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Alterations In The DNA Content In Different Tissues Of Freshwater Bivalve, *Parreysiacylindrica*es Posed To Thiamethoxam

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Abstract

Freshwater bivalves, *Parreysiacylindrica* were exposed to acute and chronic dose of thiamethoxam to study the effect of pesticide on mantle, foot, gills, digestive glands and whole body tissues. The significant depletion of DNA content was observed in the mantle, foot, gills, digestive glands and whole body tissues of fresh water bivalve, *Parreysia cylindrical* on acute and chronic treatment of thiamethoxam. The depletion was maximum in digestive glands than in mantle, foot, gills and whole body tissues. Depletion of DNA content might be due to depletion of some nuclear DNA or damage of mitochondria and their DNA due to effect of thiamethoxam.

Key words: Thiamethoxam, DNA, Bivalve, *Parreysiacylindrica*.

Introduction

Pesticides are mainly responsible for water pollution hence aquatic organisms in contaminated water bodies come in continuous and direct contact with pollutants.Pesticide after penetrating in the body of organism, affect ultrastructure of cell organelles like mitochondria, endoplasmic reticulum. It may result in imbalance of entire physiological equilibrium and various kinds of complications in the body of organism. Pollutants alter cellular functions and ultimately affecting physiological and biochemical mechanisms of animals (Satyaparmeswara et. al., 2006). Cavin, (1994) has reported that pesticide cause biochemical alterations such as inhibition of enzymes, genetic damage,metabolic disorder, hypertension and cancer due to physiological differences. The mode of action of pesticides is subjected to their chemical properties. The sum of physiological alterations generated by particular pesticide seems to be the characteristics of those pesticides. Changes in the life processes of organisms are the defensive phenomenon to combat with the stressful situation. In spite of detoxification mechanism in the body of organism to get rid of from foreign substances, toxicants induce adverse effects. Such an effect may be at cellular or even at

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molecular level but ultimately it would lead to physiological, pathological and biochemical disorders (Patil and Lomte, 1989; Jain and Kulshreshthta, 2000).

Two polynucleotide strands that winds together to form double helical structure of DNA. Nucleic acid contents are considered as an index of capacity of an organism for protein synthesis. Different hormones and stress conditions may exert control over synthesis, activity and break down of nucleic acids. The nucleic acid contents can cause alterations in genetical information and genome functioning so it is essential to study the levels of DNA and RNA periodically in different tissues of the organisms undergoing stress conditions (Khanuja, 1981). Structural changes in the DNA can be monitored using biochemical methods and usually low quantitative changes are observed on pesticide exposure. DNA strand scission can also be sensitively monitored and even more importantly the particular nucleotide position cleaved can be pin pointed by biochemical methods. This methodology has been applied effectively in monitoring both the efficiency of DNA strand scission by metal complexes, the specific sites cleaved and where the complexes are specifically bound on the helical strand(Black Marsa et. al., 1996). Genotoxicants have the capacity to alter DNA and their effects may be particularly harmful as these agents can induce changes that may be passed on to future generations and have an impact on populations. Environmental pollutants have been reported to induce DNA strand breaks in various mussel cells which can damage their functions (Nicholson and Lam, 2005).

Study of changes in DNA content is useful to know different toxicants and defensive mechanism of the body against toxic effects of pesticides. The biochemical components are indices of pollution and they determine nutritional status, health and vigor of an organism. Investigation regarding the physiological and biochemical changes in aquatic organisms such as molluscs is insufficient. Fresh water bivalves amongst the molluscs are economically important hence, an attempt is made to investigate the effect of thiamethoxam on the DNA content in different tissues of fresh water bivalve, *Parreysiacylindrica*. Some basic mechanisms of the mode of action of pesticides were studied on these model animals which can be applicable to other animals. Vital organs such as mantle, foot, gills, digestive glands and whole body tissues were used to determine the changes in DNA content of their tissues after acute and chronic treatment to thiamethoxam in the fresh water bivalves *Parreysiacylindrica*.

