

Interaction of Zinc, Cadmium, Copper, Endosulphan and Dimethoate Pesticides on Fresh Water Fish Channa Punctatus

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Abstract

The aim of the present study to find out the interaction of toxicants and pesticides on fresh water fish Channa Punctatus. We know that chemical mixtures are characteristic of life. The nature of the mixtures varies widely according to the specific chemical present and their concentration with in living system, which is controlled within relatively close limits. 90 healthy fishes divided into 6 groups of 15 each were exposed to LC50 of zinc (18.62mg/l), copper (0.56 mg/l), cadmium (11.8 mg/l) Endosulphan (3 m/l) and Dimethoate (14.84 mg/l) with selected combination separately. The sixth group of 15 fishes was kept in metal free water served as control for each experimental group. 75 fishes were divided into 5 groups of 15 fishes each and were exposed to LC50of zinc (18.62 mg/l), copper (0.56 mg/l), cadmium (11.8 mg/l) Endosulphan (3 m/l) and Dimethoate (14.84 mg/l) with selected combination separately. In this study results shows that in combination of cadmium and copper separately with Endosulfan, no mortality of fish was observed, showing antagonistic effect, while the presence of these two metals together with pesticides became very toxic (synergistic). Interaction between metals was also very toxic to fish except zinc+ cadmium combination where toxicity was reduced. Pesticides combination was found more than additive showing 50% mortality with in 42 hr instead of 48 hr, but even up to 96 hr the combination could not cause 100% mortality which suggests less than additive effect. In this study it is recommended that farmer avoid fish culture near to factories and places where pesticides used by farmer.

Keywords: Interaction; Mortality; Zinc; Cadmium; Copper; Pesticide; Endosulfan; Dimethoate; Fish; Channa Punctatus

Introduction

Man is responsible for drastically altering the natural resources through his intelligence. He has tried to improve the quality of life through industrial, agricultural and urban development and thus control his own evolutionary process. It is estimated that about 800-1000 tons of pollutants are being ejected into the atmosphere

each day by all the big cities of our country. Agriculturists in their bid to improve the quality and quantity of food products have been using fertilizers and chemical pesticides indiscriminately. Such indiscriminate use may result in nutritional imbalance of the soil, which ultimately will reflect in men and animals in that area. Therefore, increasing industrialization and developmental programs lead to continued addition of pollutants to the

environment. Environmental pollution by heavy metals has increased in recent years due to extensive use of heavy metals in agricultural, chemical and industrial processes and has become a threat to living organisms. Pesticides are used extensively throughout the world in agricultural and public health programs. The contribution of pesticides to agriculture and public health has been substantial, but their injudicious use has created serious problems.

Contamination of soil and water is caused often by chemical mixtures than by single pollutants. Common examples of mixed chemical pollutants include gasoline; crude oil and land fill leachates. Although these mixtures consist essentially of organic compounds, other pollutant mixtures may include both organic and inorganic chemicals such as B heavy metals. Even in cases where a site is contaminated by only one pollutant, the generation of degradation products, such as humic acid changes the chemical makeup of the soil and water at the site so that they are better described as mixtures. Therefore, the toxicity of mixtures or combinations acquires great significance.

The interactions among trace metals and pesticides possibly due to their competition for a common binding site are very well known and the most studied interactions are in the following groups: copper - molybdenum; copper - iron - zinc; cadmium - zinc - copper - iron; selenium - mercury - arsenic - cadmium; manganese - selenium; cadmium - Endosulfan; cadmium - Dimethoate; quinalphos - cypermethrin. The interaction between cadmium and zinc results in removal of cadmium from active sites when zinc is present in excess but when cadmium is in excess, it displaces zinc from its metallo enzymes and produces toxicity. Genus *Valgum* syndrome in Andhra Pradesh is a well known example of toxic manifestation due to metal interaction. Excessive intake of molybdenum along with fluoride led to depletion of the body reserves of the essential metal copper.

