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Bioassay Study on Fresh Water Fish with PCP and DDT

Byung-soo YANG*

Four-day bioassay method was used in this study to find out toxicity levels of DDT and PCP. From this study it was found that *M. Chrysophekadion* was the most sensitive to DDT with a 96 h-TL_m value of 0.0044 *mg/ℓ* followed by *P. Sutchi* with a 96 h-TL_m value of 0.0056 *mg/ℓ*, and the most resistant was *C. Siamensis* with a 96 h-TL_m value of 0.0133*mg/ℓ*. In the case of PCP, it was also found that *M. Chrysophekadion* and *P. Sutchi* were the first and second most sensitive to PCP with a 96 h-TL_m value of 0.065*mg/ℓ* PCP and 0.125*mg/ℓ* PCP respectively, as in the case of DDT study. *T. Nilotica* was found to be the most resistant to PCP with a 96 h-TL_m value of 0.264*mg/ℓ* PCP. The resistant power of fish to pollution varies with the species of fish and the types of pollutants. In order of decreasing sensitivity of fish to DDT based on TL_m, the following sequence is obtained *M. Chrysophekadion, P. Sutchi, T Nilotica, P. Gonionotus, K. Bicirrhis, L. Bicolor, C. Carpio, R. Heteromorpha, C. Siamensis* while in the case of PCP, *M. Chrysophekadion, P. sutchi, L. Bicolor, K. Bicirrhis, P. Gonionotus* (*R. Heteromorpha, R. Trilineata*), *C. Siamensis, C. Carpio, T. Nilotica.*

Introduction

Waste water are of both organic and inorganic nature and highly harmful to aquatic life. The increasing pollution load and demand for unpolluted water are mutually incompatible. The urgency to control water pollution has led to the establishment of "Water quality criteria" for aquatic life in many countries.

Bioassay methods for the assessment of the toxicity of pollutants to living organisms are recognized as basic tools for the detection, evaluation and abatement of water pollution.

Clearly there is a need to establish both industrial effluent standard and stream standard in order to ensure that harmful concentrations of wastes do not deteriorate biota. Safe levels of various pollutants should be based on the amount of a given toxicant that can be added to a particular waterway at a specific point without harm to aquatic life.

Pollution criteria in aquatic life are commenly measured by doing a bioassay with fish since the fish is not only relatively sensitive to pollutants in aquatic ecosystem but also plays on important role in recreational point of view for human beings. DDT and PCP are often used on bioassay because these are extremely toxic pollutants in environment.

DDT is commonly used as an organic insecticide. It is extremely toxic to fish and poses serious problems of water pollution. This substance is washed from soil and open dump of solid waste by heavy rain into rivers and lakes. DDT is well known as to

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affect the central nervous system of both man and aquatic organisms like fish. Katsuki and Yoshimo (1952) proved it with Japanese eels and Anderson and Peterson (1969) with brook trout. Holland and Butler (1966) reported that sheepshead minnows survived from DDT treatment and produced young fish which are more sensitive to DDT. Fish exposed to DDT seemed less resistant to non-specific stressors, such as exposure to saline water.

Anderson and Peterson (1969) found that sub-lethal dosage of DDT completely prevented the subsequent establishment of a simple conditioned avoidance response in trout. Trout which had learned the response, then treated with DDT, showed a decrease in performance success compared to control fish.

PCP or pentachlorophenols are used in many industrial processes, such as wood processing and germicide industry. Its presence in river water is due to industrial discharges, from runoff or agricultural uses. PCP is generally used in bioassay study rather than phenol because the phenolic compounds are more toxic than pure phenol. Kai and Juhani (1977) found that toxicity of PCP depended much on the pH of the test water. They reported that the toxicity of PCP to crayfish at pH 7.5 was 53 ppm but, when lowering the pH level to 6.5, the toxicity of PCP was increased by a factor of 5.9. Vermeer et al (1974) observed that large fish were killed following the spraying of PCP at a rate of 4*kg/ha* on the rice fields in Surina, South America, for the purposes of snail control. The concentration of PCP in dead fish ranged from 31 to 59/*μg/g*, while snail contained 37*μg/g*. The toxicity of PCP to fish is known to be acute, but the information about this subject is scant.

