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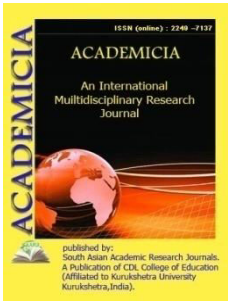

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PESTICIDES AND TOXICITY ON AQUATIC LARVAE: A STUDY ON LOKTAK LAKE CATCHMENT AREA

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ABSTRACT

This paper attempts to study on Pesticides and Toxicity on aquatic larvae of Pantalaflavecens found in the Loktak Lake Catchment Area, Manipur. It is also a part of the study on pesticide pollution in the lakes and their effect on non-target organisms. The time at which the pesticides are added into the water will be noted along with the room and water temperatures. The larvae were observed continuously for their response to the chemical. Whenever a larva dies, it was immediately removed. Therefore, a research work with an objective to develop ways and means of fighting harmful insects, pests and weeds without effecting non-target species and ecosystem is necessary.

KEYWORD: *Pesticide, Chlorpyrifos, Monochrotophos, Toxicity, Endosulphan*

INTRODUCTION

Manipur is one of the eight states of the North East India. There are a number of shallow lakes including the Loktak Lake in Bishnupur District. For the people living around the lake the Oksoilake provides water fish cash crops, vegetables, etc., throughout the year. During winter season, the dried up shallow peripheral areas are used for cultivation of the vegetable crops including cabbage, cauliflower, mustard, etc. while these shallow areas are converted in to paddy field. In Manipur, the pesticides are used by the farmers in large scale in agriculture and horticulture for the controlling of harmful insects and pests and they produced increase crop yield and quality. But on the other hand (as a result) with the increasing use of insecticides and pesticides, it causes environmental pollution, disturbance in the ecosystem and it leads to extinction of useful non-target species. It also causes many diseases to human beings through food chain system.

Description of study site

There are two seasons, the winter season is form October to March and the summer season is from April to September. The summer season is actually the rainy season in this part of the state. The state has been divided into 16 administrative districts. They are the Imphal West, the Imphal East, the Bishnupur, the Thoubal, the Kakching, the Jiribam, the Ukhrul, the Senapati, the Tamenglong, the Chandel the Churchandpur, the Pherzawl, the Kamjong, The Noney, the Kangpokpi, and the Tengnoupal.The mean annual rainfall is 117.8 cm, the lowest rainfall occurs during January 0.36 cm, and maximum rainfall during June is 23.2 cm. The highest average rainfall occurs in Tamenglong District and is about 350 cm. The vegetations vary from tropical rain forest to coniferous. Under climate and adapted conditions of high temperature, heavy rainfall, dissected topography and thin soil, the vegetation has almost gregarious growth.In Bishnupur district there are a number of shallow lakes including the Loktaklake. For the people living around the lake, the Loktaklake provides water, fish, cash crops, vegetables etc. throughout the year. During winter season, the dried-up shallow peripheral areas are used for cultivation of vegetable crops including cabbage, cauliflower, mustard etc. while these shallow areas are converted into paddy field. Loktaklake is considered as the lifeline of the people of Manipur due to its importance in their socio-economic and cultural life. It is the largest natural freshwater lake in the northeastern region and plays an important role in providing ecological and economic security to the region (Linthoi, 2013).

For enhancing the crop production in and around the lake, people use different types of pesticides that are applied without much precautions. This leads to entry of an excessive amount of these potentially harmful substances into the lake water, leading to deleterious effects on the whole lake ecosystem in general, and the organisms present in the lake water in particular. This necessitates investigations into the eco-toxicology of these substances.

METHODOLOGY

- **Collection of Test Animals:**

The aquatic larvae of different species dragonflies belonging to the order odonata and suborder Anisoptera were collected with the help of nets from Loktaklake in Bishnupur District, Manipur for use as test animals.

