PESTICIDE CONTAMINATION IN SELECT WETLANDS OF NILGIRIS DISTRICT WITH SPECIAL REFERENCE TO SEDIMENTS AND FISH

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Executive Summary

Intensive use of synthetic pesticides in agriculture and public health has resulted in serious environmental hazards. The freshwater bodies adjoining agriculture fields are continuously being contaminated by toxic chemical pesticides. Many organic pesticides and trace elements are hydrophobic; that is, in aquatic environment they tend to be associated with sediment and biological tissues rather than dissolved in water. Hence, studying sediments and fishes are the effective tools to assess the occurrence of these contaminants in the environment.

Pesticides and their transformation products (TPs) are mainly hydrophobic, persistent and bioaccumulable that are strongly bound to sediment/ soil. Of the various aquatic organisms, fishes are proven as excellent indicators of agrochemical contamination, because they can acquire organic pollutants from food, suspended particles or directly from water. Fish can be found virtually everywhere in the aquatic environment and they play a major ecological role in the aquatic food webs because of their function as a carrier of energy from lower to higher trophic levels.

In the present study, 19 sediments samples from three wetlands in Nilgiris district, namely Tarnadmund, Nedugula and Bison Swamp and fingerlings from Tarnadmund were collected and analyzed for organochlorines, organophosphates and synthetic pyrethroids.

Data are discussed to understand the status in the residue levels in the context of usage pattern and policy. Maximum concentration of γ HCH (14.19 ng/g) was recorded in sediment collected from agricultural field in Nedugula. Levels were very less in Bison Swamp (BDL to 0.45 ng/g). Σ DDT ranged from BDL to 75.01; BDL to 48.49 and BDL to 1.71 ng/g in Tarnadmund, Nedugula and Bison Swamp sediments respectively. Among cyclodiene pesticides (α endosulfan, heptachlor epoxide, chlordane and mirex) analysed, trace level of Σ heptachlor was detected only in hilly terrain of Tarnadmund (0.37 ng/g), where agricultural activities have been intense, while residues of α endosulfan, chlordane and mirex were not detected in any of the sediments and fingerlings. Phorate (13.25 ng/g) was the maximum in the pond sediments of Nedugula wetland, followed by Malathion (9.82 ng/g) in the same site. High concentrations of Phorate and Malathion residues in sediment samples of Nedugula reflect their current use on cabbage, carrot and tea. Residues of ethion was the maximum in sediments collected from the upper (2.90 ng/g) and downstream (2.61 ng/g) areas of Nedugula, while it was BDL in Bison Swamp.

Among the individual synthetic pyrethroids, Σ fenvalerate was found to be higher (20.95 ng/g) than other pesticides. Deltamethrin was not detected in any of the sediments samples analysed. Fingerlings of Danio sp could be collected only from a pond near Tarnadmund. It had β HCH (0.61 ng/g) among the HCH isomers tested. The metabolites of DDT, namely o,p DDT and p,p' DDD were detected in the range of 1.26 to 1.50 and 1.0 to 1.26 ng/g, respectively. All other OCs, OPs and SPs tested were BDL in Danio sp. It is necessary to mention that the quantum of pesticide residues detected may not reflect the threat because, the toxicity differs among pesticides. Apart from accumulation in animal tissues through food

chains, locally high rainfall and laterite soil in the district facilitate speedy leaching of toxic chemicals leading to contamination of water bodies not only in the hills but also in the plains.

It looks impossible to dispense with the use of pesticides and other chemicals to meet the growing food requirement by the ever growing human population. Hence, there is a need to promote safe and efficient use of pesticides for sustainable agricultural production in the district although the best will be to dispense with the use of chemicals and adopt organic farming and promote effective and sound Integrated Pest Management programmes.

1. GENERAL INTRODUCTION

Adequate information is available in India to say that our wetlands are contaminated either by pesticides or industrial effluents or by both. It may be noted that the consumption of pesticide in India has crossed more than 40,000 metric tons (MT) from a mere 5,000 MT of 1960s (http://www.indiastat.com retrieved on January 2011). These pesticides find their way to the wetlands either through runoff or seepage and inflict deleterious effects on the entire wetland system. Worst still is that these contaminants even make their way to the nearby drinking water sources also. Estimates show that three million people around the world suffer from pesticide poisoning every year, of which 2,00,000 die, mostly in countries such as India (Sahgal, 2003 and Vijayan *et al.,* 2004).

The intensive use of synthetic pesticides in agricultural fields and public health operation systems has resulted in serious environmental hazards (Singh *et al.,* 2004 & 2006). The freshwater bodies adjoining agriculture fields are continuously being contaminated by the toxic chemical pesticides (Bouregeois *et al.,* 1993, Nayak *et al.,* 1995 and Kalavathy *et al.,* 2001) and pose potential direct threats to freshwater organisms, particularly sensitive animals, such as fish and prawns (Saravanan *et al.,* 2003, Selvarani and Rajamanickam, 2003 and Park *et al.,* 2004).

Pesticide residues may be comprised of many substances, including specified derivatives such as degradation products, metabolites and impurities that are considered to be of toxicological importance. Several studies showed that pesticides could cause health problem such as birth defects, nerve damage and cancer (John *et al.,* 2001, Bedi *et al.,* 2005 and Aulakh *et al.,* 2006). Pesticides can be divided into many classes, of which the Organochlorines (OCs) were the first group to be invented. OC pesticides are non-polar, lipophilic, toxic and highly persistent compounds. These synthetic chemical pesticides reach the aquatic environment via soil percolation, air drift or surface runoff, leaching, disposal of empty containers, etc. Further, these OCs end up in groundwater and their transformation products may remain for years (Mudiam *et al.,* 2011). As a result, they can be biomagnified through food chains and produce harmful effects at every level (Muralidharan *et al.,* 2009).

The organophosphate (OP) groups of pesticides are being most commonly used across the world. They have replaced OCs, having the advantage of being readily biodegradable and less persistence in the environment. Their degradation process is faster than other agrochemicals (Sattar, 1990). These pesticides are potent cholinesterase (ChE) inhibitors. They can bind covalently with the serine residue in the active site of acetyl cholinesterase (AchE), thus preventing its natural function in the catabolism of neurotransmitters.

Pyrethroids are derived from natural pyrethrins with some modifications to enhance their environmental stability or alter their insecticidal activity. Since they guarantee as an effective agent against a broad range of pests, and stable under field conditions, they are used as insecticides in agriculture worldwide (Shan *et al.,* 1999 and Mak *et al.,* 2005). Many products such as Raid brand pesticides that are commonly found in retail stores for home use contain pyrethroids such as Permethrin and Deltamethrin, to eliminate household pests such as ants and spiders (De Pasquale, 2010). Pyrethroids are expected to be present in environment due to their predominant usage for the control of pests. Several studies have indicated that pyrethroids are highly toxic to a number of non-target organisms such as honeybees, freshwater fishes and aquatic arthropods even at very low concentrations (Smith and Straton 1986; Oudou *et al.,* 2004).

Pesticides and their transformation products (TPs) are mainly hydrophobic, persistent and bioaccumulable that are strongly bound to sediment/ soil. Pesticides that exhibit such behaviour include the organochlorines such as DDT, endosulfan, endrin, heptachlor, lindane and their TPs. Most of them are now banned for agriculture but their residues are still present (Vijayan *et al.* 2008, Muralidharan *et al.* 2010). Several metabolic pathways have been suggested, involving transformation through hydrolysis, methylation, and ring cleavage that produce several toxic phenolic compounds. The pesticides and their TPs are retained by soils to different degrees, depending on the interactions between soil and pesticide properties. The most influential soil characteristic is the organic matter. The higher the organic content, the greater the adsorption of pesticides and TPs. The capacity of the soil to hold positively charged ions in an exchangeable form is important with paraquat and other pesticides that are positively charged. Soil pH is also of some importance (Andreu and Pico, 2004).

