EVALUATION OF BOTANICAL PISCICIDES ON NILE TILAPIA *OREOCHROMIS NILOTICUS* L. AND MOSQUITO FISH *GAMBUSIA AFFINIS* BAIRD AND GIRARD

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INTRODUCTION

- The use of piscicides as a tool in pond management during pond preparation to get rid of predators before fish stocking is important.
- Ideally, ponds should be sundried and the pond bottom cracked dried to help get rid of fish predators. However, this practice is not always possible particularly during the wet season. Moreover, farmers who are always in a hurry to prepare their ponds always resort to the use of inorganic/chemical fish toxicants.

Introduction

- In the country today, there is no legally registered fish toxicant except for some organics such as tea seed cake and tobacco dust.
- In view of this, farmers resort to non-conventional and unregistered fish toxicants such as agro-pesticides and sodium cyanide because they are fast acting and readily available in the market. However, these chemicals may have negative effects on the environment and farmers' health.
- Hence, there is a need to explore other environment- and health-friendly fish toxicants such as botanical plants with piscicidal activity.

Introduction….. Plants as piscicides

- Plants are virtually inexhaustible source of structurally diverse biologically active substances (Istvan, 2000).
- Some plants contain compounds of various classes that have insecticidal, piscicidal and molluscicidal properties.
- Unlike synthetic chemical pesticides which leave harmful residues in the aquatic environment (Koesomadinata, 1980; Cagauan, 1990; Cagauan and Arce, 1992), botanical insecticides are believed to be more environment friendly compared to synthetic chemicals because they are easily biodegraded and leave no residues in the environment.
- Since some of these pesticidal compounds present in plants are also toxic to fishes, botanical pesticides have potential to be used as piscicide to eradicate unwanted fishes in the pond.

Introduction….. Plants as piscicides

- Many plants from different families have been applied for catching fish the world over such as of the genera *Derris, Tephrosia* and *Lonchocarpus* of the family L*eguminosae*.

- The toxic parts of plants employed as fish poisons can include roots, seeds, fruits, bark, latex or leaves.

- Plants have been reported to have molluscicidal action (Rejesus and Punzalan, 1997) hence, they may have high piscicidal action.

Plants presently used as piscicide

Plant material	Application rate	Author
Derris roots	4g/m^3 1 kg/ha	Lunz and Bearden, 1963 Chakroff, 1976
Teaseed cake	150 kg/ha	Chakroff, 1976
Camellia seed cake	50-200 kg/ha	Chakroff, 1976
Powdered croton seed	50-200 kg/ha	Chakroff, 1976

Bioassay Test

Plant	LC_{50} (g/l) Nile tilapia	Common carp
Blumea balsamifera	*1.54*	*1.37*
Vitex negundo	*4.95*	*3.53*
Azadiracta indica	*1.59*	*0.55*
Tinospora rumphii	*0.77*	*2.13*

From: Leaño and Cagauan (1994)