- **Identification:**

The identification of odonata larvae was done on Kumar (1973) and Needham (1954).

- **Preparation of Stock Solutions:**

The stock solutions were prepared by mixing 1 ml of insecticide will 1000 ml of water. This stock solution contains 0.001 ml of pesticide per 1 ml. By adding the required quantities of the above solution to 100 ml of water, different concentrations in ppm (parts per million) of the pesticides are obtained. The solution would be thoroughly mixed and will keep in a clean beaker of 1500 ml capacity with graduated mark on the beaker.

REVIEW OF LITERATURE

A number of investigations have been carried out on pesticide and Toxicity of commonly used pesticides on target as well as non-target organisms. Literature review is useful to critically

summarise the current knowledge in the area under investigation, identifying any strengths and weaknesses in previous work, helping to identify them in research and thus eliminate the potential weaknesses and bringing out the potential strengths. It provides an up-to-date understanding of the subject and its significance identifies the methods used in previous research on the topic, helps to work out how to answer the questions - and indeed, what questions need to be asked, provides comparisons for research findings (Sairam, 2016). Vijaya (2010) evaluated the toxicity of the three different groups' organophosphate pesticides and monocrotophos [Orthophosphate group] and to find out which group of the selected organophosphate was more toxic to the test species Channapunctatus.

Kumar and Chapman (1998) studied the toxicity of profenofos to eastern rainbow fish, Melanotaeniaduboulayi. Toxicity of malathion and carbaryl pesticides was determined in freshwater fish Colisafasciatus by Singh *et al.,* (2004). Chlorpyrifos toxicity was tested in Indian Carp. CatlaCatla, Labeorohita, and Cirrhinusmrigala by Tilak*et al.*, (2004).Acute toxicity of synthetic pyrethroids to Indian Major Carp.Catlacatla was studied by Tandon*et al.,* (2005). Ural *et al.,* (2005) studied the acute toxicity of Dichlorvos (OP) on fingerlings of European Catfish, Silurusglanis. Saha and Kaviraj (2008) studied the acute toxicity of synthetic pyrethroidcypemethrin in freshwater organisms. Koprucu et al. (2006) studied acute toxicity of Organophosphorus pesticide diazinon and its effects on behaviour and some haematological parameters were studied in fingerlings of European cat fish. Agrahari*et al.,* (2006) studied the effect of monocrotophos an erythropoietic activity and haematological parameters of the freshwater fish Channapunctatus. HiiYii*et al.,* (2007) also analysed acute toxicity of organochlorine insecticide endosulphon and its effect on behaviour and some haematological parameters of Asian swamp eel. Venkata (2013) said, in Indian context, usage of organophosphates has also gradually been increasing with consistent decline in application of organochlorines and currently takes share of 27% of total sales of pesticides in India. During the last 60 years, approximately 150 different OP chemicals have been used to protect crops, live-stock, and human health.

Jha and Singh (1984) have studied the toxicity of seven different insecticides against adult *Triboliumcastaneum,* a serious pest of all kinds of stored grain. Jaya (2004) stated that rapid increase in population density and advancement in agricultural technology has led to a greater release of xenobiotic compounds into the environment. The major sources of xenobiotics applied are pesticides, which are in high demand these days to control pests and diseases as well as to protect the agricultural products from microbial spoilage during transition. Certain lipophilic organocompounds accumulate in the environment reaching very high proportions that have been used fairly extensively for the control of a wide range of insect pests all over the world mentioned by Chauhan*et al.,* (2000)

Soil enzymes catalyze chemical, physico-chemical and biochemical reactions involved in nutrient cycling of soil Nannipieri and Landa, (2000). The enzyme activity is considered as an index of microbial activity in the soil Sriramchandrasekharan and Vaiyapuri, (2002). Measurement of enzyme activities are of great value in screening of the susceptibility of soil processes to agrochemical amendments Nannipieri, (1994); Weaver *et al*, (1994); Alef and Nannipieri, (1995) Besides the organochemicals like pesticides, enzyme activities are also

influenced by heavy metals Moreno *et al*, (2003), soils type Ravi and Siddaramappa, (2002) and fertilizers Ravi *et al*., (2002).