Fish is a valuable bioindicator because its detoxification enzymes (e.g. mono-oxygenases) have lower activity than in mammals and thus allows a higher toxicant bioaccumulation. Moreover, fishes can acquire pollutants from food, suspended particles or directly from water (Jayakumar and Muralidharan, 2011). Fish can be found virtually everywhere in the aquatic environment and they play a major ecological role in the aquatic food webs because of their function as a carrier of energy from lower to higher trophic levels (Muralidharan *et al.,* 2008, Jayanthi and

Muralidharan, 2009, Muralidharan *et al.,* 2009 and Muralidharan, 2010). Despite their limitations, such as relatively high mobility, fish are generally considered to be the most feasible organisms for pollution monitoring, especially pesticides in wetlands/ agro-ecosystems (Dhananjayan and Muralidharan, 2010). Hence, studying residue levels in fishes is the most effective tool to assess the contamination load in an aquatic environment.

The current study was carried out with the following objective;

To document the status of pesticide contamination (organochlorines, organophosphates and synthetic pyrethroids residue levels) in sediments and fish collected from Tarnadmund, Nedugula and Bison Swamp wetlands of Nilgiris district.

2. BACKGROUND INFORMATION

The importance and usefulness of wetlands was first brought to the notice of the world through a Convention on Wetlands held at the Iranian city of Ramsar, in the year 1971. The Convention was an inter-governmental treaty that provided the framework for national action and international co-operation for the conservation and wise use of wetlands and their resources. As of 9th March 2007 there are 154 Contracting Parties to the Convention, with 1650 wetland sites, totalling 149.6 million hectares, designated for inclusion in the Ramsar list of Wetlands of International Importance.

The worldwide consumption of pesticide is about 2 million MTs per year, of which 24 % is consumed in USA alone, 45 % in Europe and 25 % in rest of the world. In this context, the average usage of pesticides in India is about 0.6 kg/ha. Among various types of pesticides used in India, 40 % belong to OCs (Abhilash and Singh, 2009). The annual (2008-'09) consumption of chemical pesticides for agriculture in the country has been estimated at 43,860 metric tons (MT).

Consumption of pesticides in India varies with the cropping pattern, intensity of pests and disease and agro-ecological regions. Pesticide use is particularly high in regions with good irrigation and also in those areas where commercial crops are grown (i.e. cashew plantations in northern Kerala and Karnataka). The major use of pesticides in India is for cotton crops (50 - 55 %), followed by paddy and wheat (>40 %) (Shetty, 2004). Chemicals belonging to groups such as organophosphates, carbamates and synthetic pyrethroids are indiscriminately used to control crop pests unaware of their impacts on the ecosystem.

An extensive study done on inland wetlands of India by SACON indicates the threats and conservation issues of the wetlands across the country. They pointed out two overwhelming issues of immediate concerns: huge loss of wetlands in the country and, contamination of the remaining ones. There has been a loss of about 38% of wetlands across the country, since the last 10 years. And, in some districts the loss is up to 88%. A very conservative figure worked out from the existing data shows that there is a substantial loss in the number of wetlands, about 2,76,940 to 2,92,327 during the last 10 years. One of the major reasons for such drastic decline is the failure to consider wetland as a productive unit of land. Instead, it has been considered as wasteland. Land use statistics of none of the states shows a separate category of wetland.

Therefore, apparently there is no legal apparatus for protecting the wetlands. Lack of awareness of wetland values could be one of the major reasons for not recognizing wetland as a separate entity and for not giving the importance it deserves (Vijayan *et al.,* 2004). It has been reported that wetland is the most productive ecosystem and that in terms of economic and ecosystem service values it outweighs forest ecosystems almost seven times. Compounding to the loss of wetlands is that almost all the wetlands studied from 14 states in the country are polluted. In many cases, the fishes are not fit for human consumption, because of the high levels of heavy metals or pesticides. It is a cause for great concern that not even one of the several hundred fishes studied from 115 wetlands was free from pesticides/ heavy metal (Vijayan *et al.,* 2004).

Rao *et al.,* (1994) reported residues of DDT, HCH and Carbofuran associated with high organic contents in the waters of Ooty Lake. Rajukkannu *et al.,* (1989) reported residues of a few nematicides in potato. Regupathy and Kuttalam in (1990) recorded organochlorine contamination in human milk in Nilgiri district. Vijayan and Muralidharan in 1999 conducted a study on pesticide contamination in sediment, water, fishes and bird samples collected from major water bodies of Nilgiri district.

The present study was undertaken to monitor the residue levels of organochlorine, organophosphate and synthetic pyrethroid pesticide residue levels in sediments and fish collected from Tarnadmund, Nedugula and Bison Swamp wetlands of Nilgiris district.

3. MATERIALS AND METHODS

3.1. Study sites

Wetlands, namely Tarnadmund, Nedugula and Bison Swamp were selected for the present study.

Figure 1 : Viiew of TarnadMand Wetland

Figure 2: View of Nedugula wetland

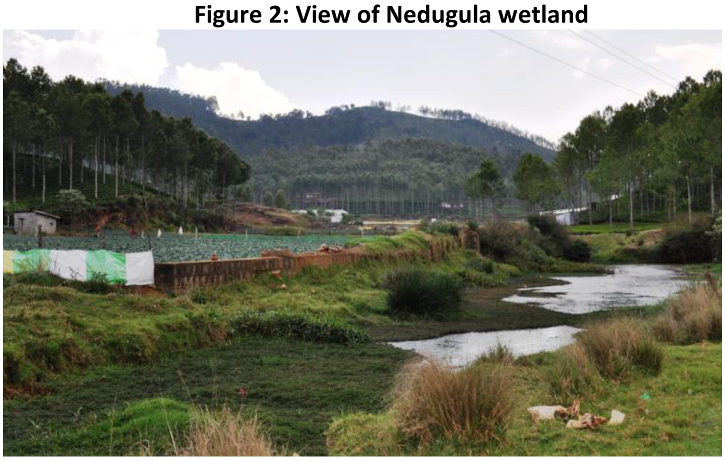
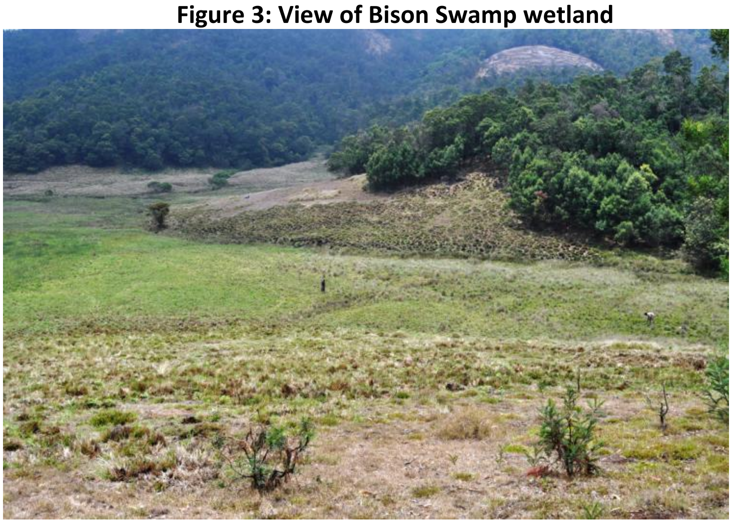


Figure 3: View of Bison Swamp wetland



3.2. Sample collection

Nineteen sediment samples were collected from three wetlands, namely Tarnadmund, Nedugula and Bison Swamp of Nilgiris district. Due to low water availability fish samples were not available in Nedugula and Bison Swamp to quantify pesticide residues. Only one species of fingerlings (*Danio* sp) could be collected in Tarnadmund and pooled together to make a sample in triplicate. All the samples were labelled and packed in clean polythene bags, transported on ice to the laboratory at Sálim Ali Centre for Ornithology and Natural History, Coimbatore. Fish samples were cleaned off dirt in tap water and whole fingerling was minced into smaller pieces and sub samples were taken from the homogenate. About 10 g of the homogenate was weighed using a top loading electronic balance (Mettler AE420) and transferred to clean specimen vials and stored in freezer at -20°C until further analysis.