No interaction or "independent joint action" occurs when the toxicants have similar mode of action. But when they act independently, the resulting toxicity of the mixture is the sum of the toxicities of individual toxicants present. Under 'interactive action' the effect may be either synergistic (more than additive) or antagonistic (less than additive). These interactions occur when one toxicant alters the toxicity of other toxicants present, which may be due to interaction between the toxicants involved or between the individual toxicant and the physiological systems of the organisms. The former case may involve

the alteration of chemical structure or chemical speciation, while the latter may involve alteration in the distribution, excretion or transformation of one toxicant by the other.

Synergism is caused by various biological activities of chemicals depending mainly on whether the agents have the same or different sites of primary action and whether or not the presence of one chemical influences the amount of other agent reaching their sites of action (interactive or non interactive). Combined treatment may result in simple addition. The compounds that belong to the same class and have a common target organ show linear additive effects, whereas compounds with the same target organ but different metabolic pathways may produce less than additive response. Synergistic mechanisms with simultaneous administration may involve one or more possibilities:

- a) Increase in uptake
- b) Enhanced metabolic activation
- c) Inhibition of DNA repair processes
- d) Increased proliferation of cells with DNA damage

Geyer, et al. [1] provides an explanation for a 40-fold difference in the acute toxicity (LC50) of gamma hexachloro cyclohexane (gamma-HCH, Lindane) in 14 different fish species, based on well recognized principles of toxicokinetics and toxic dynamics in combination with a compilation of data from the literature and some original data. The 48h median lethal concentration (48-h LC50) of gamma-HCH in 14 fish species, belonging to 6 families, range from 22 to 900 micrograms/l. A significant positive linear relationship was found between lipid content (% of wet weight) and the 48-h LC50 of gamma-HCH in these fish species, revealing that the toxicity of gamma-HCH in various fish species is decreasing with increasing total lipid content. If median lethal concentrations are normalized for 1% lipid content, then the range of 48-h LC50s is reduced to between 18 and 32 micrograms/l. It is concluded that lipids of aquatic organisms can serve (among other functions) as a protective storage site against the toxic effects of gamma-HCH and, possibly, of other lipophilic, persistent organic chemicals which are bio concentrated in body lipids. Therefore, in organisms with higher lipid content, a smaller fraction of a lipophilic chemical will reach target organs (liver, lung, central and peripheral nerves, etc.) to cause adverse effects. Results suggest that this correlation can be used to extrapolate the acute toxicity (48-h LC50) of gamma-HCH to other fish species if their lipid content is known. Toxicity of Fenvalerate a synthetic pyrethroid to some fresh water fish species was evaluated by Satyavardhan [2]. The toxicity is determined by using

both static and continuous flow through systems and also noticed some specific behavioral characteristics during the experimentation periods in different time intervals. The LC 50 values were calculated for 24h, 48h, 72h and 96h respectively. When the fish were exposed to sub lethal and lethal concentration of Fenvalerate, technical grade and 20% EC., they were migrated to the bottom of the test chamber immediately. This is because of the toxic stress. Their Schooling behavior was totally disturbed and they are swimming independently and this was followed by irregular, erratic and dangling movements with the imbalanced swimming activity. The swimming behavior was in a cork screw pattern and rotating along horizontal axis and followed by "S" jerks, sudden, rapid and non-directed sport of forward movement likely to be busted swimming. The fish were exhibited peculiar behavior that is the fish were trying to leap out from the test chamber which can be viewed as escape phenomenon. Respiratory disruption was observed due to cough and yawning this is because of toxic stress. They often barrel rolled or spiraled at regular intervals and engulfed the air through mouth before respiration ceased. A change in color of the gill lamellae from reddish to light brown with coagulation of excess mucous on the gill lamellae was observed. The symptoms of Fenvalerate poisoning in the fish include loss of schooling behavior, swimming near the upper surface, hyper activity, zigzag movement, loss of buoyancy, elevated cough, increased gill mucous secretion, flaring of the gill arches, head shaking and restlessness before death.

Materials and Methods

Healthy living specimens of the fresh water teleost fish *Channa Punctatus* were collected from the local ponds. Toxicants used in the experiments are zinc, cadmium, copper, Endosulfan and Dimethoate.

