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Piscicidal Effects of *Terminalia arjuna* Leaf, Bark and Fruit Extract on a Fresh Water Predatory Catfish, *Heteropneustes fossilis*

Suely Akter¹ Hossain M. Zabed¹ Munira Nasiruddin² Xianghui Qi^{1*}

1. School of Food & Biological Engineering, Jiangsu University, Zhenjiang, Jiangsu, 212013, China

2. Department of Zoology, University of Chittagong, Chittagong-4331, Bangladesh

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ABSTRACT

Piscicidal effects of several solvent extracts (distilled water, 50% ethanol, absolute ethanol and 80% methanol) of *Terminalia arjuna* leaves, barks and fruits were studied against a common fresh water predatory fish, *Heteropneustes fossilis* under laboratory conditions in terms of Behavior and mortality of fishes after 24 h. Fishes exposed to plant extracts showed agitating movement with quick surfacing, loss of balance, mucus secretion, and finally died. The LC50 values of distilled water, 50% ethanol, absolute ethanol and 80% methanol extracts were found to be 311.726, 236.141, 183.541, 478.794 ppm for leaves, 117.894, 96.998, 38.990, 304.193 ppm for barks and 1400.033, 949.209, 555.201, 875.158 ppm for fruits, respectively. *Chi-square* values were found to be insignificant at $P < 0.05$ in almost all plant extracts, indicating that observed and expected mortalities did not vary significantly in relation to doses, except 80% methanol extract of barks and absolute ethanol extract of fruits. The *F*-values of treatments were significant at $P < 0.01$, except 80% methanol extract of fruits, in which *F*-values were insignificant in all replicates at $P > 0.01$. Based on LC50 values, order of piscicidal activity followed the pattern, bark > leaf > fruit extracts. Order of piscicidal activity for the extracts was like absolute ethanol > 50% ethanol > distilled water > 80% methanol for leaf and bark, while for fruit extracts, trend was like absolute ethanol > 80% methanol > 50% ethanol > distilled water.

1. Introduction

Predatory fishes can create problems in pisciculture ponds by competing with feed on fry and fingerlings of stocked fish, which ultimately affect fish production, as well as farm economy^[1,2]. In Indian subcontinent and even other parts of world, air breathing predatory fishes, such as *H. fossilis*, *Channa punctatus* cause more serious troubles, because they dwell in the bottom of the pond or remain buried in the mud for a con-

siderable period. Therefore, it becomes difficult to eradicate these fishes from the water body by physical methods such as drag netting. It is possible to dewater in winter and dry the bottom in the sun for several days in shallow ponds. However, dewatering a deep pond is impossible and costly and also commercially it is not acceptable. Rather, several synthetic chemicals have been used for long time to eradicate these unwanted fishes from the water body.

Even though chemicals are target-specific and effec-

*Corresponding Author:

Xianghui Qi,

School of Food & Biological Engineering, Jiangsu University, Zhenjiang, Jiangsu, 212013, China;

Email: qxh@ujs.edu.cn

tive, their impact on the environment is mostly deleterious. The indiscriminate usage of chemical piscicides has given rise to many serious problems including persistency, toxic residues, increased cost of application, environmental pollution, aquatic ecosystem disturbance and hazard from handling^[3,4]. Various form of lives (flora and fauna) are affected from the contamination of freshwater by these synthetic piscicides^[5-8]. For these reasons, application of these compounds is not advisable and an alternative to synthetic chemicals is the use of plant piscicides, which are less expensive, biodegradable and environmentally safer^[9-11].

Plant derivatives are referred to as botanicals and when poisonous to fish are called piscicides^[12]. Such piscicidal plants contain different active ingredients known as rotenone, alkaloids, resin, tannin, saponin, nicotine and diosgenin^[13]. A new ellagitannin (arjunin), four known tannins and two phenolic acids were isolated from *Terminalia arjuna*^[14]. Many plant parts from different families have been applied for catching fish. Some plants contain compounds of various classes that have insecticidal, piscicidal and molluscicidal properties^[15-19]. These plant derivatives are the effective piscicides and not hazardous for the environment^[20-23].

In the present investigation, piscicidal effects of various anatomical parts of *T. arjuna*, namely leaves, barks and fruits extracts were tested against a freshwater predatory catfish, *H. fossilis*.