Figure 4 : Sediment sample collection

3.2.1. Sample processing

3.2.1. i. Sediment

Samples were processed adopting standard operating protocol (Codex Alimentarius WHO/FAO, 2003). Samples were air dried and ground with pestle and mortar, and sieved through 0.5 mm sieve prior to further processing (Je *et al.,* 2003). About ten g of dry sediment samples were mixed with anhydrous sodium sulphate (Qualigens fine chemicals, Mumbai) and then transferred into a 250 ml pre-washed conical flask, about 150 ml Dichloromethane (Merck Specialties, Mumbai) was added and the samples were sonicated in Ultrasonic water bath for 60 min at 30°C. After sonication they were allowed to stand in 30°C water bath overnight. The samples were again sonicated for 60 minutes at 30°C, and then filtered through prewashed glass wool placed on the Pasteur pipette (Hoof and Hsieh 1996). The extracts were condensed using rotary evaporator (Buchi) to a specific aliquot on a florisil (60-120µ mesh size SD Fine chemicals, Mumbai) and silica gel (60-100µ mesh size, Qualigens fine chemicals, Mumbai) packed glass column for clean up. The eluate was condensed in a vacuum evaporator and stored in deep freezer at -20˚C until final analysis was carried out.

3.2.1. ii. Fingerlings

QuEChERS (Quick, Easy, Cheap, Effective, Rugged and Safe) multiresidue extraction method was followed with suitable solvents and reagents for extracting pesticide residues. Ten grams of tissue was taken and ground with mortar and pestle to get a homogenous mixture. Then the homogenate was transferred into a 50 ml centrifuge tube (Oakridge, Tarsons) and 20 ml of Acetonitrile (Merck Specialties, Mumbai) was added to it and shaken vigorously for a minute. To the centrifuge tube, 4 g of anhydrous Magnesium sulphate ($MgSO_4$ - Himedia, Bangalore), 1g of Sodium chloride (NaCl - (Merck Specialties, Mumbai) salts were added and centrifuged at 10,000 rpm for 10 mins. Then the organic layer of the samples (4 ml) was transferred into a 15 ml centrifuge tube which contained 100 mg of PSA and 600 mg of $MgSO_4$ and again centrifuged at 5,000 rpm for 5 mins. From the above extract, 2 ml was transferred into test tube to evaporate to dryness and reconstituted with 1 ml of HPLC grade n-

Hexane and stored in deep freezer at -20˚C until final qualitative analysis was carried out.

Figure 5. Extraction of pesticide residues - in progress

3.3. Chemical Analysis

Quantitative analysis of OC, OP, and SPs were made with Agilent Model 7890A Series Gas Chromatograph equipped with 7000 GC-Quadruple and HP-5 fused silica capillary column (15m x 0.25mm I.D x 0.25μm film thickness) coated with 5% phenyl methyl siloxane with Helium (IOLAR) as carrier gas (1.2 ml/min) and chromatographic operating conditions were as follow: detector 325^0C; injector: 325^0C; oven temperature was programmed as 70^0C-1min; 8^0C/min-280^0C-9.2 min. All the samples were analyzed for organochlorine pesticides, namely alpha-Hexachlorocyclohexane (α-HCH), beta-Hexachlorocyclohexane (β-HCH), delta Hexachlorocyclohexane (δ-HCH), gamma-Hexachlorocyclohexane or lindane (γ-HCH), o,p-Dichlorodiphenyltrichloroethane (DDT), o,p-Dichlorodiphenyldichloroethylene (DDE), p,p-Dichlorodiphenyldichloroethylene (DDE),o,p-Dichlorodiphenyldichloroethane (DDD), p,p-Dichlorodiphenyldichloroethane (DDD), α-Endosulfan, heptachlor, Heptachlor Epoxide (HE), Chlordane and Mirex, organophosphate pesticides, namely Phorate, Malathion, Phenthoate, Primiphos ethyl and Ethion and Synthetic Pyrethroids, namely Permethrin I, Permethrin II, Cypermethrin I, Cypermethrin II, Cypermethrin III, Cypermethrin IV, Deltamethrin I, Deltamethrin II, Fenvalerate I and Fenvalerate II. An equivalent mixture manufactured by Sigma-Aldrich chemicals (Accustandard - United States) was used as standard. Concentrations of individual compounds were quantified from the peak area of the sample to that of the corresponding external standard. Recoveries of the compounds from fortified samples (50 μg/kg) ranged from 91 to 102% and the results were not corrected for per cent recovery and expressed in wet weight basis. Analysis were run in batches of 10 samples plus four quality controls (QCs), namely reagent blank, matrix blank, QC check sample and random sample in duplicate. The minimum detection limit for each pesticide was given in table 1. The limits of detection (LOD) for the method were in the range of 0.25-0.75; 0.29-0.62 and 0.09-0.40 ng/g for the organochlorine, organophosphate and pyrethroid compounds respectively. All concentrations are expressed on dry weight basis for sediments and wet weight basis for fingerling.

Table 1: Detection limit of pesticides analyzed in the present study

S.No	Name of the pesticide/ group	Detection limit (ng/g)
	Organochlorines (OCs)	
1.	α HCH	0.27
2.	β HCH	0.43
3.	γ HCH	0.33
4.	δ HCH	0.32
5.	*o,p* DDT	0.75
6.	*o,p* DDE	0.32
7.	*p,p* DDE	0.33
8.	*o,p* DDD	0.30
9.	*p,p* DDD	0.58
10.	α Endosulfan	0.38
11.	Heptachlor	0.25
12.	Heptachlor Epoxide	0.49
13.	Chlordane	0.33
14.	Mirex	0.25
	Organophosphates (OPs)	
15.	Phorate	0.33
16.	Malathion	0.36
17.	Phenthoate	0.44
18.	Primiphos ethyl	0.62
19.	Ethion	0.29
	Synthetic Pyrethroids (SPs)	
20.	Permethrin I	0.21
21.	Permethrin II	0.31
22.	Cypermethrin I	0.30
23.	Cypermethrin II	0.40
24.	Cypermethrin III	0.34
25.	Cypermethrin IV	0.34
26.	Deltamethrin I	0.09
27.	Deltamethrin II	0.31
28.	Fenvalerate I	0.38
29.	Fenvalerate II	0.27

4. RESULTS AND DISCUSSION

4.1. Total pesticide residues in sediments of select wetlands in Nilgiris district

Nineteen sediments samples from three wetlands of Nilgiris district, namely Tarnadmund (TAR), Nedugula (ND) and Bison Swamp (BSW) and *Danio* sp. fingerlings from Tarnadmund were collected and the data have been analysed to check the overall load of pesticides in the study sites with reference to group, namely organochlorines, organophosphates and synthetic pyrethroids. Further, within the groups, levels of individual pesticides and their isomer and metabolites have been compiled to understand the problem with reference to their usage.