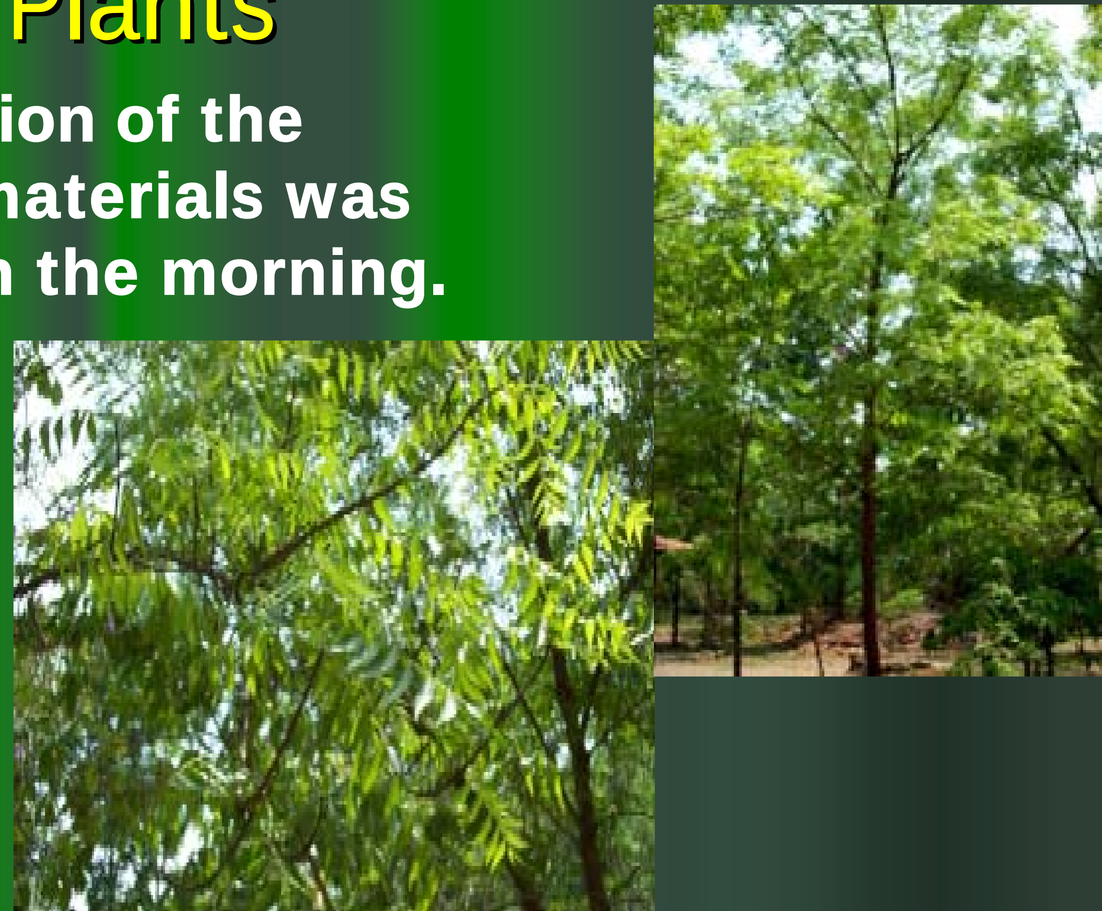
Objectives of the Study

- **The study assessed the piscicidal activity of ten locally available plants to two freshwater fishes: Nile tilapia (*O. niloticus* L.) and mosquito fish (*G. affinis* Baird and Girard*)*.**
- **It focused on the laboratory determination of lethal concentrations (LC_{50} and LC_{100}) through a static bioassay test.**

Methodology

Test Plants

Collection of the plant materials was done in the morning.



Adelfa
Nerium indicum Mill.
Apocynaceae
Leaves

Neem
Azadirachta indica
Meliaceae
Leaves

Test Plants

Makabuhai
Tinospora rumphii Boerl.
Menispermaceae
Leaves

Physic Nut
Jatropha curcas L.
Euphorbiaceae
Stems

Test Plants

Sambong

Blumea balsamifera L.
Asteraceae
Leaves

Calamansi

Citrus mitis Blanco
Rutaceae
Leaves

Test Plants

Agave

Agave cantala Roxb.

Agaveceae

Leaves

Lagundi

Vitex negundo L.

Verbaceae

Leaves

Test Plants

Ampalaya/Bitter gourd
Momordica charantia L.
Cucurbitaceae
Leaves

***Gliricidia sepium* (Jacq.) Steudel**
Leguminosae
Leaves

Experimental set up

Rectangular plastic containers each measuring 16 cm x 11 cm x 5 cm and provided with net covers were used. Each container was filled with 500 ml distilled water. The water was aerated first to full oxygen saturation for 20 minutes before use.