Microbial transformation of sulphur regulates the bioavailability, toxicity and environmental impact of these elements in the biosphere. Germida and Siciliano, (2003) reiterated that now days, large scale application of the pesticides in agricultural soils seldom affect the sulphur oxidizers, in particular species of fhiobacilli. Kalpana (1996) analysed to investigate the short-term toxicity of three pesticides such as quinalphos (organophosphorus), endosulfan (organochlorine) and sevin (carbamate) to two micro-green algae viz. Ankistrodesmusconvolutus and Scemdesmusbijugatus while most of the reports available in this area concentrate on toxicity effects based on long-term experiments.Tayyaba (1986) referred the indiscriminate use of various types of pesticides in the modern world has led to much greater emphasis on the possibility of serious environmental contamination which is a fast developing menace to the survival of mankind. Muni (2016) stated that in current scenario, water in most Ramsar wetlands has been heavily degraded, mainly due to agricultural runoff of pesticides and fertilizers, and industrial and municipal wastewater discharges, all of which cause widespread eutrophication. Bindumol (2018) opined that Pesticide is defined as any substance or mixture of substances intended for preventing, destroying or controlling any pest, including vectors of human or animal diseases.Fogg and Boxall (2004) studies on the efficacy of bio beds to treat contaminated water from pesticide spillages, leakages, decontamination of tractors and sprayers were carried out.Arias-Estevez *et al*., (2008) reviewed the influence of physical and chemical characteristics of the soil system, such as moisture content.Giannouli and Antonopoulos, (2014) analysed, Pesticide leaching model can be used to describe soil water dynamics and pesticide transport to soils.Janci (2009) mentioned that a wide range of pesticides is used in agricultural field for crop protection and to control unwanted agricultural pests like brown plan hoppers, green leaf hopper, leaf roller and stem borer. Bailey *et al.,* (2000) also stated that Pesticides applied to forest, cropland, roadsides and gardens are inevitably lost to water bodies.

Discussion and analysis

Due to their abundance and comparatively easier handing, the aquatic larvae of Pantalaflavecens (Fig 1, 2&3) selected as the experimental animal. The larvae were collected with the help of round scoop nets locally known as Longthrai and triangular scoop nets locally known as Thelongfrom the sallow area of Loktaklake during January 2007. Larvae are active, resting on the bottom with rotting leaves and vegetation and weed – dwellers in habits. They are also sluggish and bottom dwelling in habit, and have a covering of mud over their body. Soon after collection the larvae were transferred into polythene bags containing clear water from the lake itself. The bags were than filled with oxygen and the upper ends were tightly bound by a thread and brought to the laboratory. The larvae were released in aquarium containing water from the lake. They were released in aquarium containing water from the lake. They were acclimatized for 1 week before conducting toxicity experiments.