Total OCP residue load was found to be the maximum (157.20 ng/g) in midstream sediments of Nedugula, followed by downstream in Tarnadmund (135.57 ng/g), while it was below detection limit (BDL) in midland and highland areas of Bison Swamp. Total organophosphate residues were found to be higher in pond sediments in Nedugula (23.54 ng/g) followed by near stream in Tarnadmund (8.63 ng/g). Total synthetic pyrethroids were found to be higher in sediments collected from midstream in Nedugula (62.79 ng/g) followed by downstream in Tarnadmund (39.62 ng/g), while the levels were found to be the lowest in Bison Swamp (1.13 ng/g). The mean variations in the total organochlorines, organophosphates and synthetic pyrethroids residue levels in sediments are presented in Fig. 3-5.

4.1.1. Residues of organochlorine pesticides in sediments in Tarnadmund, Nedugula and Bison Swamp wetlands of Nilgiris district.

Many organic pesticides and trace elements are hydrophobic; that is, in aquatic environments they tend to be associated with sediment particles and biological tissues rather than dissolved in water. For this reason, sampling sediment and fish is an effective way to assess the occurrence of these contaminants in the aquatic environment.

a). HCH and its isomers

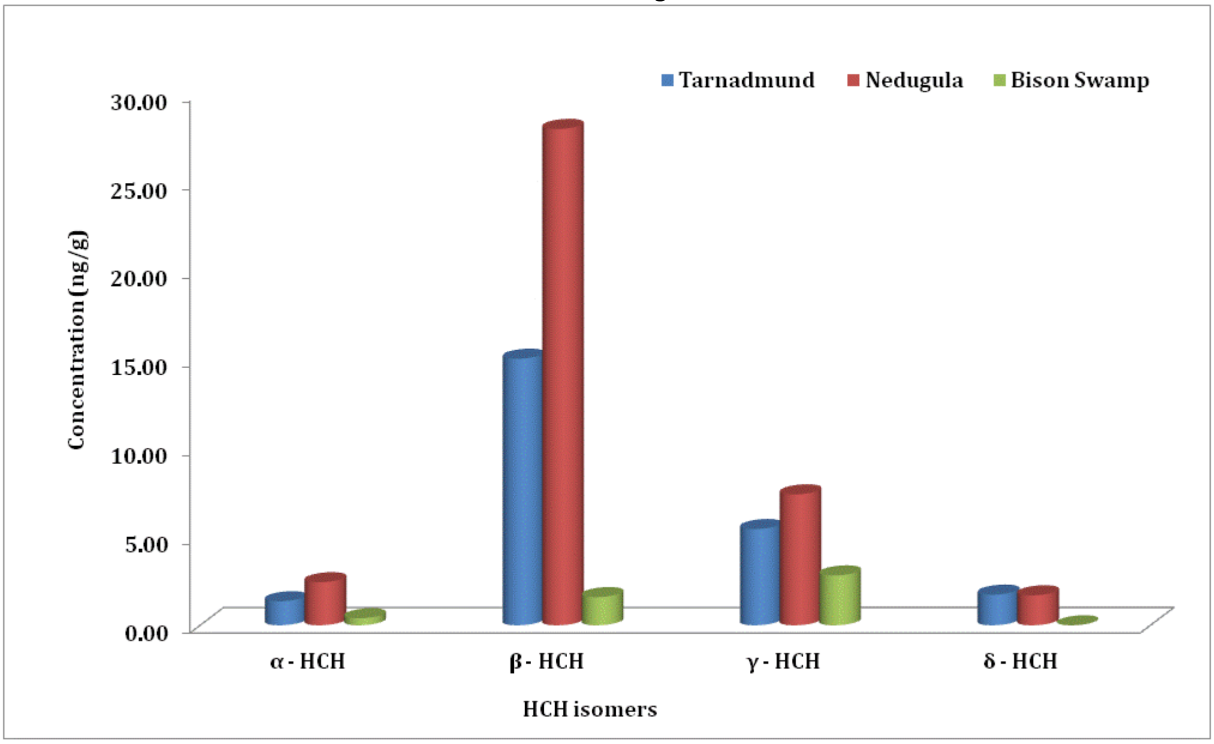
Four isomers of HCH, namely alpha (α), beta (β), gamma (γ) and delta (δ) were detected in the sediments collected from the study sites in Nilgiris district. Variations in the HCH isomer residue levels in sediments are presented in Fig. 1. Alpha HCH was recorded the maximum in sediments collected from the pond (4.06 ng/g) and midstream area of Nedugula (4.03 ng/g), while it was minimum in Bison Swamp (BDL - 0.45 ng/g). Predominance of α HCH in environmental samples reflects the recent use of technical mixtures of HCH (Doong *et al.,* 2002). Maximum concentration of γ HCH (14.19 ng/g) was recorded in sediment collected from agricultural field in Nedugula, followed by midstream (13.39 ng/g) and pond sediments (12.60 ng/g) of the same area. Levels were very less in Bison Swamp (BDL to 0.45 ng/g). The levels of γ HCH was higher (10.98 ng/g) in downstream sediments of Tarnadmund as well. δ HCH levels were the maximum in downstream area of Tarnadmund (2.65 ng/g). In Nedugula, δ HCH was detected in the pond (2.14 ng/g) and midstream area (2.10 ng/g) whereas levels were below detection limit (BDL) in Bison Swamp. β HCH (89.15 ng/g) was the maximum in the midstream sediments in Nedugula wetland. Residues of total HCH (Σ HCH) in sediments ranged between BDL and 108.71 ng/g.

Levels of α HCH in the sediments of Nedugula (BDL - 4.06 ng/g) and Tarnadmund (BDL -3.09 ng/g) are higher than the levels reported by Vijayan and Muralidharan, (1999) in Pykara, while levels in Bison Swamp are lower. Further, levels of Σ HCH in 70 % of the samples are higher than the levels reported by Vijayan and Muralidharan, (1999) from sediments of various reservoirs, namely Avalanche, Uppar Bhavani, Emerald, Kamaraj Sagar, Pykara and Maravakandy, Ooty lake and rivers such as Coonoor and Moyar in Nilgiris district. Σ HCH levels are lower than the levels recorded in the coastal sediments of Chennai, Cochin and Visakapattinam (Senthilkumar *et al.,* 2001) and the river sediments of Gomati, Lucknow, India (Malik *et al.,* 2009).

Technical HCH is a broad spectrum organochlorine insecticide that was available for decades and used throughout the world for agricultural and non-agricultural purposes because of its effectiveness and low cost (Li *et al*. 1998; Raina *et al.,* 2008; Carvalho *et al*. 2009b). It has more than five isomeric forms (α, β, γ, δ and ε etc), which are all toxic, recalcitrant and exhibits both acute and chronic toxicity, particularly β isomer, acts as an environmental estrogen (Walker *et al*. 1999). Its use continued unabated until HCH was also banned in 1997 in India, but restricted use was allowed for lindane (99% γ HCH). It is still used illegally for control of mosquitoes and other insect pests (Raina *et al.,* 2008 and Lal *et al.,* 2010).