2. Materials and Methods

2.1 Collection of the Fish Samples

Healthy and live specimens of *H. fossilis* of either sex belonging to a single population (length 9.8-15.4 cm and weight 16-33.47 g) were collected from the local market at Hathazari, Chittagong, Bangladesh. After collection, fishes were kept in plastic containers filled with tap water and brought to the laboratory immediately, and then stocked in a clean glass aquarium bearing 10 L of tap water (Figure 1). Water was renewed every alternative day. Aeration was maintained for oxygen supply in the aquarium. The fishes were regularly fed with rice cake, algae, plankton and oligochaetes. Before experiments, fishes were observed for any disease or abnormality and only healthy fishes were used in the experiments.

2.2 Collection of Plant Materials

Leaves, seed bearing fruits, and barks of *T. arjuna* plant were collected from the University of Chittagong campus near the Faculty of Science, cleaned, dried under the diffused sunlight. Dried plant materials were then ground

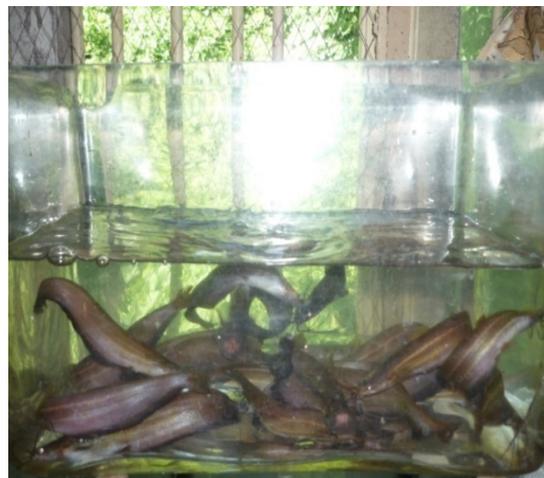


Figure 1. Photograph showing fishes of *H. fossilis* adapted in laboratory aquarium

separately to make fine powder in a power-driven grinder and sieved followed by storing at 4°C for further usage (Figure 2).

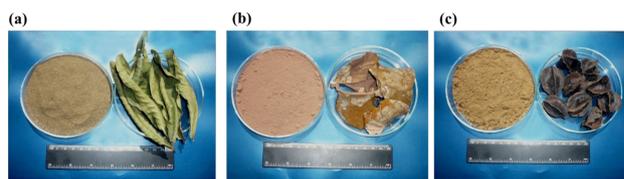


Figure 2. Anatomical parts of dried and ground parts of *Terminalia arjuna*; (a) Leaves (b) barks and (c) fruits

2.3 Extraction of Plant Materials

Four distinct solvents were used for extraction of plant materials, including distilled water, 50% ethanol, absolute ethanol and 80% methanol. Ten grams of powder from each type of plant material was mixed with 100 ml of each solvent separately in a 500 ml conical flask. The flask was shaken vigorously in a magnetic stirrer at room temperature for 3-4 hours to ensure complete extraction of the toxic components. After 3-4 hours the extracted solvent was filtered through fine muslin cloth. The process was repeated twice. Filtered solution obtained after extraction was designated as “Stock solution” and used in all further the experiments. The desired actual concentrations of different test solutions were obtained by appropriate dilution of the stock solution as described elsewhere^[24].

2.4 Exposure of Fish to Extracts

H. fossilis fishes were exposed to extracts of *T. arjuna* plant parts at five different concentrations of each extract under laboratory conditions (Table 1). The tests were carried out in a series of glass aquaria (30 cm×23cm×23cm)

each containing 5L of tap water and calculated amount of extract. In each test, a set of 5 fishes were randomly exposed to each concentration of relevant extract for 24 h. All exposure tests were done in triplicate and conducted under the laboratory conditions. The temperature and pH of water were recorded before and 24 hours after exposure [25]. A control set was maintained with the same number of fishes released in same volume of water in each set of experiments without adding any extract. Behaviors of fishes exposed were recorded in terms of movements and abnormalities during the experiments. The rate of fish mortality was counted in percentage only those fishes which were killed within 24 h of exposure.