Preparation of plant extracts

- Plant materials for assay were prepared in water-extracted form.
- Fresh plant material was weighed using the Mettler balance and then processed in a food blender.
- Distilled water was added to the chopped plant material before grinding.
- The ratio of plant material to the volume of the distilled water added was 1:2 or 100 g of plant material added to 200 ml of distilled water.
- The extracts and solid plant materials were separated by hand squeezing using cheesecloth.
- The plant extract was used immediately after extraction to ensure its freshness.

Fish species

Nile tilapia *Oreochromis niloticus*

average weight: 0.17 g

Mosquito fish *Gambusia affinis*

average weight: 0.19 g

Fishbase 2000

Test concentrations used for the different plants tested on Nile tilapia and mosquito fish

Plant	Test concentration ($ml.l^{-1}$)	
	Nile tilapia	Mosquito fish
Agave	0, 10, 30, 50, 70, 100	0, 20, 50, 100, 150, 200
Neem	0, 2, 4, 6, 8, 10	0, 2, 4, 6, 8 , 10
Madre-de-cacao	0, 8, 40, 60, 90, 125	0, 50, 80, 100, 140, 200
Ampalaya	0, 1, 2, 3, 4, 5	0, 2, 6 10, 12, 15
Adelfa	0, 0.1 , 0.5, 1.0, 1.5, 2.0	0, 2, 4, 6, 8, 10
Makabuhai	0, 0.1, 0.2, 0.4, 0.6, 0.8	0, 2, 4, 6, 8, 10
Sambong	0, 40, 60, 80, 120, 150	0, 10, 40, 80, 100, 120
Lagundi	0, 10, 15, 20, 30 40	0, 5, 10, 20, 36, 50
Kalamansi	0, 0.5, 1, 2, 5 10	0, 0.5, 1, 2, 3, 5
Physic Nut/tuba	0, 5, 10, 14, 20, 30	0, 20, 50, 60, 80 , 90

Bioassay test

- Standard static bioassay procedures were employed based on APHA, AWWA and WPCF (1971; 1980).
- Fish mortalities were observed and recorded at 24, 48, 72 and 96 hours from stocking.
- Dead fish were removed immediately. A fish is considered affected by the plant toxicant when it manifests erratic swimming behavior, hyperactivity, hyperventilation and pronounced ataxia coinciding with decreased capacity to respond to visual stimuli. A fish is considered dead when it does not respond to mechanical prodding.

Lethal concentrations

- The lethal concentrations (LC_{50} and LC_{100}) of each of the test plants were determined by plotting concentrations of the plant against fish mortality within 24 hours, 48 hours, and 96 hours after exposure to the treatment.
- Interpolation between two concentrations where the mortality occurred at less than and greater than 50 % was done.
- LC_{50} or median lethal concentration is the concentration at which 50 % survived and 50 % died of the test fish. It is the basis of most toxicity and tolerance tests.
- LC_{100} is the lowest concentration at which 100 % of the fish died. It is the basis of the piscicidal activity of test plants because the purpose of using a piscicide is to ensure total eradication of unwanted fishes.
- Trendline analysis using linear regression in Microsoft Excel was used to estimate LC_{50} and LC_{100}.

Water quality parameters

Water temperature	Digital pH meter
pH	YSI DO meter Model 55
dissolved oxygen	YSI DO meter Model 55

Data Gathered

- Fish mortalities at 24, 48, 72 and 96 hours
- Median lethal concentration (LC_{50})
- Lethal concentration (LC_{100})
- Water quality parameters such as temperature, dissolved oxygen and pH

Results and discussion

Toxicity expressed as 96-hr LC_{50}

Results and discussion

Lethal concentrations for Nile tilapia

Plant	LC_{50}			LC_{100}		
	24-hrs	48-hrs	96-hrs	24-hrs	48-hrs	96-hrs
Agave	52.86	40	30	74.29	52.5	130
Neem	6.4	3.22	2.57	12.4	4.89	4
Madre de Cacao	75	73.93	52	90	90	72
Ampalaya	1.71	1.5	0.45	2.59	2.44	1.82
Adelfa	0.62	0.069	0.083	1.06	0.18	0.5
Makabuhai	0.69	0.53	0.44	0.82	0.68	0.64
Sambong	97.14	88.42	5.11	125.71	120	84.44
Lagundi	23.99	15.67	2.93	31.5	20.67	20.77
Calamansi	3.5	3.33	3.12	5	5	5
Jatropa	18.67	17.27	12.8	26.67	25.45	21.8