The presence of β isomer, the most persistent isomer (Phillips *et al*. 2005), can be attributed to the degradation of α HCH. Wu *et al*. (1997) demonstrated that the transformation of α HCH occurs under both toxic and anoxic conditions, while β HCH increased resistance to microbial degradation and lower volatility (Babu Rajendran *et al*. 2005; Phillips *et al*. 2005). Considering that the half-life of lindane is 30-300 days in water and 2 years in soils (INE, 2004), its detection in this study suggests its continuing use for either agricultural or public health purpose. In addition, β HCH and lindane isomers exceeding the sediment quality guidelines (SQG), the threshold effect level (TEL), the probable effect level (PEL) (Buchman 2008) and the maximum permissible concentration (MPC) (Crommentuijn *et al.* 2000) in sediments collected from streams of Tarnadmund and downstream, agricultural field and pond areas of Nedugula indicate possibility of adverse effects to organisms. It is known that lindane causes endocrine disorders (CEC, 2003). Although technical HCH is restricted in many countries, such as Canada, the USA, European Union and Mexico, lindane is still used to treat seeds and control ectoparasites in cattle. Lindane is also used to a lesser degree as a pharmaceutical to treat lice and scabies in developing countries (Li 1999; INE 2004).

Figure 6 : Mean concentration of HCH isomers in sediments collected from select wetlands in Nilgiri district.



b). DDT and its metabolites

Three isomers of DDT (*o,p* DDT, *o,p* DDE and *o,p* DDD) and 2 metabolites (*p,p'* DDE and *p,p'* DDD) were detected in the sediments collected from the study sites in Nilgiris district. Variations in residue levels of DDT isomers and metabolites in sediments are presented in Fig. 2.

Residues of o,*p* DDT were detected in 70 % of the samples. Residues of *p,p'* DDD was the maximum (36.11 ng/g) in the downstream sediments of Tarnadmund, followed by of o,*p* DDT (33.27 ng/g) in the same site. The levels of o,*p* DDD, o,*p* DDE and *p,p'* DDE were in the ranges of BDL to 3.91; BDL to 0.92; BDL to 0.94 ng/g respectively in Tarnadmund samples. Levels of these chemicals were < 0.5 ng/g in other sites. This indicates the past usage of this pesticide in this region. Σ DDT ranged from BDL to 75.01; BDL to 48.49 and BDL to 1.71 ng/g in Tarnadmund, Nedugula and Bison Swamp sediments respectively.

Wong *et al.* (2010) detected the highest concentration of DDT in urban soils of an endemic malarial region, Maxico (360 ng/g), while the concentrations found in agricultural soils of the same region (mean of Σ DDT = 10 ± 21 ng/g) were lower than the present study. However, levels of Σ DDT in >50 % of the samples are higher than the levels reported by Vijayan and Muralidharan, (1999) from various reservoirs, namely Avalanche, Uppar Bhavani, Emerald, Kamaraj Sagar, Pykara and Maravakandy, Ooty Lake and rivers such as Coonoor and Moyar in Nilgiris district. Σ DDT levels are lower than the levels recorded in the coastal sediments, Chennai, Cochin and Visakapattinam (Senthilkumar *et al.,* 2001) and in river sediments of Gomati, Lucknow, India (Malik *et al.,* 2009).

The Σ DDT concentration obtained in this study is higher than the concentration reported by Babu Rajendran *et al.,* (2005) in sediments (0.04 to 4.79 ng/g) collected from the Bay of Bengal, India; Pandit *et al.,* (2006) in coastal marine sediments (5.68 ng/g) of Mumbai and lower than the sediments (BDL to 480 ng/g) collected from select locations along the Mumbai trans-harbour line, Mumbai (Vijayan *et al.,* 2008).

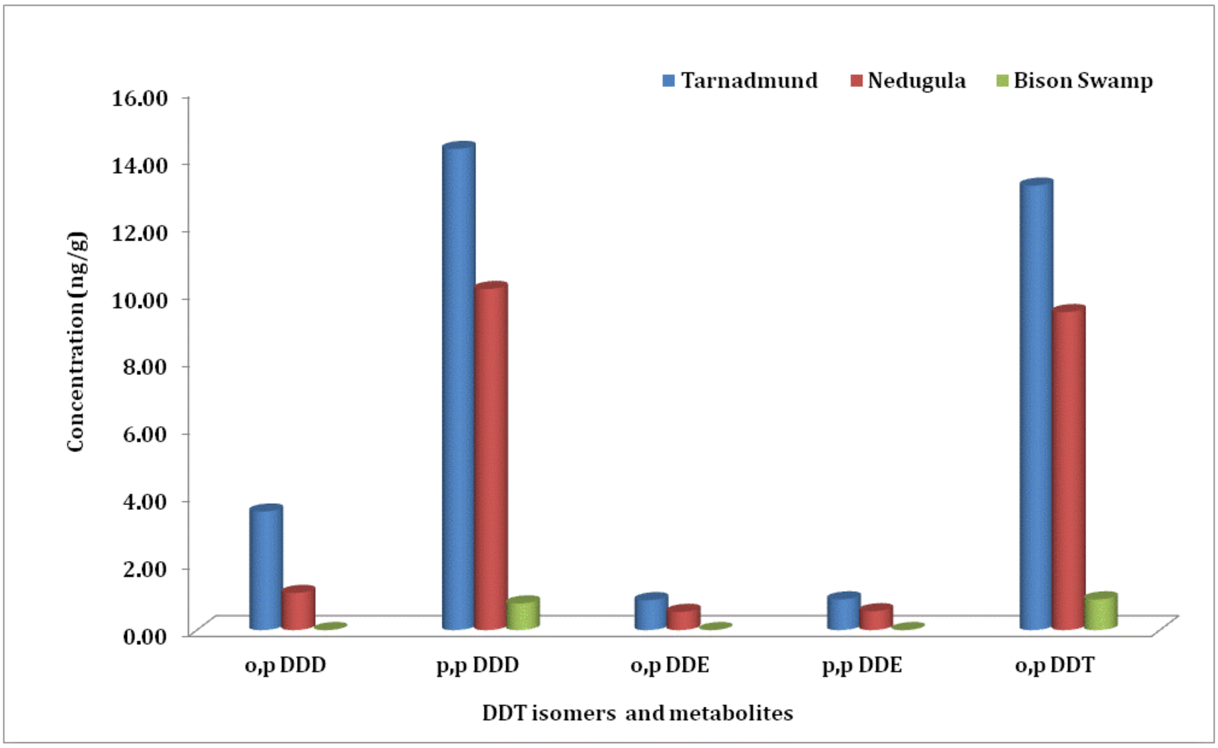
The DDE/ DDT ratio in sediments measured in the Tarnadmund and Nedugula indicates slower degradation rates in these wetlands. The high incidence of DDE has ecological implications because this metabolite is more persistent and toxic than DDT. Furthermore, DDE is known to alter metabolic functions in organisms by acting as an antiandrogen, binding to androgen receptors and inhibiting transcriptional activation, which causes reproductive abnormalities (WHO 2004). The concentrations of *p,p'* DDE (BDL - 0.94 ng/g) did not exceed the sediment quality guidelines (SQG), threshold effect level (TEL), probable effect level (PEL) (Buchman 2008) and maximum permissible concentration (MPC) (Crommentuijn *et al.* 2000).

However, as per the Canadian sediment quality guidelines on PEL, the concentrations of total DDT exceeded the 6.75 ppb level in 27 % of the samples.

Technical DDT is typically composed of 77.1% *p,p'* DDT, 14.9% *o,p* DDT, 4% *p,p'* DDE, and some other trace impurities; thus, the ratio of parent compounds to metabolites has been used to infer the age of contaminant residues in sediments (Zhu *et al.* 2005). In many parts of India, DDT has also largely lost its effectiveness (Sharma, 1999).