Table 1. Concentration of different solvent extracts of *T. arjuna* leaf, bark and fruit applied on *H. fossilis*

Plant parts	Concentration of extracts used (ppm)			
	Distilled water extracts	50% Ethanol extracts	Absolute ethanol extracts	80% methanol extracts
Leaves	1500, 1000, 500, 250 and 100	1000, 500, 250, 100 and 50	750, 500, 250, 100 and 50	1250, 1000, 750, 500 and 250
Barks	400, 300, 200, 100 and 50	300, 200, 100, 50 and 25	200, 100, 50, 25 and 10	800, 600, 400, 200 and 100
Fruits	2000, 1750, 1500, 1250 and 1000	1500, 1250, 1000, 750 and 500	1250, 1000, 750, 500 and 200	1750, 1500, 1250, 750 and 500

2.5 Statistical Analysis

Data obtained from the experiments were analyzed statistically. Dose concentrations were transferred to logarithms. Mortality data were subjected to probit analysis following the methods describe earlier [26]. Probit analysis was used to determine the LC₅₀ values for the extracts. The regression equation was calculated from the empirical probit, working probit, weighting probit, values of which were taken from the standard tables [26]. The value of Chi-square (χ^2) was determined and compared with the tables of statistics for (n-1) degrees of freedom at P<0.05 [27]. Analysis of variance (ANOVA) of percentage mortality of fishes was made to estimate the variation among treatments at P<0.01 [28]. Toxicity values were calculated on the basis of potency, which is a reciprocal of the equitoxic doses. Relative potency of the equitoxic toxicants was obtained by taking the highest LC₅₀ values of a toxicant as unit and comparing with the respective LC₅₀ values of other toxicants. The comparative analysis amongst the different plant extracts were made in terms of LC₅₀ values of each toxicant for the fish sorted out by the probit analysis programme.

3. Results

3.1 Effects of Extracts on Behaviors of *H. fossilis*

Behaviors and movements of fishes significantly fluctuated due to the exposure of fishes to the plant extracts, compared to the Behaviors of control fishes. Behaviors of fishes were noted in terms of movements, physical abnormalities and mortality. In the control, behaviors of fishes were found to be normal throughout the experiment. Fishes moved gently by regular opening of their operculum without generating any straightened barbels. They remained healthy, active and were physically well balanced with soft fins and barbels throughout the experiment. The fins were observed normal and body color was not changed. Moreover, no mortality was recorded in the control group of fishes.

On the other hand, fishes in the treatment groups showed vigorous movement, surfacing and excitement, particularly when they were exposed to the higher doses of extracts. They became slowly inactive, began to lose their balance and settled down at the bottom of the aquaria. The fins and barbels were straight. Fishes died at different time intervals with mucus secretion, and some were floating approximately at 45-90° angles (Figure 3). In contrast, fishes exposed to the lower doses of extracts showed lesser excitements with a fewer of abnormalities. They moved slowly towards the surface and swam around. Fishes tried to jump out from the aquaria but their fins were paralyzed.

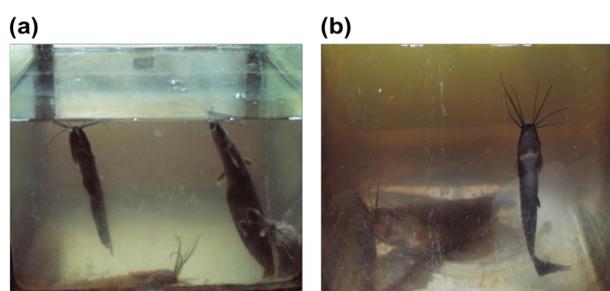


Figure 3. Photograph showing the dead fishes with mucus secretion (a), and floating at 90° angles and lying laterally flattened at the bottom of the aquaria (b)

3.2 Effect of Extracts on the Mortality of *H. Fossilis*

As shown in Table 2, mortality of fishes in the treatment groups significantly varied depending on the solvents used for extracting plant materials, dose of extracts, and anatomical parts of plants, namely leaves, fruits, and barks. The result of the percentage mortality was recorded at an interval of 24 hours of exposure to different concentrations of the extracts. To determine the result of the tox-

icity test for each type of extracts probit analysis method was used.

The highest mortality of *H. fossilis* exposed to distilled water, 50% ethanol, absolute ethanol and 80% methanol extracts of leaves were found to be 86.66%, 86.66%, 93.33% and 93.33%, while the lowest were 26.66%, 20%, 26.66% and 33.33%, respectively. However, maximum mortality was recorded in the fishes exposed to bark extracts in all the four solvent extracts at each of highest doses, estimating to 93.33%, while minimum 33.33% with extract distilled water, 20.0% with both 50% and absolute ethanol and 26.66% with 80% methanol extracts. Similarly, highest 86.66% mortality was found in distilled water, 50% ethanol and 80% methanol extract of fruit and 93.33% for absolute ethanol extract. Minimum mortality exposed to this plant part was 20.0% for 50% ethanol and 26.66% for the rest three solvent extracts.

