Gnathonemus petersii* (Mormyridae), *Hippopotamyrus psittacus* (Mormyridae), *Labeo parvus* (Cyprinidae) and *Tilapia zillii* (Cichlidae), were mostly affected by extract, than the bottom dwellers like *Protopterus annectens* (Protopteridae), *Gymnarchus niloticus* (Gymnarchidae), and *Heterobranchus longifilis* (Clariidae). Toxicity studies showed that aqueous extract of *Raphia farinifera* fruits is used as fish poison (Ichthyotoxicity), it thus serve as an alternative means of harvesting fish locally from Nigeria fresh water.

Keywords: Ichthyotoxicity, *Raphia farinifera*, Fresh water, Phytochemical, Doses.

INTRODUCTION

Raphia farinifera (Gaertn.) Hyl Belongs to the family Arecaceae (Palmae), and it is a native of Madagascar also found along Africa's eastern coast over woody marsh lands or river banks (Burkill, 1985). It is commonly called Raffia palm; it has about 20 species which are native to tropical regions of Africa as well as Central and South America (Cracker and Simon, 1986).

The plant grows up to 16m tall with leaves which are compound pinnate about 25m long and 3m wide. It is monocarpic, flowering once and then dying after the seeds are matured (Dalziel,1956). Economically, it has been used for various purposes in DR Congo as ropes, sticks, roof covering and in textile (Burkill, 1985). In Nigeria, fibre from the plant is used in making local bed, building construction and source of palm wine. Chemically, it was reported to contained glycosides and tannins (Gill, 1992). Despite the overwhelming use of the plant, no report of the use of this plant fruits as fish poison

was recorded. The need to find a safe and stable means through which fresh water fish can be harvested has become a source of concern to large fish farmers, hence, justifies why this research was carried out.
This present study was carried out to evaluate Ichthyotoxicity activity of *Raphia farinifera* fruit extract on selected Nigeria fresh water fishes.

MATERIALS AND METHODS

Collection and Preparation of Fruits

Ripped fruits of *Raphia farinifera* were collected from the tree at the bank of River Anambra in Ogurugu, Enugu State, Nigeria. They were soaked in a big basin filled with water and left for seven days. After this, the fruits were poured into local mortar, pounded into coarse form to remove the imbricate scales, and covered with polythene (water proof) for 48 hours.

Phytochemical Screening of Extract Solution

Using appropriate procedures (Evans, 2006), the solution from pounded fruits was tested for the presence of some phytoconstituents.

Ichthyotoxicity Testing of Extract Stocking of Fish Families

Fifteen basins were filled with water to equal volumes, and labelled accordingly to represent each fish families. Another basin was stocked with all the species of fish, and used as control. The basins were stocked in the order of 1 to 16 as follows: 1. *Clarias anguillaris* (Clariidae); 2. *Ctenopoma muriei* (Anabantidae); 3. *Garra ornate* (Cyprinidae); 4. *Gnathonemus petersii* (Mormyridae); 5. *Gymnarchus niloticus* (Gymnarchidae); 6. *Heterobranchus longifilis* (Clariidae); 7. *Heterotis niloticus* (Arapaimidae); 8. *Hippopotamyrus psittacus* (Mormyridae); 9. *Hydrocynus forskahlii* (Alestidae); 10. *Labeo parvus* (Cyprinidae); 11. *Oreochromis niloticus* (Cichlidae); 12. *Pellonula leonensis* (Clupeidae); 13. *Protopterus annectens* (Protopteridae); 14. *Synodontis filamentosa* (Mochokidae); 15. *Tilapia zillii* (Cichlidae); 16. collection of all the fishes.

These fishes were collected from the National Research Institute for Fresh Water Fisheries (NRIFWF) New-Bussa, Niger State, Nigeria. All fish collected were matured; no fingerlings were used in the experiment.

Spraying Techniques of Extract

The coarsely grind fruits of *R. farinifera* were mixed slightly with mud so that it can sink easily into bottom of the basins, and sprayed using ordinary hand; covered with hand glove. Basins were agitated to ensure that the extract was properly spread in the water containing fish families. Each of the basins which contained 20 fishes (except the control that contain 15 fishes) were left to stand for 20 minutes and observed.

Mortality rate in each basin was noted by picking out those fish that do not survive the intoxication. The experiment was repeated three times by increasing the concentration of the extract applied in the basins. Concentration was measured using graduated cylinder 50 to 1000 cm^3 volumes. Histopathological examinations were carried out on the fish species that do not survive ichthyotoxicity testing from each basin on organs such as gills, eyes, scales, lungs, liver and lateral lines.

Acute and Chronic Toxicity Testing of Extract

Aqueous extract of *R. farinifera* was later on subjected to acute (LD_{50}) and chronic toxicological studies (*in vivo*) using 13 male albino mice (Lorke, 1983). This is to know if the fish are safe for eating by humans. Histopathological examinations of organs and behavioural changes in the animals were observed after extract administration orally and intraperitoneally (I.P). The experiment was brought to end after 12 weeks and the survived animals were sacrificed following international guidelines (Bombardell, 1978).

RESULTS AND DISCUSSION

Results of preliminary phytochemical studies showed that the extract of fruits of *R. farinifera* contained saponins, alkaloids, anthraquinones, flavonoids and tannins (Table 1).