In India, the use of DDT in agriculture was banned in 1989 with a mandate to use a maximum of 10,000 tons of DDT per annum for the control of malaria and Kala-azar and this policy is strictly adhered to till date (Dash *et al.,* 2007). DDT can accumulate and biomagnify in organisms (Walker 2001) due to its lipophilicity (K_{ow} = 5.7 - 6.36) and persistence ($T_{1/2}$ = 10 - 15 years). Their long persistence is the reason that residues of DDT and its metabolites can still be detected in the environment. DDE is the primary metabolite found in the environment and is more persistent than DDT. The degree of persistence of DDE varies considerably, as persistence depends on soil type and temperature. The estimated half-life of DDE in tropical and temperate soils is 10 and 30 years, respectively (Hwang *et al.* 2006), while the half-life of DDT varies from 6 weeks to 1.5 years in tropical soils and from 10.5 to 35 years in temperate soils (Fogh *et al.* 2001). The concentrations of p,p'-DDE observed in this study are lower than to those found by Carvalho *et al.* (2002) in the Pabellon Lagoon, Maxico (0.3 - 26 ng/g).

Figure 7: Mean concentration of DDT isomers and metabolites levels in sediments collected from select wetlands in Nilgiri district.

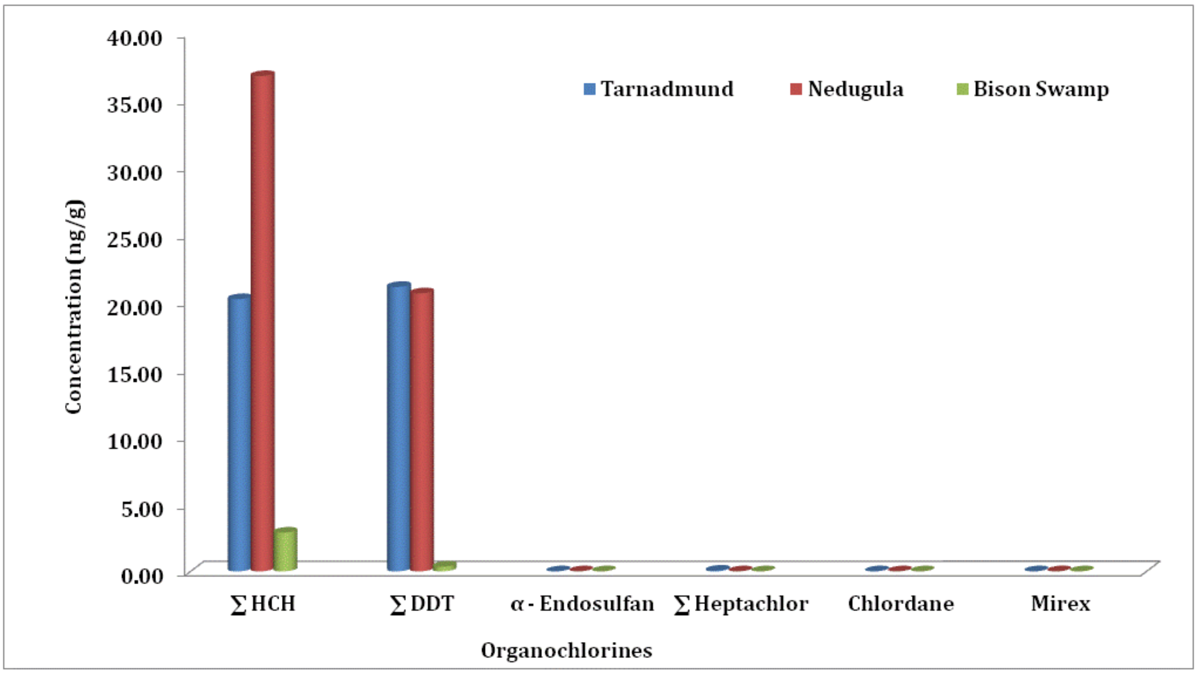


c). Cyclodiene insecticides

Four cyclodiene pesticides (α endosulfan, heptachlor epoxide, chlordane and mirex) have been included in the present study. Of all the samples analysed, trace level of Σ heptachlor was detected only in hilly terrain of Tarnadmund (0.37 ng/g), where agricultural activities have been intense, while residues of α endosulfan, chlordane and mirex were not detected in any of the sediments from the other sites. Further, Σ heptachlor levels are lower than the levels reported by Vijayan and Muralidharan, (1999) from various reservoirs, namely Avalanche, Uppar Bhavani, Emerald, Kamaraj Sagar, Pykara and Maravakandy, Ooty lake and rivers such as Coonoor and Moyar in Nilgiris district and coastal sediments of Chennai, Cochin and Visakapattinam (Senthilkumar *et al.,* 2001) and Gomati river, Lucknow India (Malik *et al.,* 2009). The Supreme Court vide its order dated May 13, 2011 had banned the manufacture, sale, use and export of endosulfan in India. The court had later allowed exports of the stocks but the ban on its use, manufacture and sale within India continued. In April this year the pesticide manufacturers once again appealed to the apex court that they be allowed to manufacture endosulfan from the stock of raw material, Hexachlorocyclopentadiene (HCCP), available with them and use it within the country. The court ordered the centre to revert with a disposal plan of the banned pesticide. The centre has recommended that no further exports of HCCP be allowed in the country. However, the endosulfan manufactures should be allowed to manufacture endosulfan, with the available stock of HCCP, and sell it within India except Kerala and Karnataka (http://www.cseindia.org; Retrieved on 15th October 2012).

The levels of total OCs in the present study are lower than the levels reported by Kumarasamy *et al.,* (2012) in sediments of Tamiraparani river basin, Tamil Nadu during summer. Further, the levels of the OC pesticides and metabolites recorded in the present study are lower than the levels recorded in the sediments of Mahala reservoir, Jaipur (Misra, 1989; Misra & Bakre 1994); Unnao district, Uttar Pradesh, northern Indo-Gangetic region (Singh *et al.,* 2007) and sediments of paddy agroecosystem in Padayetti village, Kerala (Muralidharan & Ganesan 2011).

Figure 8 Levels of organochlorine pesticide residues in sediments collected from select wetlands in Nilgiri district.



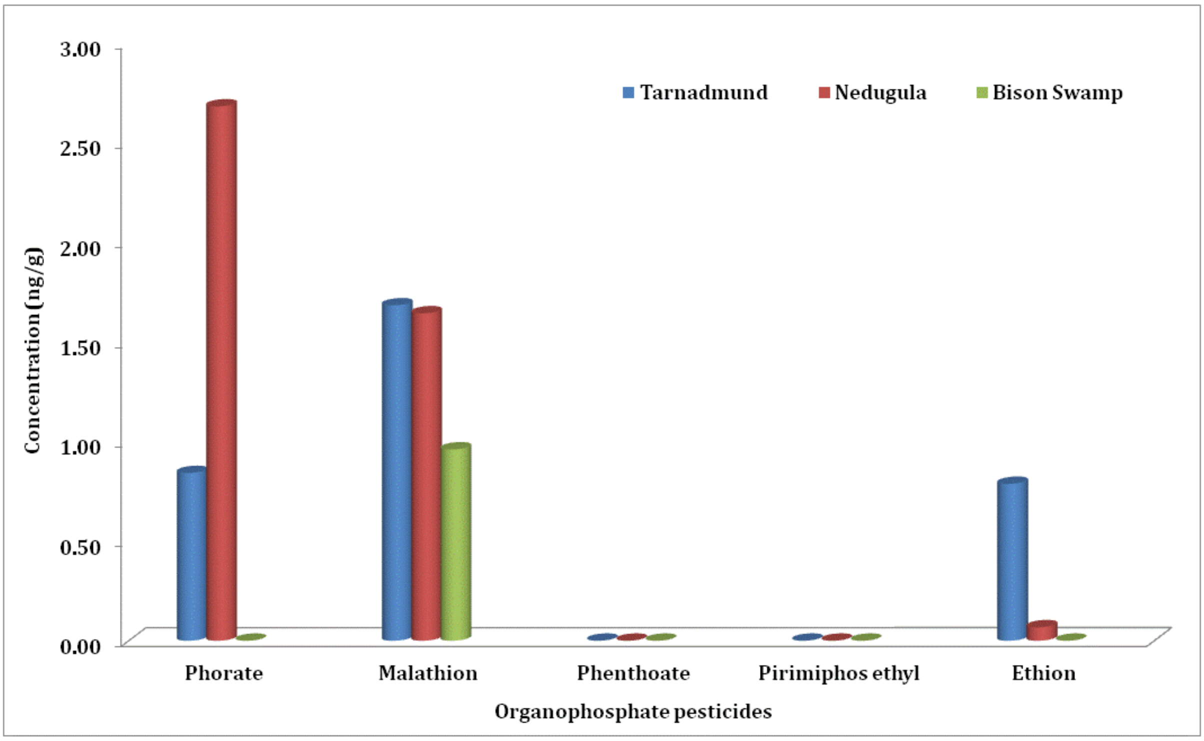
4.1.2. Residues of organophosphate pesticides in sediments in Tarnadmund, Nedugula and Bison Swamp wetlands of Nilgiris district.