Healthy living specimen of freshwater teleost fish *Channa Punctatus* were collected from the local ponds and acclimatized to the laboratory conditions for 10 days in batches of 10 each were transferred to 4 glass aquarium and exposed for 96 hour to 3 different toxicants and 2 pesticides at their various concentrations (Zinc 18.62 mg/l, Cadmium 11.8mg/l, Copper 0.56 mg/l, Endosulfan 3.1ug and Dimethoate 14.84). After 96 hours exposure the mortality of the fish at each concentration was recorded. The fish were considered dead when they did not respond on being probed with a glass rod and respiration ceased. Table 1 lists the combinations of toxicants and their concentration used in toxicity studies. The sequence of administration of metals and pesticides

was as follows: each aquarium containing 10 fish in tap water received first a corresponding concentration of the toxicant listed in Table 1 as the additive. Within the next 20 mm. a corresponding volume of solution listed as the 'toxicant' was added.

Toxicant	LC50 mg/l	Additive	lc50 mg/l
Zinc	18.62	Cu	0.56
		Cd	
		Endosulfan	0.56
		Dimethoate	11.8
		Cu+Cd	3.1 ug/l
		Cu+Endosulfan	14.84
		Cu+Dimethoate	
		Cd+Endosulfan	
		Cd+Dimethoate	
		Cd+Cu+Endosulfan	
		Cd+Cu+Dimethoate	
		Cd+Cu+Endosulfan+ Dimethoate	
Cd	11.8	Cu	
		Endosulfan	
		Dimethoate	
		Cu+Endosulfan	
Cu	0.56	Endosulfan	
		Dimethoate	
Endosulfan	3.1 ug	Dimethoate	
Dimethoate	14.84	Cd+Cu	

Table 1: Volume of solution listed as the 'toxicant'.

All experiments were run simultaneously for 96 hr and dead fishes were counted after every 8 hr intervals. A control experiment was also run for 96 hr in toxicant free tap water. All experiments were repeated three times at temperature varying between 20-25°C. From these experiments six combinations, zinc+cadmium, zinc+copper, cadmium+Endosulfan, zinc+Endosulfan, zinc+cadmium+ copper+Endosulfan, zinc+cadmium+copper+Dimethoate were selected.

Exposure of Test Fish

Healthy living specimens of the fresh water teleost fish *Channa Punctatus* collected from the local ponds of Rohtak or purchased from fish market were quickly transported to laboratory. The fish were maintained in glass aquaria and fed twice daily with pelleted diet (prawn powder, fish powder, and minced liver in 2:2:1 ratio). The water in aquaria was continuously aerated in order to maintain the dissolved oxygen concentration above 7 ppm.

Individual Exposure

90 fishes divided into 6 groups of 15 each were exposed to LC50 of zinc (18.62 mg/l), copper (0.56 mg/l), cadmium (11.8 mg/l) Endosulfan (3 m/l) and Dimethoate (14.84 mg/l) with above selected combination separately. The sixth group of 15 fishes was kept in metal free water served as control for each experimental group.

Group Exposure

75 fishes were divided into 5 groups of 15 fishes each and were exposed to LC50 of zinc (18.62 mg/l), copper (0.56 mg/l), cadmium (11.8 mg/l) Endosulfan (3 m/l) and Dimethoate (14.84 mg/l) with above selected combination separately.

Results and Discussion

The volume of information on the metal ion requirements by living organism is rapidly increasing.

New elements are being constantly included in the lists of requirements for bio elements as well as their toxic effects. Gradually more attention is being paid to interactions of metal ions and pesticides within the biological systems. These, if known and properly applied, may be useful tools in the fight against certain diseases and in detoxification in fishes. The interaction between chemicals in the natural environment does not act alone but in combination. Therefore, it is absolutely essential for a better ecotoxicological approach to investigate their interaction. The present study deals with the percentage mortality after 96 hr of exposure period due to the addition of LC50 of heavy metals and pesticides in different groups as shown in Table 1.

Percentage Mortality

Results of the investigation, with interaction of heavy metals and Pesticides are presented in Table 2.

