Potential of *Moringa oleifera* (Lam.) fresh root-bark extract as an organic piscicide in aquaculture pond management

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Abstract

This study examined the effectiveness of *Moringa oleifera* fresh root-bark extract as an organic piscicide to control predatory fish in ponds. Acute-lethal toxicity (LC₅₀) of *Moringa oleifera* extract for 96-h exposure for *Oreochromis niloticus* fingerlings was determined at 26.45 mg Γ^1 . The extract was more toxic at higher concentrations of 100 mg Γ^1 , with fingerlings showing abnormal swimming, restlessness and uncoordinated behaviour before death. *Moringa oleifera* extract could be used as an organic piscicide in aquaculture pond management. Baseline information on its toxicity to fish could serve as a tool in fisheries management to wipe out predatory fish in ponds prior to stocking.

Keywords: Toxicity, Oreochromis niloticus, Fisheries management, LC50

Introduction

Elimination of unwanted fish is a common practice among fish farmers prior to stocking with desirable fish fingerlings. However, the synthetic piscicides used to eliminate unwanted fish are persistent and could get into the food chain. Thus the use of environmentally safe organic piscicides as an alternative is being embraced in aquaculture because they are biodegradable over short time periods, and there is the possibility that fish killed by them are edible with no health hazards (Chiayvaresajja *et al.* 1997; Agbon *et al.* 2004; Akinbulumo *et al.* 2004; Akinwade *et al.* 2007). Some organic piscicides have been identified and used by different researchers around the world as fish pond management tools to eradicate predators in fish ponds (Adewole 2002; Tiwari *et al.* 2003; Adeogun 2004; Agbon *et al.* 2004; Tiwari & Singh 2004; Tiwari 2005). These organic piscicides include extracts from plants such as *Parkia biglobosa, Raphia hookeri, Derris elliptica, Euphorbia tirucalli* and *Nerium indicum*.

Currently, there is scanty information in the literature as regards the use of fresh rootbark extracts of *Moringa oleifera* as an organic piscicide in aquaculture pond management in the developing world. *Moringa oleifera* is a tree belonging to the family Moringaceae, usually growing outside the forest areas of south-western Nigeria (Adesina *et al.* 2008). The tree is under-utilized and contains several toxic compounds, such as phenol, tannins, saponins, glucosinolates, oxalic acid, lectins, moringine and moringinine (alkaloids), pterygospermin, spirochin and benzyisothiocyanate (Berger *et al.* 1984; Grabow *et al.* 1985; Makkar & Becker 1997; Fuglie 2001; Fahey 2005; Wise 2006). *Moringa oleifera* is suggested as an alternative to synthetic piscicides because it is commonly available, is less expensive for fish farmers and has a lower toxicity against non-target species. Its piscicidal potential is embedded in the rootbark which contains the alkaloids that act on the nervous system of fish and other terrestrial animals (Fuglie 2001; Adesina 2008).

Therefore the objective of this research was to determine the acute lethal toxicity (LC_{50}) of *Moringa oleifera* fresh root-bark extract to *Oreochromis niloticus* fingerlings exposed for 96 h.

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Materials & Methods

180 Oreochromis niloticus L. fingerlings were obtained from the fish farm of the Wildlife & Fisheries Management Department, University of Ibadan, Nigeria. The fingerlings selected for the experiment had a mean total length of 5.41 ± 0.25 cm and weight 5.4 ± 0.28 g, with no record of prior exposure to the toxicant. The fishes were held in translucent plastic 55-litre aquaria covered with transparent mosquito net to prevent fish from jumping out of the aquaria during the experiment: aeration of the dechlorinated water was constantly maintained. The fish were acclimated for 14 days, and fed twice daily with commercial floating feed containing 30% protein; they were fed 3% of their body weight as a maintenance ration (Bhagwant & Bhikajee 2001; Omitoyin *et al.* 2006). Unused feed and faeces were siphoned out daily to prevent water contamination (Oyelese & Faturoti 1995).