Regression equations for distilled water, 50% ethanol, absolute ethanol and 80% methanol extracts of leaves, barks and fruits are shown in (Figure 4). Chi-square values, F-value for treatment and for replication, LC₅₀ values and relative potency of different solvent extracts of *T. arjuna* leaf, bark and fruit are shown in (Table 3). Distilled water extracts of different plant parts showed the maximum LC₅₀ values and absolute ethanol extracts gave the lowest value. Maximum LC₅₀ value was found 1400.03 ppm in distilled water extract of fruit after 24 h exposure, while the lowest value was 38.90 ppm and found in absolute ethanol extract of bark. Highest relative potency (35.91) was found in absolute ethanol extract of bark. Based on the LC₅₀ and relative potency, it was observed that absolute ethanol extract of bark was the most toxic

amongst the different extracts having the lowest LC₅₀ and highest relative potency, while the least toxic was the distilled water extract of fruit.

Table 2. Average mortality of *H. fossilis* exposed to various concentrations of *T. arjuna* extracts of leaves, barks and fruits extracts after 24 h (Total number of fishes in each treatment was 15 (3×5=15))

Solvent	Leaf extract		Bark extract		Fruit extract	
	Dose (ppm)	% mortality	Dose (ppm)	% mortality	Dose (ppm)	% mortality
Distilled water	1500	86.66	400	93.33	2000	86.66
	1000	66.66	300	73.33	1750	66.66
	500	60.00	200	53.33	1500	53.33
	250	46.66	100	40.00	1250	33.33
	100	26.66	50	33.33	1000	26.66
50% Ethyl alcohol	1000	86.66	300	93.33	1500	86.66
	500	60.00	200	60.00	1250	53.33
	250	53.33	100	46.66	1000	46.66
	100	26.66	50	26.66	750	40.00
	50	20.00	25	20.00	500	20.00
Absolute ethyl alcohol	750	93.33	200	93.33	1250	93.33
	500	60.00	100	73.33	1000	66.66
	250	53.33	50	53.33	750	53.33
	100	33.33	25	33.33	500	40.00
	50	26.66	10	20.00	200	26.66
80% Methanol	1250	93.33	800	93.33	1750	86.66
	1000	66.66	600	53.33	1500	73.33
	750	60.00	400	46.66	1250	53.33
	500	46.66	200	40.00	750	46.66
	250	33.33	100	26.66	500	26.66

Table 3. Toxicities of different solvent extracts of *T. arjuna* leaf, bark and fruit on *H. fossilis* exposed for 24 hours*

Toxicity parameters	Distilled water extract			50% ethanol extract			Absolute ethanol extract			80% methanol extract		
	Leaf	Bark	Fruit	Leaf	Bark	Fruit	Leaf	Bark	Fruit	Leaf	Bark	Fruit
Dose range (ppm)	100-1500	50-400	1000-2000	50-1000	25-300	500-1500	50-750	10-200	200-1250	250-1250	100-800	500-1750
Chi-square value	5.02	7.82	7.96	5.98	8.22	9.28	7.70	4.96	26.25	8.51	15.52	5.75
F-value (Treatment)	11.18	21.08	11.25	17.16	35.09	19.00	44.28	30.30	17.41	16.43	14.20	6.83
F-value (Replicate)	3.37	2.15	0.17	0.8421	6.90	0.29	7.43	2.15	0.71	6.02	0.00	0.44
LC ₅₀ (ppm)	311.73	117.89	1400.03	236.14	96.99	949.21	183.54	38.90	555.20	478.79	304.19	875.16
Confidence limit (lower)	129.41	61.28	1183.99	134.18	61.97	735.49	95.62	23.25	361.32	252.20	167.93	575.40
Confidence limit (upper)	544.62	177.15	1605.35	418.78	151.24	1222.90	315.83	60.72	749.33	667.70	498.83	1141.86
Relative potency	4.49	11.88	1.0	5.93	14.43	1.48	7.63	35.91	2.52	2.92	4.6	1.60
Ranking	7	3	12	5	2	11	4	1	9	8	6	10

Note: * Degrees of freedom for χ^2 -test is 4; Level of significance for χ^2 -test is $P > 0.05$; Level of significance for F-test (treatment) is $P < 0.01$; Level of significance for F-test (replicate) is $P > 0.01$; Degrees of freedom for F-test (both treatment and replicate) is $V_1 = 4$; $V_2 = 8$.