Among the three wetlands, residues of total OP in sediments were found to be the maximum in Nedugula (mean 4.39 ng/g; n=7), followed by Tarnadmund (mean 3.31 ng/g; n=7) and minimum in Bison Swamp (mean 0.96 ng/g; n=5). The levels were in the ranges of BDL to 8.63 ng/g; BDL to 23.54 ng/g and BDL to 2.91 ng/g in Tarnadmund, Nedugula and Bison Swamp respectively.

Among the individual OP pesticides analysed in sediments, Phorate and Malathion were detected in 18 and 55% of samples respectively. While Ethion was detected only in 13% of samples, Phenthoate and Primiphos ethyl were not detected in any of the samples analysed. Phorate (13.25 ng/g) was the maximum in the pond sediments of Nedugula wetland, followed by Malathion (9.82 ng/g) in the same site. High concentrations of Phorate and Malathion residues in sediments samples of Nedugula reflect the current use on cabbage, carrot and tea. Residues of ethion was the maximum in sediments collected from the upper (2.90 ng/g) and downstream (2.61 ng/g) areas of Nedugula, while it was BDL in Bison Swamp. The residues of phorate ranged from BDL to 3.51 and BDL to 13.25 ng/g in Tarnadmund and Nedugula sediments respectively. While in Bison Swamp it was BDL.

Sediments collected from Bison Swamp did not have any residues of OP. Variations in organophosphate pesticides were given in Fig. 4. Total OP residues measured in the present study (BDL to 23.54 ng/g) were lower than the levels (0.03 to 1294 ng/g) reported by Parra *et al.* (2012) in sediment samples collected from Culiacan Valley, Mexico.

Figure 9 : Levels of organophosphate pesticides in sediments collected from select wetlands in Nilgiri district.



4.1.3. Residues of synthetic pyrethroids in sediments in Tarnadmund, Nedugula and Bison Swamp wetlands in Nilgiris district.

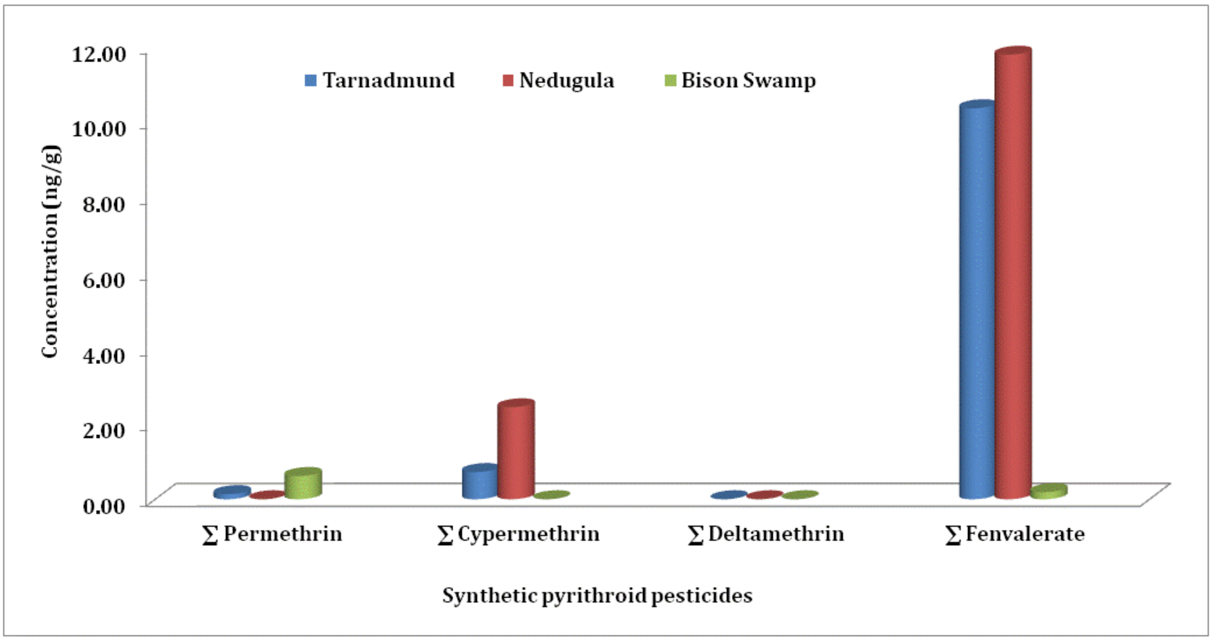
Levels of synthetic pyrethroids in sediments in the study sites differed (Fig. 5). Total synthetic pyrethroids were found to be higher in sediments collected from midstream in Nedugula (62.79ng/g) followed by sediment collected from downstream in Tarnadmund (39.62ng/g).

Among the individual synthetic pyrethroids, Σ Fenvalerate was found to be higher (48.10 ng/g) than other pesticides. Deltamethrin was not detected in any of the sediments samples analysed. Sediment samples collected from Bison Swamp recorded minimal concentration of the synthetic pyrethroids, trace amount of Σ Permethrin (mean 0.61 ng/g) and Σ Fenvalerate (mean 0.8 ng/g) were recorded in sediment collected from streams of Bison Swamp; whereas levels of other SPs were BDL.

Residues of Σ Fenvalerate (48.10 ng/g) and Σ Cypermethrin (10.90 ng/g) were the maximum in sediments collected from middle stream area of Nedugula while it was the lowest in Bison Swamp (BDL - 0.30 ng/g).

Some of these compounds could pose potential threats to the aquatic ecosystems. The present study clearly describes the levels of contaminants in Tarnadmund, Nedugula and Bison Swamp wetlands in Nilgiri district. Hence, it is necessary to continuously monitor the environmental contaminants levels to understand the transport, environmental fate and effects.