Lethal concentrations for Mosquito fish

Plant	LC_{50}			LC_{100}		
	24-hrs	48-hrs	96-hrs	24-hrs	48-hrs	96-hrs
Agave	70.83	62.5	3.12	102.08	100	50
Neem	6	3.43	3	8.31	4.86	4.07
Madre de cacao	98.75	120	92	117.5	140	112
Ampalaya	9.75	9.16	9.16	13.5	12.32	12.32
Adelfa	5.87	2.95	2.85	7.87	4	4
Makabuhay	5	4.93	4.7	6	6	6
Sambong	55	20	20.43	80	41.43	40
Lagundi	37.65	21.14	14	50	38.28	20
Calamansi	1.71	2.44	2.38	3	3	3
Jatropa	56.67	44	28.57	81.67	74	50

Ranking of 96-hr LC_{50} values for Nile tilapia

- adelfa (0.083 ml/l)
- makabuhai (0.44 ml/l)
- ampalaya (0.45 ml/l)
- neem (2.57 ml/l)
- lagundi (2.93 ml/l)
- calamansi (3.12 ml/l)
- sambong (5.11 ml/l)
- physic nut (12.8 ml/l)
- agave (30 ml/l)
- madre de cacao (52 ml/l)

Ranking of 96-hr LC_{50} values for mosquito fish

- calamansi (2.38 ml/l)
- adelfa (2.85 ml/l)
- neem (3 ml/l)
- agave (3.12 ml/l)
- makabuhai (4.7 ml/l)
- ampalaya (9.16 ml/l)
- lagundi (14 ml/l)
- sambong (20.43 ml/l)
- physic nut (28.57 ml/l)
- madre de cacao (92 ml/l)

Piscicidal effect as 24-hr LC_{100}

Ranking of 24-hr LC_{100} values for Nile tilapia

- makabuhai (0.82 ml/l)
- adelfa (1.06 ml/l)
- ampalaya (2.59 ml/l)
- calamansi (5 ml/l)
- neem (12.4 ml/l)
- physic nut (26.67 ml/l)
- lagundi (31.5 ml/l)
- agave (74.29 ml/l)
- madre de cacao (90 ml/l)
- sambong (125.71 ml/l)

Ranking of 24-hr LC_{100} values for mosquito fish

- calamansi (3 ml/l)
- makabuhai (6 ml/l)
- adelfa (7.87 ml/l)
- neem (8.31 ml/l)
- ampalaya (13.5 ml/l)
- lagundi (50 ml/l)
- sambong (80 ml/l)
- physic nut (81.67 ml/l)
- agave (102.08 ml/l)
- madre de cacao (117.5 ml/l)

Fish behavior

Test fish stocked in the higher concentrations of the test plant extracts exhibited erratic swimming behavior and rapid opercular movement. Later, the test fishes lose their balance, after which death occurred.

Tolerance

Direct comparison of the lethal concentration values suggests that mosquito fish more frequently had higher LC_{50} and LC_{100} than Nile tilapia. This indicates that mosquito fish is more tolerant to the test plants compared to Nile tilapia.

CONCLUSION

- The results of the study showed that locally available plants in the Philippines have the potential to be used as piscicide which may be an alternative to harmful chemicals that are widely used today to eradicate unwanted fishes in the ponds.
- However, the bulky application rates of the botanical piscicides might be a constraint.
- Commercialization might consider exploring it to develop synthetic compounds from these plants.

Recommendations

- use of other extraction methods
- use of other fish species for bioassay test
- use of other plant parts
- use of other plant species
- actual testing on the use of plant piscicide in earthen ponds

Thank you!
Salamat po!