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Materials and Methods

The fresh water bivalve,*Parreysiacylindrica* were collected from the Jamda dam which is nearly 30 Kms away from Chalisgaon, Dist. Jalgaon (M.S). After collection, the bivalves were acclimatized in the laboratory condition at room temperature for 4-6 days. The active acclimatized bivalves of approximately same size were selected for experimentation. Before starting the experiment, these bivalves were divided into three groups such as A, B and C. 1. 'A' group of bivalves were maintained as control. 2. 'B' group of bivalves were exposed to sub lethal dose (14.2114ppm $LC_{50/2}$ of 96 hrs) of thiamethoxamupto 96 hours 3. 'C' group of bivalves were exposed to chronic dose (2.8422ppm $LC_{50/10}$of 96 hrs) of thiamethoxam, upto 21 days. The control and experimental bivalves of A and B groups were dissected after 24 and 96 hours and animals from C group of chronic treatment after 7 and 21 days. Their mantle, foot, gills, digestive glands and whole body were removed. These tissues were dried in oven at 75^0 C to 80^0 C till constant weight was obtained and blended into dry powder. This powder was used for the estimation of DNA content. DNA content of the tissue was estimated by using Diphenylamine method of Burton (1956).

Results and Discussion

Changes in DNA contents in mantle, foot, gills, digestive glands and whole body tissue of freshwater bivalve, *Parreysiacylindrica* after exposure to acute and chronic concentrations of pesticide, thiamethoxam for 24 and 96 hours and 7 and 21 days are summarized in table No. 1.It was observed that after chronic exposure of thiamethoxam, there was significant decrease in the DNA content in mantle, foot, gill, digestive gland and whole body tissue of experimental bivalves as compared to those of control bivalves.

Table 1- Profiles of DNA in different tissues of fresh water bivalve, *Parreysiacylindrica* after acute and chronic exposure to thiamethoxam (Values are given in mg/100 mg of dry tissue).

Tissue	Control				Thiamethoxam (14.2114 ppm)		Thiamethoxam (2.8422ppm)	
	24hrs	96hrs	7 days	21 days	24hrs	96hrs	7 days	21 days
Mantle	1.345 ±0.0456	1.340 ±0.098	1.455 ±0.021	1.421 ±0.055	1.276 ±0.0421 (-5.1301) p<0.05	1.056 ±0.089 (-21.1940) p<0.05	1.250 ±0.0684 (-14.0893) p<0.05	1.043 ±0.0645 (-26.60) p<0.05
Foot	2.577 ±0.062	2.549 ±0.0586	2.612 ±0.087	2.598 ±0.055	2.343 ±0.11 (-9.08)	2.045 ±0.094 (-19.77)	2.245 ±0.124 (-14.05)	2.058 ±0.0984 (-20.78)

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					p<0.05	p<0.05	p<0.05	p<0.05
Gill	1.238 ±0.084	1.19 ±0.095	1.249 ±0.071	1.238 ±0.046	1.11 ±0.068 (-10.3392) p<0.05	0.898 ±0.049 (-24.53) p<0.01	1.025 ±0.0431 (-17.93) p<0.05	0.887 ±0.0213 (-28.35) p<0.05
Digestive gland	2.318 ±0.085	2.298 ±0.113	2.297 ±0.074	2.256 ±0.123	1.835 ±0.065 (-20.83) p<0.05	1.59 ±0.091 (-30.80) p<0.05	1.487 ±0.0124 (-35.26) p<0.01	1.197 ±0.0981 (-46.94) p<0.001
Whole body tissue	2.371 ±0.122	2.365 ±0.136	2.412 ±0.124	2.398 ±0.134	2.212 ±0.124 (-6.7060) p<0.05	1.844 ±0.123 (-22.02) p<0.05	2.034 ±0.109 (-15.67) p<0.05	1.795 ±0.0954 (-25.14) p<0.05

Each value is the mean of five observations ± S.D.