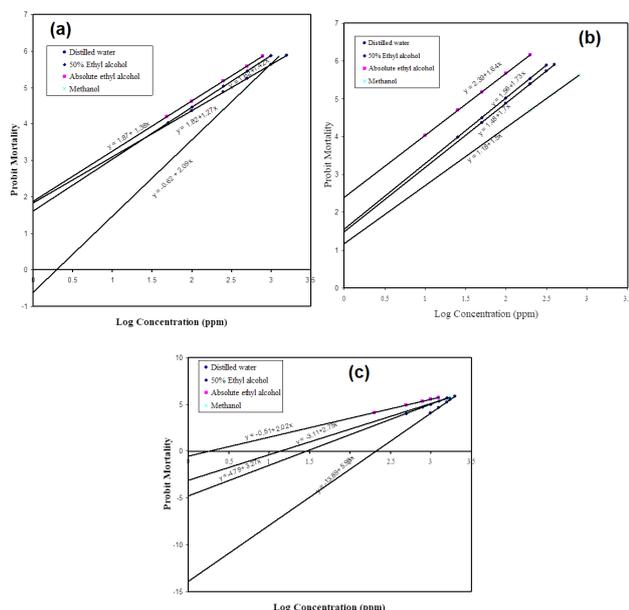


Figure 4. Regression graphs generated for determining LC₅₀ of extracts for distilled water, 50% ethanol, absolute ethanol and 80% methanol extracts of *Terminalia arjuna* leaf (a), bark (b) and fruit (c) against *Heteropneustes fassilis* after 24 hours of exposure

3.3 Effect of the Extracts on Temperature and pH Values of Water

Water quality parameters such as temperature and pH of the experimental set up were monitored using standard methods [25], and data obtained before and after experiments are presented in Table 4. At the beginning of experiment and after 24 hours of exposure, temperature and pH values of water fluctuated significantly. In case of leaf extract, temperature after experiment increased slightly for distilled water extract, while it decreased for 50% ethanol, absolute ethanol and 80% methanol extracts. Likewise, temperatures were found to be decrease for all solvent extracts of barks and fruits.

During experiments, pH of water was 7.2 at initial stage. In leaf extract, pH decreased during experiments due to adding water extracts of leaves but before the experiments it was alkaline. pH of water before the experiments in 50% ethanol, absolute ethanol and 80% methanol extracts were slightly alkaline but after the experiments the extracts became acidic. In bark and fruit extracts, pH decreased slightly after experiments with distilled water, 50% ethanol, absolute ethanol and 80% methanol extracts.

Table 4. Water quality parameters measured before and after the experiment

Plant parts	Solvent	Temperature (°C)		pH	
		Before experiment	After experiment	Before experiment	After experiment
Leaf	Distilled water	20.8±0.19	21.0±0.12	7.3±0.02	7.1±0.03
	50% ethanol	22.9±0.12	21.0±0.11	7.2±0.04	6.8±0.02
	Absolute ethanol	21.7±0.02	20.0±0.03	7.2±0.02	6.9±0.02
	80% methanol	23.4±0.11	21.0±0.12	7.1±0.03	6.9±0.02
Bark	Distilled water	22.3±0.12	20.0±0.06	7.1±0.02	6.9±0.02
	50% ethanol	24.3±0.10	23.0±0.12	6.8±0.02	6.7±0.04
	Absolute ethanol	20.5±0.09	19.0±0.19	7.0±0.02	6.9±0.02
	80% methanol	21.7±0.10	20.0±0.12	6.9±0.02	6.8±0.03
Fruit	Distilled water	19.4±0.16	18.0±0.03	7.2±0.04	6.9±0.02
	50% ethanol	23.2±0.12	21.0±0.11	7.1±0.02	6.7±0.02
	Absolute ethanol	21.9±0.19	20.0±0.12	6.9±0.02	6.8±0.03
	80% methanol	20.8±0.04	19.0±0.16	7.0±0.02	6.9±0.02

4. Discussion

Present study showed that behavior, mortality and activity of *H. fossilis* were affected due to the exposure of fishes to the extracts of *T. arjuna* leaves, barks and fruits. Upon exposure to the extracts, *H. fossilis* showed vigorous movement and were repeatedly rising towards the surface probably for taking air. With erratic movement and having no control on the balance, the exposed species became paralyzed and straightened. They showed mucus secretion and floated at various angles to the surface. Then they slowly settled down to the bottom of the aquaria-water and ultimately died after different intervals. Exposed fish showed signs of respiratory distress and increased opercular movement were observed before death occurred. The findings are in close conformity with those of several previous studies [20,22,23,29]. Researchers have reported their observations about the physical response of the fishes in different toxicants such as agitating swimming was observed in *Salmo gairdneri*, *Peudapocrytes dantatus*, *Gambusia affinis* and *Aphanius mento* [30,31].