Fresh roots of *Moringa oleifera* were collected from the nursery unit of the Department of Forestry Resources Management, University of Ibadan. The bark was removed, cut into pieces with a knife, and later ground with a pestle and mortar to obtain macerated samples. Aqueous extracts were obtained using a cold-water extraction technique in order to simulate local procedures (Ofogba *et al.* 1998). 50 g of macerated freshly prepared samples of root-bark were weighed into a conical flask, 100 ml distilled water added and shaken before filtering using a dry whatman filter paper, and then the filtrate poured into a 100-ml graduated measuring cylinder. The cold-water extract was stored at 4 ^oC inside a refrigerator for immediate use.

A preliminary toxicity test was conducted (Odiete 1999; Reish & Oshida 1986; Pandey *et al.* 2008) using a spacing factor of 10 to determine the lethal concentrations (LC₅₀) of over 24 h. The following concentrations of fresh extract were prepared by dissolving 50 g in 100 ml distilled water, and serially diluting to produce 5000 mg Γ^1 per 5 l water (Odiete 1999): 0, 0.1, 1, 10, 100, 1000 mg Γ^1 . The fish were starved for 24 h before the experiment. Five fish were tested on each concentration, and the mortality and clinical behaviour monitored and recorded after the toxicant was introduced into the aquaria. The definitive toxicity test referred to as acute toxicity was carried out based on the results of the prelimary test, but a spacing factor of 1.8 and a duration of 96 h were used (as earlier described by Odiete 1999).

Ten fingerlings of *Oreochromis niloticus* starved for 48 h were released into each aquarium of 55 litres capacity, and one of the following concentrations of extract introduced: 0, 10, 17, 31, 56 and 100 mg I^{-1} . Fish mortality was monitored and recorded hourly for the first 4 h, every 4 h for the next 24 h, and subsequently every 24 h until the time limit of 96 h (Ayotunde & Ofem 2008). The inability of fish to respond to external stimuli was used as an index of death (Ayotunde & Ofem 2008). Fish behaviour such as erratic swimming, air gulping, loss of reflex, discolouration and moulting was monitored during the experiment. The experimental design was a complete randomized design with six treatments and three replicates per treatment.

Water quality monitoring was carried out prior to, during and after the experiment. The physico-chemical parameters of the water (dissolved oxygen, temperature and pH) were measured using the APHA (1998) method of water quality assessment. Mortality data were analysed using probit software of USEPA (2000). The median lethal concentration (LC_{50}) at the selected period of exposure and associated 95% confidence interval for each replicate toxicity test was subjected to logit and probit analysis (Finney, 1971). Water quality parameters were subjected to one way analysis of variance.

Results

Control fish showed no abnormal behaviour during the experimental period of 96 h. Fish treated with the highest concentration of 100 mg l^{-1} of *Moringa* extract showed various toxic

reactions, such as erratic swimming, restlessness, air gulping, increased opercular beat. These abnormal behaviours progressively (through time) manifested themselves in the other lower concentrations of 10, 17, 31, and 56 mg I^{-1} . Increases in concentration and exposure time resulted in loss of scales and haemorrhages in exposed fish. Dead fish were covered with excessive mucus, and fish died with stiff fin rays. The highest mortality was recorded at 100 mg I^{-1} over 96 h (Table 1).

Concentration	Mortality out
of extract	of 30
$(mg l^{-1})$	
0	0
10	8
17	9
31	11
56	21
100	30

Table 1: Acute toxicity of Moringa oleifera fresh root-bark cold-water extract applied
to Oreochromis niloticus fingerlings for 96 h. Each replicate involved 10
fish. The slope of the probit analysis of mortality against concentration was
 2.12 ± 0.60 (95% confidence limits 0.958 to 3.295). LC₅₀ was estimated at
26.5 mg l⁻¹ (95% confidence limits 15.3 to 42.0).