DNA content is the index of capacity of an organism for protein synthesis in the different stress conditions and it was affected by pesticides.The pesticides and related chemicals originating from human activity or agricultural farming and they are discharged directly or indirectly into the waters. The discharge of agricultural runoff, sewage to the aquatic environment and the occasional failing of sewage treatment plants to eliminate some chemicals has raised concerns about the future water quality in rivers, lakes, estuaries and ocean. These ecosystems are very important for many aquatic organisms, because they serve as a nursery area and as source of food and nutrients for fish and birds (Allanson and Baird, 1999). The many aquatic organisms like fishes, bivalves, prawns and crabs of the freshwater ecosystem are adversely affected by pollutants. Pollution is recognized as one of the most important factors causing major declines in populations of freshwater species in various parts of the world (Winfield, 1992; Lawton and May, 1995; Maitland, 1995; Mason, 1996). According to Williams et.al., (1993) nearly 70% of the freshwater mussel (Bivalvia: Unionidae) species in the United States are considered as endangered, threatened or of special concern. Pesticides cause stress to the aquatic organisms (Newell, 1973; Madhyastha, 1996) and change the metabolic activities causing metabolic disarrangement in the living system of aquatic organisms. Pesticide induced alterations in biochemical composition of organs and the physiological state of metabolic activity of an organism reflects in the use of their biochemical energy to counteract the toxic stress (Swami et. al., 1983). Pesticides due to their toxicity produce biochemical changes in the tissues and organs of exposed animals (Shastry and Sharma, 1979).Any kind of toxic stress leads to changes in biochemical and physiological mechanism in the body of organism. In order to study the physiological

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changes after pesticide exposure, the study of alterations in the biochemical constituents is the most fundamental tool.

Biochemical studies are good parameters which help to see the effect of pesticide on biochemical composition of different tissues of bivalve. In terms of energy conservation, the organism would be expected to make compensatory adjustments for gain and loss of energy in such a way that loss is minimized and gain is maximized in case of change in environmental condition (Bayne,1973).Observations recorded in this study showed that the body parts of *Parreysiacylindrica* might have got capacity to change the metabolic substance during the variable stress depending upon type of effect of the stress factor, same results was observed by Mane and Muley (1989) in *Lamellidensmarginalis*.

Significant DNA strand breakage in the foot tissue of*Anodontagrandis* exposed to lead was observedby Black et al., (1996). The metals may be carcinogenic because of their capacity to generate reactive oxygen species and other reactive intermediates or react directly with DNA (Brien et al., 2003). The capacity of carcinogenic metals to facilitate DNA damage through inhibition of DNA repair enzymes (Hartwing et al., 2002) or binding to histones. It is assumed that the ultimate goal of free radicals or metals is DNA and that the mechanisms of carcinogenesis must include genotoxic effects on organisms. According to Mayer et al., (1998) metal such as Ni (II) is only weakly genotoxic and mutagenic. Rao et al., (1998) studied the effect of Fluoride on the nucleic acid contents of freshwater crab, *Barytelphusacunicularis* and observed that the level of DNA in muscles and hepatopancreas were found to be increased initially and then a gradual decrease was observed in gills, testes and ovaries. The decreased in levels of DNA and RNA were observed by Patil and Lomte (1989) in *Mythimaseperata* under toxic stress of pesticides. The cellular degradation rapid histolysis and decreased rate of protein synthesis are possible reasons behind this. As compared and supported by above literature, the present investigation of the acute and chronic exposure of thiamethoxamdepletion was observed in DNA levels in mantle, foot, gills, digestive glands and whole body tissues of fresh water bivalve, *Parreysiacylindrica*. It might be due to the depletion of some nuclear DNA or damage of mitochondria and their DNA.

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