Most works on bioassay have been done with only single specis of fish under different conditions. These results would give some information in evaluating pollutant toxicity to fish but may not be sufficient enough for setting water pollution criteria because there is possibility that the sensitivity of fishes to pollutants may vary with the different specises of fish. Hence, the objective of this study is to evaluate the sensitivites and/or toxic levels of the diffenent species of fish to different organic pollutants (PCP and DDT) under same condition and to find the most sensitive fish out of 10 species of tested fish chozed according to Ira and Lloyed (1976).

Method of Experiment and Apparatus

The bioassay tests were conducted according to Standard methods (1975). The method consists of preparing various concentrations of organic substances in a selected dilution water, adding the experimental animals, and making observation at selected time intervals. Fig. 1 shows the pattern of bioassay test.

1. Test Animals and Acclimatization

Ten species of the test fish were selected based on Ira and Lloyd (1976) criteria. Names and sizes of the tested fish are grven in Table 1. Six hundred test fish were placed in three 180-ℓ tanks for acclimatization during a period of one week. During the acclimatization period, water in the tanks was renewed twice a week and aerated with compressor-generated air

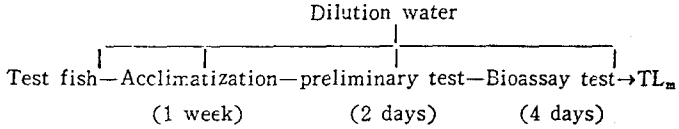

Fig. 1 Schematic diagram of experimental process.

Table 1 Names and sizes of the tested fish

Name of Fish		Weight (*mg*)	Length (*cm*)	Age (week)
Scientific Name	Common Name			
Chanda siamensis	Glass fish	250–300	2.5–3.5	6
Ctprinus carpio	Common carp	250–350	2.5–3.5	4
Kryptopterus bichirris	Glass catfish	200–250	3.0–4.5	7
Labeo bicolor	Redtail blackshark	300–350	2.5–3.5	5
Morulius chrysophekadion	Black shark	250–350	3.0–3.5	5
Pangasius stuchi	Catfish	300–350	3.0–5.0	5
Puntius gonionotus		300–350	3.5–4.5	6
Rasbora heteromorpha	Harlequins	200–250	2.5–3.0	5
Rasbora trilineata	Resbora scissor tail	200–250	3.0–4.0	6
Tilapia nilotica	Tilapia	350–450	4.0–5.5	5

through submerged air-lift filter. The air-lift filter was changed once every two days to avoid clogging of the filter media.

The fish were fed with fish food twice a day but were not fed two days before the bioassay test. The amount of food given was approximately 5% of the total fish weight.

2. Four-Day Bioassay

Duplication test jars for a short-term bioassay were used as shown in Fig. 2. Twelve test jars (6 jars for each run) each containing 10-ℓ of dilution water were placed in a water bath filled with tap water up to the level of dilution water in the jars. One thermostirrer was installed in each bath in order to keep the temperature of dilution water at 29±1°C. Each jar was equipped with an air diffuser stone placed at the bottom of the jar and compressed air was allowed to flow through the dilution water at the rate of 20ℓ/min.

The doses of organic substances were freshly prepared from their stock solutions and then added into each jar according to the concentration range found in the preliminary test. When the temperature and dissolved oxygen in the test solution reach 29±1°C and 7 mg/l respectively, 10 fish were randomly picked by net from the acclimatization tank and placed into each jar and also in the control. Each test was conducted for 96 h without renewing the test solution. The number of dead fish was recorded at the time intervals of 24, 48, 72, and 96 h. Any dead fish was removed immediately from the test solution. No food was given during the test because NH_3 released from the decomposition of the food would become toxic to the test fish. The experiment was repeated when more than two fish died in the control jar.

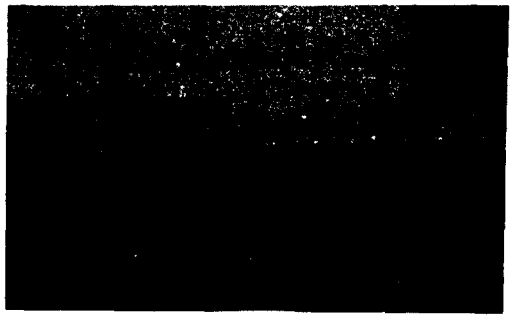

Fig. 2 Duplicate test jar in water bath (side view).