Discussion

The water quality parameters obtained in the present study are within the range of water quality recommended for culturing tilapia species (Olaifa *et al.* 2008; Ayotunde & Ofem 2008; Adesina 2008). Fingerlings of *Oreochromis niloticus* exhibited a range of abnormal behaviours at higher concentrations of the *Moringa* extract; normally, fish exposed to toxicants exhibit different behavioural changes (Santhakumar & Balaji 2000; Fafioye 2001; Ayuba & Ofojekwe 2002, Chung–MinLiao *et al.* 2003; Adeogun 2004; Adesina 2008; Ayotunde & Ofem 2008; Olaifa *et al.* 2008; Pandey *et al.* 2008) that adapt them to the toxin. Here, fish exposed to higher concentrations exhibited toxic reactions that later resulted in death (Fafioye 2001; Fafioye *et al.* 2004; Omitoyin 2006). Mortality was dose-dependent and increased with time.

The LC₅₀ value for *Oreochromis niloticus* fingerlings (26.5 mg l^{-1}) is quite high; estimates vary in different fish species and in the same species under different conditions (Omitoyin *et al.* 1999; Omitoyin *et al.* 2006; Pandey *et al.* 2008). Verma *et al.* (1982) recorded values of 5.14, 4.8, 4.67 and 4.57 mg l^{-1} for 24, 48, 72, and 96 h respectively in *Heteropneustes fossilis* exposed to dimethoate; at different seasons the 96-h values were 14.39 and 2.98 mg l^{-1} for the dry and wet season respectively (Pandey *et al.* 2008). The 96-h LC₅₀ of pawpaw seed powder was 18 mg l^{-1} (Ayotunde & Ofem 2005). Some species are very resistant to some taxons: for example, 5878, 4865 and 610 mg l^{-1} were recorded for *Sarotheordon galilaeus* exposed to ethanolic extracts of cocoa bean shell (Olaifa *et al.* 2008). A 96-h LC₅₀ value for dimethoate on *Clarias batrachus* was 65mg l^{-1} (Begum & Vijayraghvan, 1995).

Differences observed in the values of LC_{50} from the current research as compared to other values obtained elsewhere show that fish become more sensitive to any toxicant with greater exposure times; increased mortality is also know to result from increased temperatures (Patra *et al* 2007). Variation in LC_{50} thus depends on a number of biological and physico-chemical factors, reported by many earlier workers (Pandey *et al.* 2008).

In conclusion, fish exposed to fresh root-bark extract of *Moringa oleifera* at different concentrations exhibited toxic responses which eventually lead to death. This extract could be used as an organic piscicide in aquaculture pond management, and baseline information on its toxicity to fish could serve as a fisheries management tool to wipe out predatory fishes in ponds prior to stocking.

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الملخص العربي

الاستخدامات الممكنة لمستخلصات الجذور الطبيعية لأشجار البان (Lam.) كمبيد عضوي للأسماك يسهم في التحكم في أحواض تربية الأحياء المائية

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الملخص العربي

بحثت هذه الدراسة فعالية الخلاصة الطبيعية لجذور أشجار المورينجا في العمل كمبيد عضوي للتحكم في الأسماك المفترسة في الأحواض. تم تحديد الجرعة المميتة (LC50) لخلاصة أشجار المورينجا بعد 96 ساعة من تعرض زريعة البلطي النيلي لها بحوالي26،45 ملجم/ لتر⁻¹. ولقد كانت خلاصة النبات أكثر سمية على زريعة تلك السمكة عندما أعطيت لها بتركيز أعلى يبلغ 100 ملجم/ لتر⁻¹ ، مما أدى لسباحتها بشكل غير طبيعي وإصابتها بالأرق والسلوك غير المنسق وبعد ذلك الموت. وبهذا يمكننا القول بأنه يمكن استخدام خلاصة أشجار البان كمبيد عضوي للأسماك يساعد على المندق أحواض تربية الأحياء المائية، حيث أثبتت المعلومات الأساسية عن سمية تلك الخلاصة إمكانية المنعر وبعد المصايد للقضاء على الأسماك المفترسة في أحواض تربية الأسماك قبل تخزينها.