Table 1. Phytoconstituents of R. farinifera Fruits Extracts

Constituents	Observation
Saponins	*+ + + +
Alkaloids	+ +
Tannins	+ +
Flavonoids	+ + +
Anthraquinones	+

INDICATORS: ++++ (Highly present), +++ (High present), ++ (Moderately present), + (Low present), * indicated by the level of foam and haemolysis.

In table 1, the intensity of colour change was used to indicate whether the constituents were highly, high, moderately or low present. Saponins were observed to be highly present in the extract while anthraquinones were low in the extract. Ichthyotoxicity evaluation of the fruits extract showed that most of the surface dwelling fish families were affected more than the bottom dwellers. However, fish whose operculum was not tightly closed were also killed by the extract. It is possible that the extract deprived dissolved oxygen from the fish thereby making their gills change colour from red to pale red initially to almost whitish 20 minutes later (Table 2).

Table2. Effects of *R. farinifera* Fruit Extract on Fish

Fish name	Family	Fish Died (Fish Survived)
Clarias anguillaris	Clariidae	[a]10 (10)
Ctenopoma muriei	Anabantidae	15 (5)
Garra ornate	Cyprinidae	16 (4)
Gnathonemus petersii	Mormyridae	**20 (0)
Gymnarchus niloticus	Gymnarchidae	* 6 (14)
Heterobranchus longifilis	Clariidae	8 (12)
Heterotis niloticus	Arapaimidae	[a]10 (10)
Hippopotamyrus psittacus	Mormyridae	**20 (0)
Hydrocynus forskahlii	Alestidae	18 (2)
Labeo parvus	Cyprinidae	**20 (0)
Oreochromis niloticus	Cichlidae	18 (2)
Pellonula leonensis	Clupeidae	15 (5)
Protopterus annectens	Protopteridae	*2 (18)
Synodontis filamentosa	Mochokidae	17(3)
Tilapia zillii	Cichlidae	**20 (0)
All the fish (control)	-	15(15)

INDICATORS: **Mostly affected fish, *lowest mortality rate, [a] mortality of ≥ 10 was considered susceptible to extract ichthyotoxicity effects.

Those fish species that were not affected adversely as a result of intake of extract doses had very thick scales or very tightly fitted scales and mechanism that protect their lateral lines from osmotic shock of the extract because, the presence of saponins had been reported to produce lateral line osmotic shock in water thereby breaking the linkage along lateral lines in aquatic organisms (Udom, 2006). It is possible that higher concentration of saponins in the extract caused abnormal lateral lines in families Mormyridae, Cyprinidae and Cichlidae. Saponins in itself had been used as fish poison because it converts the aglycone portion of the glycoside to genins when in solution (Kar, 2007). These genins then combined with the haem portion of the RBC to form partial or total clot of the blood thereby impairing normal blood circulation in the organism (Bannerman, 2002).

An increase in dose of the extract resulted in blockage of the blood capillaries supplying blood to the gills and other respiratory organs in fish, and this was responsible for the change in colour of the gills in the dead fish families. There were sharp decreases in the level of dissolved oxygen (DO) in the basins because saponins, tannins and alkaloids, when in aqueous solution form soluble complexes of basic salts which react with oxygen in water to form poison

substance such as genins and Acetogenins; these chemicals when taken in by fish change their metabolic systems [Ayensu and Baba, 2008).

Ukwubile, (Ukwubile, (2012), reported that alkaloids in crude drugs whether in higher or lower quantities have either beneficial or harmful effects in animals. It is possible that the presence of alkaloids in this plant extract was beneficial to some families like Protopteridae and Gymnarchidae that witnessed little or no mortality at all (Table 2). In all cases, fish that dwell mainly on surface of water were affected mostly than those at the bottom, even with slight agitation of the basin so that the extract can reach both parts of water. The fact that aqueous extract of R. farinifera fruits produced mortality effect in all the families showed that it contains phyto constituents which are capable of killing fish and other aquatic organism.

In this present study, toxicity study revealed that aqueous extract of the fruits at lower concentration was safe. This is because, acute toxicity study of extract in mice at doses 10, 100, and 1000 mg/kg bow of extract in phase one produced no deaths at all. However, results of chronic toxicity study showed necrosis of the liver, arteriosclerosis, partial calculus of the lungs, kidneys and spleen, as well as oedema during Histopathological examinations (table 1).

The study therefore, showed that since the LD50 was found to be greater than 5000 mg/kg (I.P), the extract was safe and the fish killed by the extract can be eaten. Thus, fruits extract of *Raphia farinifera* represent another source for which fresh water fish can be harvested easily.

CONCLUSION

Use of chemicals that are synthesized by artisans to harvest fish from fresh waters had produced cumulative effects on lives in water for the past years (Sofowora,1986). However, fruits extract of the plant represents a safe and easy way of harvesting fish from a large fresh water body, its use should be controlled. It is pertinent to mention at this juncture that the use of *R. farinifera* fruit extract at higher doses produced 100 % mortality rate in fish families, which can destroy both egg and fingerlings.

The study showed that the presence of higher concentration of saponins and other phytoconstituents in the fruits extract were responsible for ichthyotoxicity activity of the plant.

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