Results and Discussion

1. Mode of Interpretation of Results

The standard technique using TL_m values will be adopted in the interpretation of experimental data. Estimation of TL_m values was made by using semi-log paper and plotting the pollutant concentrations on the log-scale and the percent of survival on the arithmetic scale. A straight line was drawn between two points representing the percent survival at the two successive concentrations that were lethal to more than half and less than half of the test organisms. The concentration at which this line crossed 50% lethality line gave the estimated TL_m value.

The TL_m is the concentration of a toxic material in water that kills 50% of the test animals during a specific time interval. The TL_m was used because the concentration required to effect a response in 50% of the test fish is more reporducible than any other value.

All the calculations of TL_m value are reported in terms of the initial amounts of organic added to the dilution water. Since solubility of these substances is greatly influenced by the characteristics of the dilution water, their concentration to which the fish are exposed are not definitely known.

2. Dilution Water Characteristics

The characteristics of dilution water used for this study are summarized in Table 2.

From these characteristics it appears that the dilution water is suitable for fresh water fish. The pH, alkalinity, turbidity, total solids, COD, BOD, are all within the acceptable range when compared with the water quality criteria for freshwater organisms, as reported by Federal water pollution control administration(1968).

Table 2. Characteristics of dilution water.

Parameter	Concentration Range
pH	7.6–7.8
Hardness, mg/l as $CaCO_3$	80–10
Methyl orange alkalinity	80–110
Turbidity, JTU	15±2
Total solids, mg/l	120–160
COD, mg/l	15–25
BOD_5, mg/l	1–2
Total non-filterable solids, mg/l	5–10
Cd^{++}	less than 1 $\mu g/l$
Cu^{++}	less than 1 $\mu g/l$
DO, mg/l	5.5–6.3

3. Toxicity of Organic Pollutants

In this part of study, attention was given to the sensitivity of fish *via-ã-vis* the organic pollutants (DDT and PCP) without making any attempt to ascertain the specific causes of response of tested animals. TL_m values are reported in terms of the initial amount of DDT and PCP added to the dilution water. Since these salts react with the dilution water, the concentrations of DDT or PCP which the fish were exposed to were not definitely known.

1) DDT Toxicity

A series of DDT concentrations to be tested were found after knowing the critical range. A critical range is the initial dosages between the highest concentration at which all the fish died off and the lowest concentration at which all the fish survived during two days test.

Known quantities of stock solution were mixed with the solution water before the introduction of fish. The experimental environment was not renewed during four consecutive days of test. The mortality rate of some species went up to 70-80% during the first day of experiment, at high concentrations of pollutants.

Average TL_m values obtained are plotted in Fig. 3 and shown in Table 3.

Table 3 TL_m values of DDT in *mg/ℓ*

Tested fish	24h-TL_m	48h-TL_m	72h-TL_m	96h-TL_m
Chanda siamensis	0.0259	0.0183	0.0148	0.0133
Cyprinus carpio	0.0160	0.0138	0.0123	0.0106
Kryptopterus bicirrhis	0.0120	0.0101	0.0094	0.0090
Labeo bicolor	0.0120	0.0100	0.0100	0.0092
Morutius chrysophekadion	0.0061	0.0054	0.0050	0.0044
Pangasius sutchi	0.0100	0.0071	0.0058	0.0056
Puntius gonionotus	0.0190	0.0150	0.0100	0.0088
Rasbora heteromorpha	0.0187	0.0148	0.0126	0.0125
Tilapia nilotica	0.0113	0.0080	0.0067	0.0060

From this table, it can be seen that *M. Chrysophekadion* was the most sensitive fish to DDT with a 96 h-TL_m value of 0.0044*mg/ℓ* DDT followed by *P. Sutchi* with a 96 h-TL_m value of 0.0056*mg/ℓ* DDT, and the most resistant was *C. Siamensis* with a 96 h-TL_m value of 0.0133 *mg/ℓ* DDT. It can be remarked that an insignificant amount of DDT is harmful to fish, at all levels of TL_m. Somtavila (1977) found that a 96 h-TL_m value of *T. Mossambica* was 0.285*mg/ℓ* DDT which is much higher than what was found in this study. This may be due to the size of the fish and also the species.