Fig. 1 – Larvae of Odonata

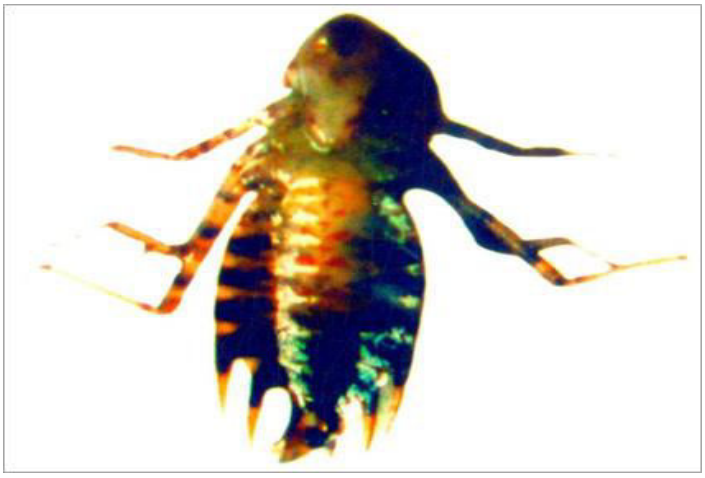

Fig. 2 – Ventral view of PantalaFlavescens

Fig. 3 – Dorsal view of Pantalaflavescens

Acclimatization

The acclimatization of the larvae was done for 1 week in the laboratory of the Zoology Department, NaoremBirahari College, Khunadrakpam in an aquarium filled with water from the Loktak Lake itself. During this period the larvae were fed with plankton collected from a local pond of college campus by plankton net. Acclimatization was necessary to make the larvae become adapted to the changed habitat with a restricted moving space.The odonate larvae were released into the aquarium in the morning. They were provided natural food (plankton) collected using a plankton net from a pond in the College campus. Aquatic plants like *Azolla, Potamogeton, Pistia* etc. were also added to provide a resting substratum to the larvae. The room temperature was 18^0 C & the water temperature was 16^0 C. Any larvae found dead were taking out using a long forceps.When the temperature was below 15 degree the larvae were found in group and hanging together on the roots and leaves of aquatic plants present in the aquarium. During this period, electric bulbs were placed over the aquarium to provide some heat.

Pesticides used

Monocrotophos ($C_7H_{14}NP$) – 36%: Monocrotophos is a systemic insecticide but also act as a stomach and contact poison. It is marketed as colourless crystals with mild ester smell. Monocrotophos is miscible with water and all organic solvents. It is rapidly hydrolyzed in alkaline solutions than in acid. It is a hazard to wild life if the food is contaminated with residues above 0.5 PPm. Its acute oral LC 50 value for rat is 20 mg/kg. It is effective against a wide variety of insect pests via, thrips, aphids, leaf mines, cotton boll arrow, white flies and some mites etc.

*Endosulphan*16% - EC: Endosulphan is a dark brown liquid consisting of Endosulphan technical 35 w/w, solvents, emulsifiers and stablizers. It is used as a broad spectrum insecticide acaricide to control aphides, jassids, thrips, beetles, weevils, foliar feeding larvac, mites, borers, cutworm bugs, white flies, scale etc.

Endosulphan 16% E.C. is an organophosphorous insecticide effective against mining on crops. Endosulphan is absorbed through the skin and has an acute oral LC 50 in rats of 30-79 mg/kg. It acts as a stomach and contact poison of low toxicity to beneficial insects like honey bees, parasites and predatory animals when uses at recommended dosages. It is toxic to fish but is well tolerated by crops.

Chlorpyrifos($C_9H_{11}Cl_{13}NO_3$) 20% EC: It is white crystalline substance with mild mercaptanodour. It is practically insoluble in water but readily soluble in most organic solvents. Its is a contact and stomach poison with vapour action also and effective against many pests with particular reference to rice pests, borers, aphids, mosquitoes, soil inhibiting insects and some mites. It is formulated as 20 EC and available in the name of **Dursban.** Its acute oral and dermal LC 50 values to rat and rabbit are 135 mg/kg and 1000 to 2000 mg/kg respectively.

Dimethoate($C_5H_{12}O_3PS_2N$) 30% EC: Dimethoate is a white crystalline solid, melting points is from 121.8 to 123.67 degree and technical material contains 94 to 96 % chemical. It is a systemic and contact poison effective against sucking insect pests and mites of a wide variety of crops and fruits trees. It is highly toxic to insects but less toxic to mammals, its acute oral and dermal LC 50 values for rats are 200 to 3000 and 700 to 1150 mg/kg respectively. It is generally formulated as 30% EC and sold in the name of **Roger**.