Different solvent extracts of *T. arjuna* leaves, barks and fruits caused death of *H. fossilis* in 24 h time period and range of highest mortality was found to be 86.66-93.33% at the dose of 750-1500 ppm. *Azadirachta indica* and *Mesua ferrea* leaf extracts studied and found the mortality rate as 80-90% [23]. In case of bark parts studied it was found that the highest toxicity occurred with 200 ppm (present observation) of absolute ethanol extract of *T. arjuna* and the least toxicity was with 1250 ppm which was supported by the findings with *Acacia auriculaeformis*

absolute ethanol extract^[23]. Of the fruit parts studied it was seen that the most toxic extract was the absolute ethanol extract of *T. chebula*^[32]. Present study with *T. arjuna* is similar to this finding.

The LC₅₀ values of the extracts varied from solvent to solvent and extract to extract. In case of leaf extracts, LC₅₀ values showed trend of toxicity in the order absolute ethanol > 50% ethanol > distilled water > 80% methanol. Previous study with leaf extracts of *Mesua ferrea* also showed the highest LC₅₀ (49.321 ppm) in absolute ethanol extract and while the least toxicity was the distilled water extract of *A. auriculaeformis* with LC₅₀ of 2602.657 ppm^[23]. Whereas in the present investigation, the most toxic extract was absolute ethanol extract of *T. arjuna* with LC₅₀ of 183.541 ppm and the least toxic extract was the 80% methanol extract of *T. arjuna* with LC₅₀ of 478.794 ppm. In case of bark extracts, LC₅₀ values showed the trend of toxicity in the order absolute ethanol > 50% ethanol > distilled water > 80% methanol. Of all the bark extracts studied it was seen that the most toxic extract was absolute ethanol extract of *T. arjuna* with LC₅₀ of 38.990 ppm (present observation), while the least toxicity was the distilled water extract of *A. indica* with LC₅₀ of 1118.041 ppm^[23]. In case of fruit extracts of the present observation the trend of toxicity was absolute ethanol > 80% methanol > 50% ethanol > distilled water. Of all the fruit extracts studied it was found that the highest toxicity was absolute ethanol extract of *S. indicum* with LC₅₀ of 18.62 ppm^[33], while the least toxicity was the distilled water extract of *T. arjuna* with LC₅₀ of 1400.033 ppm (present observation). In comparison with all other extracts tested for LC₅₀, absolute ethanol extracts of *T. arjuna* bark and leaf of the present observation showed high toxicity and fruit extract showed less toxicity. Toxicity of plant parts of *T. arjuna* followed the pattern bark > leaf > fruit. In all the cases, dose response slopes were more or less identical, which is suggestive of a common mechanism for the cause of death. The LC₅₀ is of great significance as biological constants in the data analysis^[34,35].

The relative potency values from the experiment, it was observed that amongst the leaf, bark and fruit extracts on *H. fossilis*, absolute ethanol extract *T. arjuna* of bark which had high mortality with highest relative potency value (35.907 ppm) compared with distilled water extract of *T. arjuna* of fruit which had lowest relative potency value (1.000 ppm). *T. arjuna* leaf showed medium toxicity with medium relative potency value (7.627 ppm) of the absolute ethanol extract. The comparative relative potencies of the parts were observed in the order *T. arjuna* bark > leaf > fruit with relative potency values 35.907 > 7.627 > 1.000 respectively. With 50% ethanol extracts the

comparative relative potencies were in the order *T. arjuna* bark > leaf > fruit with relative potency values 14.433 > 5.928 > 1.474 respectively. With distilled water extract the comparative relative potencies were in the order *T. arjuna* bark > leaf > fruit with relative potency values 11.875 > 4.491 > 1.000 respectively. Whilst with 50% 80% methanol extract the comparative relative potencies were in the order *T. arjuna* bark > leaf > fruit with relative potency values 4.602 > 2.924 > 1.599 respectively. With this data presentation, it makes the deduction that *T. arjuna* bark extracts were the most toxic with comparison to the extracts of the other two (leaf and fruit) plant parts and *T. arjuna* fruit extracts were the least toxic.