Toxicant	LC50 mg/l	Additive	lc50 mg/l	percent mortality						
				18hr	24hr	42hr	48hr	66hr	96hr	
Zinc	18.62	Cu	0.56		25	50				
		Cd				25	50			
		Endosulfan	0.56	No mortality observed						
		Dimethoate	11.8	25				50	75	
		Cu+Cd	3.1 ug/l	50	100					
		Cu+Endosulfan	14.84		50	100				
		Cu+Dimethoate		75	100					
		Cd+Endosulfan				50	100			
		Cd+Dimethoate		50	75	100				
		Cd+Cu+Endosulfan		100						
Cd	11.8	Cd+Cu+Dimethoate		100						
		Cu		25	75	100				
		Endosulfan		No mortality observed						
		Dimethoate				25	100			
		Cu+Endosulfan		25		75	100			
		Cu	0.56				25			
		Endosulfan	3.1 ug				50	75	100	
		Dimethoate	14.84				50		75	
		Dimethoate	14.84	Cd+Cu		50	75	100		

Table 2: Variation in percent mortality of *Channa Punctatus* in combination of metals and pesticides after 96 hr.

It has been observed that in some pairs of heavy metals and pesticides, the effects were synergistic or antagonistic or no interaction was observed among them. In pairs of Zinc+cadmium, zinc + Dimethoate and copper+

Endosulfan antagonistic effect was observed. No interaction was observed in zinc+copper combination. Synergistic effect was noted in pairs of cadmium+copper, cadmium+Dimethoate, copper+Dimethoate,

Endosulfan+Dimethoate, zinc+cadmium+Endosulfan and zinc+copper + Endosulfan strong antagonism is noticed between copper and Endosulfan as only 25% mortality was observed after 66 hr. No mortality occurred in the combination of zinc+Endosulfan and cadmium+Endosulfan even after 96 hr. A powerful synergism of 100% mortality after 18 hr was noted in the mixtures of zinc+cadmium+copper+Endosulfan, zinc+cadmium+copper+Dimethoate and zinc+cadmium+copper+i+Endosulfan+Dimethoate. In pairs of zinc+copper, zinc +cadmium, zinc+ Dimethoate and Endosulfan + Dimethoate mortality never reached 100% after 96 hr exposure period. There was only one combination (zinc+cadmium+Endosulfan) in which 100% mortality was observed after 48 hr. There was 100% mortality in the mixtures of zinc+copper+Endosulfan, zinc+cadmium+Dimethoate, cadmium+copper and cadmium+copper+ Dimethoate after 42 hr, while zinc+copper+cadmium and zinc+copper + Dimethoate produced 100% mortality with in 24hr. Present results supported by the studies of Brown, et al. [3]. Results of their experiments ranged from antagonistic to synergistic responses. Though the overall trend was for zinc to reduce toxicities of other metals, toxicant mixture studies have also been Undertaken in fish exposed to such chemical groups as PCBs [4], pesticides [5,6], the importance of studying the effects of metals combination has become an area of interest. It has been demonstrated that the responses of organisms to metal combination can be different from those on the solitary action of metals. For example, two or more metals may act antagonistically where one metal may ameliorate the toxicity of the other metal [7]. Conversely metals may act synergistically where the toxic response is greater than the sum of individual toxicities. Shivraj, et al. [8] studied the toxicity of cadmium and copper to a freshwater fish, *Puntius arulius*. The effects of pesticides on blood characteristics and histological changes in erythrocytes of the fish species *Cyprinus carpio* and *Puntius ticto* were studied by Satyanarayana, et al. [9-18]. The fishes were exposed to sub lethal concentrations of different chlorinated pesticides namely aldrin, dieldrin, DDT, BHC and chlordane for 10, 20 and 30 d in continuous flow-through test. Results showed an increase in hemoglobin content of both *Cyprinus carpio* and *Puntius ticto* in case of aldrin and dieldrin.

Conclusion

From these observations it is clear that in combination of cadmium and copper separately with Endosulfan, no mortality of fish was observed, showing antagonistic

effect, while the presence of these two metals together with pesticides became very toxic (synergistic). Interaction between metals was also very toxic to fish except zinc+ cadmium combination where toxicity was reduced. Pesticides combination was found more than additive showing 50% mortality with in 42 hr instead of 48 hr, but even up to 96 hr the combination could not cause 100% mortality which suggests less than additive effect.

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