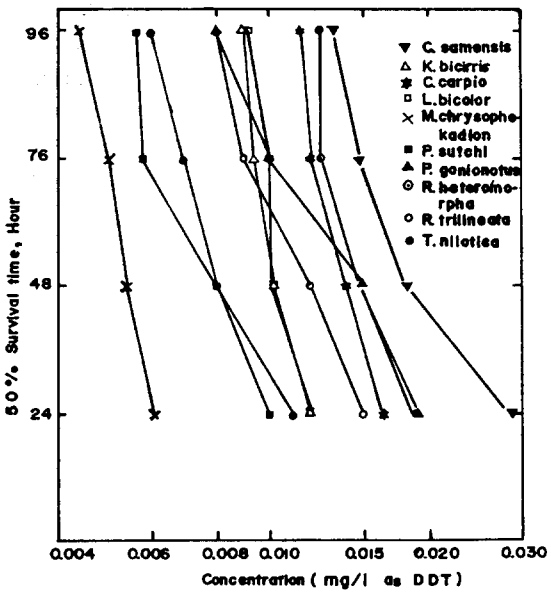

Fig. 3 Median survival curve of different species of fish.

In order of decreasing sensitivity of fish to DDT based on TL_m values, the following sequence is obtained; *M. Chrysophekadion* >*P. Sutchi*> *T. Nilotica* >*P. Honionotus* >*K. Bicirrhis* >*L. Bicolor* >*C. Carpio*> *R. Heteromorpha* >*C. Siamensis*.

2) PCP Toxicity

In PCP, the same mortality rate was found as in the case of DDT study. Fish increased their swimming activity, came up to water surface and then died. Average TL_m values are plotted in Fig. 4 and summarized in Table 4.

Table 4 TL_m Values of PCP in mg/l

Tested fish	23 h-TL_m	48 h-TL_m	72 h-TL_m	96 h-TL_m
Chanda siamensis	0.301	0.273	0.245	0.203
Cyprinus carpio	0.570	0.405	0.300	0.243
Kryptopterus bicirrhis	0.270	0.215	0.200	0.170
Labeo bicolor	0.197	0.180	0.165	0.154
Morulius chrysophekadion	0.098	0.073	0.068	0.065
Pangasius sutchi	0.178	0.163	0.153	0.125
Puntius gonionotus	0.283	0.228	0.211	0.180
Rasbora heteromorpha	0.260	0.226	0.180	0.180
Rasbora trilineata	0.283	0.228	0.211	0.180
Tilapia nilotica	0.395	0.356	0.308	0.264

It was found that *M. Chrysophekadion* and *P. Sutchi* were the first and second most sensitive to PCP with a 96 h-TL_m value of 0.065 mg/l PCP and 0.125 mg/l PCP respectively, as in the case of DDT study.

T. Nilotica with a 96 h-TL_m value of 0.265 mg/l PCP was found to be the most resistant to PCP in this experiment. Ira and Lloyd(1976)

conducted a study with goldfish and obtained a TL_m value of 0.21 mg/1 PCP which is nearly the same as the value obtained for the most resistant fish in this study.

TL_m value of PCP depends very much on pH value as reported by Kai and Juhani (1977). When pH changed from 7.5 to 6.5 toxicity of PCP will increase about sixfold.

DDT toxicity was found to have a pronounced impact and operate in a narrower range than PCP. The toxicity of DDT range from 0.0044mg/1 to 0.0259 mg/1 and PCP from 0.065 mg/1 to 0.570 mg/1. *M. Chrysophekadion* and *P. Sutchi* were found to be the most sensitive to both DDT and PCP.

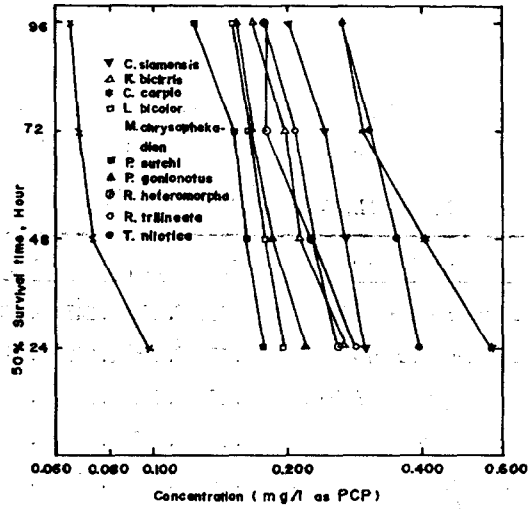

Fig. 4 Median survival curve of different species of fish.