Water quality parameters are very important for any water body, as these parameters have direct influence on either health of humans or animals of this environment^[36]. Present study was conducted with tap water in laboratory condition. Slight fluctuation in temperature was noticed during the experiment, which was mainly related with the day temperature. pH of water was found to decrease slightly at the end of the experiment due to release of CO₂ by the test fishes, which is form carbonic acid. This finding was similar with the observation of several investigators^[37,38].

5. Conclusion

Botanical piscicides are believed to be more environmentally friendlier because they are easily biodegraded and leave no residues in the environment. These piscicides are helpful to control the predatory and weed fishes of aquaculture. Present study evaluated the piscicidal potential of three anatomical parts of *T. arjuna* (leaves, barks and fruits) extracts, using various solvents. All extracts showed good piscicidal effects on the predatory catfish of aquaculture system (*H. fossilis*). Among the plant parts, barks showed the highest piscicidal activities against this catfish, followed by leaves and fruits. On the other hand, absolute ethanol extracts showed the highest potential not only for requiring lower concentration, but also for resulting highest mortality of fishes.

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References

- [1] Singh D, Singh A. Piscicidal effect of some common plants of India commonly used in fresh water bodies against target animals. *Chemosphere*, 2002, 49: 45-49.
- [2] Olaifa F E, Hamzat R A, Oyetyoyan O O. Acute toxicity of ethanol extracts of cocoa bean shell on *Sarotherodon galilaeus* juveniles. *J Fish Int*, 2008, 3: 56-60.
- [3] Baskaram P, Palanichamy S, Visalakshi S, et al. Effects of mineral fertilizers on survival of the fish *Oreochromis mossambicus*. *Environ Ecol*, 1989, 7: 463-465.
- [4] Kalavanthy K, Sivakumar A A, Chandran R. 2001. Toxic effect of the pesticide dimethoate on the fish *Sarotherodon mossambicus*. *J Ecol Res Bioconserv*, 2001, 2: 27-32.
- [5] Santharan K R, Thayunmanavum B, Krishnaswamy S. Toxicity of some insecticides to *Daphnia carinata* King and important link in the food chain in fresh ecosystem. *Indian J Ecol*, 1975, 3: 70-73.
- [6] Pant P C, Singh T. Inducement of metabolic dysfunction by carbamate and organophosphorus compounds in a fish, *Puntius conchonioides*. *Pestle Biochem Physiol*, 1983, 20: 294-298.
- [7] Hodson P V. The effect of metal metabolism on uptake, disposition and toxicity in fish. *Aquat Toxicol*, 1988, 11: 3-18.
- [8] Johl M S, Dua A. Elemental lepidological and toxicological studies in *Channa punctatus* (Block) upon exposure to an organochlorine pesticide, endosulfane. *Bull Environ Contam Toxicol*, 1995, 55: 916-921.
- [9] Suely A., Zabed H., Ahmed A.B.A., Mohamad J., Nasiruddin M., Sahu J.N., Ganesan P. Toxicological and hematological effect of *Terminalia arjuna* bark extract on a freshwater catfish, *Heteropneustes fossilis*. *Fish Physiol Biochem*, 2016, 42: 431-444.
- [10] Sudhanshu T, Singh A.. Piscicidal activity of alcoholic extract of *nerium indicum* leaf and their biochemical stress response on fish metabolism. *Afr J Trad Med*, 2004, 1: 15-29.
- [11] Kumar A, Prasad M R, Srivastava K, et al.. Branchial histopathological study of catfish *Heteropneustes fossilis* following exposure to purified neem extract, Azadirachtin. *World J Zool*, 2010, 5: 239-243.
- [12] Burkil H N. The useful plants of west tropical africa, 2nd Ed, Vol 1, Families A-D, Royal Botanical Garden, Kew, 1985.
- [13] Wang S, Huffman J B.. Botanochemicals: Supplements to petrochemicals. *Econ Bot*, 1991, 35: 369-382.
- [14] Fayez E K, Mahmoud I N. A tannin anti-cancer pro-motor from *Terminalia arjuna*. *Phytochem*, 1998, 47: 1567-1568.
- [15] Tripathi S M, Singh D K. Molluscicidal activity of *Punica granatum* bark and *Canna indica* root. *Braz J Med Bio Res*, 2000, 33: 1351-1355.
- [16] Rao I G, Singh A, Singh V K, et al. Effect of single and binary combinations plant-derived molluscicides on different enzyme activities in the nervous tissue of *Achatina fulica*. *J App Toxicol*, 2003, 23: 19-22.
- [17] Prajapati V, Tripathi A. K, Khanuja S P S, et al. Anti-insect screening of medicinal plants from Kukrail forest, Lucknow, India. *Pharmaceut Biol*, 2003, 41: 166-170.
- [18] Neuwinger H D. Plants used for poison fishing in tropical Africa. *Toxicon*, 2004, 44: 417-430.
- [19] Singh A, Singh S K. Molluscicidal evaluation of three common plants from India. *Fitoterapia*, 2005, 76: 747-751.
- [20] Latifa G A, Hamid A, Sharma G.. Piscicidal activity of dry bark of *Diospyros ebenum* (koen) on *H. fossilis* (Bloch) and *Anabas testudineus* (Bloch). *Bang J Life Sci*, 2002, 14: 31-36.
- [21] Latifa G A, Begum M T, Bachar S C. Piscicidal activity of dry barks of *Leucaena leucocephala* (Lam. De Wit) on *Channa punctatus* (Bloch) and *Channa striatus* (Bloch). *Bangladesh J Zool*, 2004, 32: 247-251.
- [22] Nasiruddin M, Azadi M A, Sultana N. Histopathological effects of dry seed extracts of four medicinal plants on *Heteropneustes fossilis* (Bloch). *Chitt Univ J B Sci*, 2007, 2: 113-126.
- [23] Nasiruddin M, Azadi M A, Rahman I A S. Toxicological effect of *Acacia auriculaeformis* (A. Cunn. Ex. Bench) and *Mesua ferrea* (L.) plant parts on *Heteropneustes fossilis* (Bloch). *Bangladesh J Zool*, 2009, 37: 103-112.
- [24] American Public Health Association (APHA). Standard methods for the examination of water and waste water. APHA Press, Washington DC, 1976.
- [25] American Public Health Association (APHA). Standard methods for the examination of water and waste water. APHA Press, Washington DC, 1998.
- [26] Finney D J. Probit analysis, 3rd ed. Cambridge University Press, London, 1971.
- [27] Fisher R A, Yates F. 1963. Statistical tables for biological, agricultural and medicinal research, 6th ed. Oliver and Boyd Ltd, Edinburgh, 1963: 47-50.
- [28] Chowdhury S M. 1969. Introduction to statistical theory, part 11. Parch Made Industries and Press, Lahore, Pakistan, 1969.
- [29] Latifa G A, Begum A. Piscicidal activity of the dry stem of *Euphorbia neriifolia* (Lin. 1753) on *Het-*