Melathion ($C_{10}H_{10}O_6PS_2$) 50% EC: It is purified compound of brownish liquid and chemically it is dimethyl Phosphorodithioate. It is soluble in water, slightly soluble in mineral oils and soluble in most organic solvents. It is non-stable in alkaline medium. Melathion is mostly used as a contact insecticide though it acts as a stomach poison and exhibits some fumigant action also. It is used for the control of a wide variety of pests of crops such as aphids, hoppers, bugs, caterpillars, flea, beetle and mites, etc. It is the safest insecticide available so for, its acute oral and dermal LC 50 for rat being 1400 to 1900 and 4000 mg/kg respectively. At the recommended dosages, it can be applied to most edible crops within one to three days of harvest.

Dichlorvos ($C_4H_7O_4Cl_2P$) 76% EC: Chemically it is 2, 2 dichlorovinyl diethyl phosphate. It is one of products resulting from the alkaline treatment of trichlorfon. Dichlorovos is a thin colourless liquid with pleasant smell, boiling at 120^0C. It is insoluble in water but readily soluble in most organic solvents. Dichlorvos gets hydrolysed slowly in neutral and acidic media and rapidly in alkaline medium. It is a contact and stomach poison with some fumigant action and has good penetration properties. It brings about rapid knock down of the insects. It is highly volatile compound, therefore, can be used on all crops just before harvest.

Dichlorvos is very effective in baits and in aerosol formulations for the rapid knock down of flies, mosquitoes and moths etc. It's acute oral and dermal LD 50 values for rat are 25-30 & 75 to 900 mg/kg respectively. It is available in the form of 100 EC with the trade names Nuvan and Vapona etc.

Stock solutions:

Since experimental requirement of the pesticide was very small, preparation of a diluted stock solution was necessary. The stock solution was prepared by mixing 1ml of the insecticide with 1000 ml of water. This stock solution contains 0.001 ml of the pesticide per 1 ml. By adding the required quantities of the above solution to 1000 ml of water, different concentrations in ppm [parts per million] of the pesticide were obtained. The solution was thoroughly mixed and kept in a clean beaker of 1500 ml capacity with graduated mark on the beaker.

Experimental procedure:

Toxicity experiments were set up by using round plastic bottles of 2000 ml capacity. Water from Loktak Lake was used in the experiments. The bottles were marked serially as 1, 2, 3, 4, 5 and control 10 healthy and active odonate larvae were put in each bottle for 24 hours (1 day) before adding pesticides.Different quantities of the freshly prepared pesticide stock solution were added in each bottle except the control. The final volume in the bottles was 1000 ml and the bottles contain different concentration of pesticides in ppm. The mouth of the bottle was covered with white thin cloths & fastened using rubber band.

CONCLUSION

The time at which the pesticide was added into the water was noted along with the room and water temperatures. The larvae were observed continuously for their response to the chemical. Whenever a larva dies, it was immediately removed. The total number of larvae died within a period of 24 hours (1 day) was noted for observation of LC_{50}. Larvae which have ceased to exhibit any movement were counted as dead.From the above findings, it can be concluded that the seven common organophosphorous pesticides used in the experiments have significant toxic

effects on aquatic larvae of *Pantalaflavescens*. From the value of the LC_{50} for the different pesticides it is also evident that the Pantalaflavescens is very sensitive to the presence of the pesticides in the ambient water.Pesticides applied in the surrounding agricultural areas naturally reach to the lakes water during rainy season along with surface run off. The pesticides exert toxic effects on the various organisms living in the aquatic. This is possible because most of the pesticides remain undergraded for up to three months in the area applied and if rainfall and flooding follows within one month of the application, the pesticides will reach to the lake water. Thus organisms residing in the lake are exposed to pulses of pesticides from to time. If the pesticides contamination occurs before any degradation, insects larvae are exposed to these pollutants.

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