It could be noticed that the degrees of sensitivities of fishes to pollutants somewhat vary with the type of the species of fish and the chareteristics of pollutants. These results well agree with Yang's report (1978). However, the general trends in the sensitivities, the fish resistant to the specific pollutant is resistant to other pollutants, with exception of dissolved oxygen.

Conclusion

Based on the results of this study, the following conclusion could be made.

1. The toxicity of DDT to fresh water fish is considerably higher than that of PCP.
2. In addition to influence of time and physical and chemical factors on the toxicity, the comparative toxicity of organic pollutants (PCP and DDT) varies with the test species.
3. *M. Chrysophekadion* was the most sensitive fish to DDT with a 96 h-TL_m value of 0.004 *mg/ℓ* followed by *P. Sutchi* with a 96 h-TL_m value of 0.0056*mg/ℓ*, and the most resistant was *C. Siamensis* with a 96h-TL_m value of 0.0133*mg/ℓ*.
4. *M. Chrysophekadion* and *P. Sutchi* were the first and second sensitive fish to PCP with a 96h-TL_m value of 0.065*mg/ℓ* PCP and 0.125*mg/ℓ* PCP, respectively. *T. Nilotica* is found to be the most resistant to PCP with a 96h-TL_m value of 0.264*mg/ℓ* PCP.

References

Anderson J.M. and Peterson M.R. (1969): DDT Sub-lethal effects on brook trout nervous system. Science, N.Y. 164 (38-78) p. 440-441

Holland H.T., Coppage, D.L. and Butler, P.A. (1966): Increased sensitivity of pesticides in sheepshead minnows, Trans, Am. Fish. Soc. 95, p. 110-112

Ira, A. and Lloyd L. and Smith Jr. (1976): Father minnows and goldfish as standard fish in bioassays and their reaction to potential reference toxicants, J. Fish. Rec. Board. Can. Vol 33, 209-214

Kai, K. and Juhani S. (1977): Toxicity of penta chlorophenol and 2, 3, 6 trichlorophenol to the cray fish env. Pollut. (12), Science Publishers Ltd, England

Katsuki Y. and S. Yoshino (1952): Response of the single lateral-line nerve fiber to the linearly rising current stimulating the endorgan, Japan. J. Physiol. 2, 219-239

Somtavila S. (1976): The effect of temperature

on the toxicity of DDT and eldrin to tilapia mosambica special report submitted to marine science department, Chulalongkorn University, Bangkok, Thailand

Standard methods for the examination of water and wastewater (1975): APWA, AWWA and WPCF, 14th Ed., American public health association Inc., New York.

Vermur K. et al (1974): Pesticide effects on fishes and birds in rice fields of surinam, south america environ poll. 7, 217

Water pollution control administration federal (1968): Water quality criteria, report of the national technical advisory committee to the secretary interior, April 1, 1968, Washington, D. C. Federal water pollution control administration.

Yang, byungsoo(1978): Bioassay studies on Chao Phya River Fish. Master thesis No. 1409, AIT, Bangkok, Thailand, p.30-37

PCP와 DDT의 독성이 담수어에 미치는 영향

梁 秉 洙

하천 오염에서 문제시되고 있는 유기오염 물질인 PCP 와 DDT의 환경기준을 평가하고 담수어 어종간의 독성에 대한 저항성을 연구하였다.

각 어종에 대하여 24, 48, 72 및 96시간의 TL_m 값을 산출하였던 바 오염물질에 대한 저항성은 오염물질의 성상에 따라 상이한 양상을 보여주었다.

실험한 10종의 담수어중 PCP 및 DDT에 저항성이 약한 어종은 *M. Chrysophekadicn* 으로 96 h-TL_m 값은 0.065 *mg/ℓ* 과 0.0044 *mg/ℓ* 으로 각각 나타났으며 PCP에 저항성이 강한 어종은 *T. nilotica* 로서 96 h-TL_m 값은 0.264 *mg/ℓ* 이었으나 DDT에 대해서는 *C. Siamensis* 로서 96 h-TL_m 값은 0.0133 *mg/ℓ* 로 평가 되었다.