- eropneustes fossilis (Bloch) and *Channa punctatus* (Bloch). *Bangladesh J Sci Res*, 1993, 11: 217–225.
- [30] Wedemeyer G. 1970. Stress of anaesthesia with MS-222 and benzocaine in rainbow trout (*Salmo gairdneri*). *J Fish Res Board Can*, 1970, 27: 909-914.
- [31] Sharma A P, Al-Nasiri S K, Bhatt M N. 1978. Toxicity and efficacy of MS-222 on three fishes in Iraq. *Bangladesh J Zool*, 1978, 6: 107-111.
- [32] Chakraborty M. Piscicidal, histopathological and hemolytic effects of fruit, bark and leaf extracts of *Terminalia chebula* (Retz) on *H. fossilis* (Bloch). Dissertation for Master Degree. Department of Zoology, University of Chittagong, 2005.
- [33] Khalil M I. 1984. Study of the piscicidal property of the indigenous *Sapium indicum* fruits (Fam: Euphorbiaceae). Dissertation for Master Degree. Department of Zoology, University of Dhaka, 1984.
- [34] Zbinden G, Flury-Roversi M. 1981. Significance of the LC50 test for the toxicological evaluation of chemical substance. *Arch Toxicol*, 1981, 47: 79-99.
- [35] Wallance-Hayes A. Principles and methods of toxicology. Raven press, New York, 1982.
- [36] Zayed H., Suely A., Faruq G., Sahu J.N.. Water quality assessment of an unusual ritual well in Bangladesh and impact of mass bathing on this quality. *Sci Total Environ*, 2014, 472: 363-369.
- [37] Ameen M, Chowdhury A K A, Khan H R, et al. Insecticidal properties of *D. elliptica* (wall) roots against the larvae of *Culex fatigans* (Diptera: Culicidae). *Dhaka Univ Stud B*, 1983, 31: 1–11.
- [38] Latifa G A, Shafi M, Pervin S I et al. Piscicidal property of dry root of *Tephrosia purpurea* (Pers) on *H. fossilis* (Bloch) and *C. punctatus* (Bloch). *J Asiatic Soc. Bangladesh Sci*, 1988, 14: 48-50.