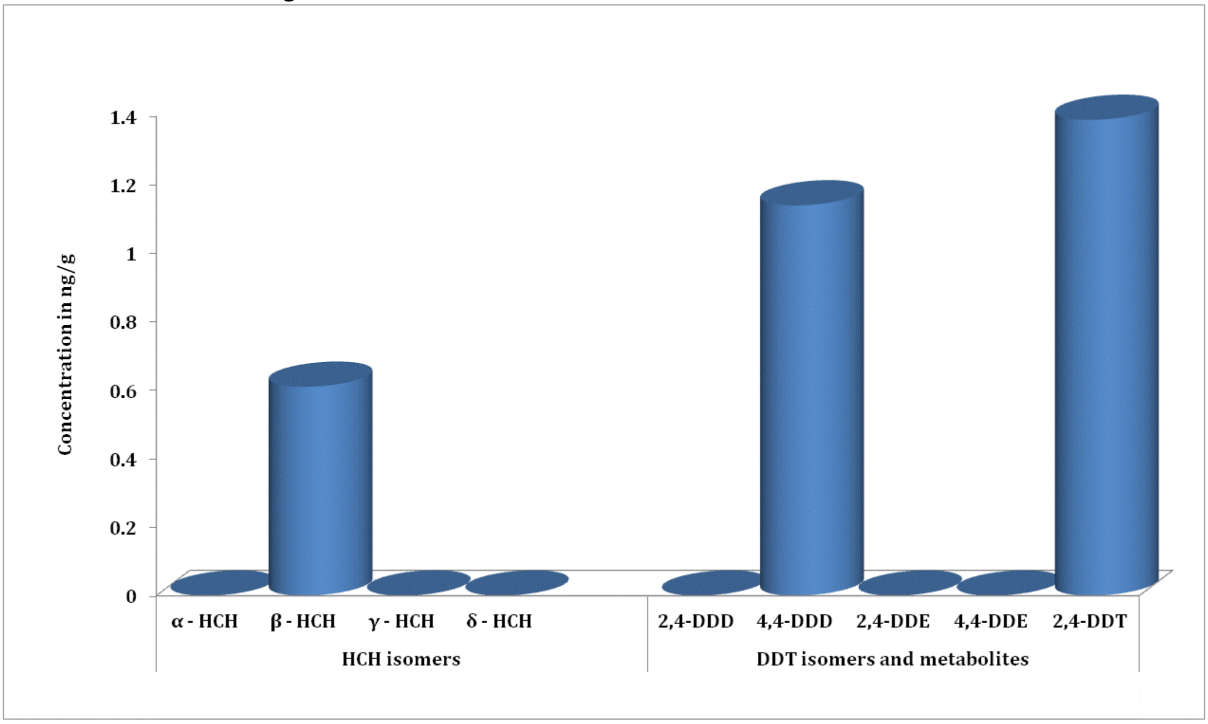
Figure 10 : Levels of synthetic pyrethroid pesticides in sediments collected from select wetlands in Nilgiri district.



4.1.4. Residues of organochlorine, organophosphate and synthetic pyrethroid pesticides in *Danio* sp. fingerlings collected from Tarnadmund wetland in Nilgiris district.

As referred in the methodology, fingerlings of *Danio* sp could be collected only from a pond near Tarnadmund. Levels of HCH and DDT isomers and metabolites levels in fingerlings are presented in Fig. 6. Among the HCH tested only β HCH (0.61 ng/g) was detected in the fingerlings. The metabolites of DDT, namely *o,p* DDT and *p,p* DDD were detected in the range of 1.26 to 1.50 and 1.0 to 1.26 ng/g, respectively. All other OCPs (α endosulfan, chlordane and mirex) tested were BDL.

Figure 11 : Mean concentration of HCH and DDT isomers and metabolites in Danio sp fingerlings collected from Tarnadmund, Nilgiri district.



Levels of Σ DDT are lower than the levels reported by Vijayan and Muralidharan, (1999) in around 1050 fishes comprising six species of fishes studied from various reservoirs (BDL to 65.52 ng/g), namely Avalanche, Uppar Bhavani, Emerald, Kamaraj Sagar, Pykara and Maravakandy, Ooty lake and rivers such as Coonoor and Moyar in Nilgiris district. Vijayan and Muralidharan, (1999) recorded δ-HCH (77.68 ng/g), α-Endosulphan (16.24 ng/g) and *p,p'*-DDD (5.25 ng/g) in the fishes collected from Kamaraj Sagar reservoir, Ooty. In the present study concentration of δ-HCH and α-Endosulphan was found to be below detection limit. However *p,p'*-DDD was recorded in the range of 1.00 to 1.26 ng/g. The residues of DDT recorded in the present study are 5-fold lower than the levels (BDL- 12.3 ng/g) recorded in nine freshwater fishes of different inland wetlands in Karnataka by Dhananjayan Muralidharan (2010).

Residues of OCs in fishes have often been used as indicators of contamination of aquatic environments (Muralidharan *et al.,* 2009). Fishes are most effective indicators of environmental contaminants present in the water as they have greater potential to bioaccumulate and biomagnify particularly the persistent OCs (Muralidharan and Dhananjayan 2010).

Although organochlorine pesticides are found to be higher in sediment samples collected from Tarnadmund than the other places, *Danio* sp. fingerlings collected from Tarnadmund had low levels of organochlorines (2.25-2.76ng/g). *Danio* sp. fingerlings were not found to be contaminated with either organophosphate or synthetic pyrethroids. The levels were only below detectable limits. Even if the fishes were expected to have higher pesticide concentrations than the sediment samples, many factors might have been responsible for the present condition. Size and age of the fish, route of entry and concentration available for intake are very important factors for bioaccumulation. Incidentally we could collect only fingerlings which obviously did not stay in the water for the long to pickup contaminants.

Although many of the OC group of pesticides have been banned, still we find unsafe level of residues in the environment. Further, many short-lived pesticides belonging to OP and SPs are being used quite extensively in the name of replacements. But the impact created by these ephemeral substances on the aquatic ecosystems and particularly fishes are not assessed. These fishes are also considered to be conveyers of chemical carcinogens to the human population.

As the OC pesticides have adverse effects not only on the fishes and other aquatic ecosystems, but also on the public health, their regular monitoring is strongly advised. Although we have phased out many OCs due to their ill effects, still there are many being used for agricultural crop pests and public health purposes. Further, whatever we used till recently are expected to be in the environment for many years.

Figure 12 : OP and SP pesticides being intensively applied for potato, carrot and cabbage

Figure 13 : Agricultural runoff pollutes the wetlands

5. SUMMARY AND CONCLUSION

- Wetlands in Nilgiris, as in rest of India are deteriorating due to severe anthropogenic pressure, changes in land use/ land cover, developmental activities and improper use of agrochemicals.

- Nineteen sediments samples from three wetlands in Nilgiris district, namely Tarnadmund, Nedugula and Bison Swamp and fingerlings from Tarnadmund were collected and analyzed for organochlorines, organophosphates and synthetic pyrethroids. Data are discussed to understand the trend in the residue levels in the context of usage pattern and policy.

- Among the sediments collected from three wetlands, the Σ OCP residues were the maximum in Nedugula (mean 57.43 ng/g; n=7), followed by Tarnadmund (mean 41.39 ng/g; n=7) and minimum in Bison Swamp (mean 3.22 ng/g; n=5).

- Residues of Σ HCH in sediments ranged between BDL and 108.71 ng/g. Among the HCH isomers, β HCH (89.15 ng/g) was the maximum in midstream sediments of Nedugula; whereas concentration of γ HCH (14.19 ng/g) was found to be high in sediment collected from agricultural fields in Nedugula, followed by middle stream (13.39 ng/g) and pond sediments (12.60 ng/g) in the same area. The levels were comparatively less in Bison Swamp.

- β-HCH and lindane exceeded the sediment quality guidelines (SQG), threshold effect level (TEL), the probable effect level (PEL) (Buchman 2008), and the maximum permissible concentration (MPC) (Crommentuijn *et al.* 2000) in sediments collected from stream area of Tarnadmund and downstream, agricultural field and pond areas of Nedugula indicating possibility of adverse effects to organisms. It is known that lindane causes endocrine disorders (CEC, 2003).

- Residues of *p,p'* DDD (36.11 ng/g) and o,*p'* DDT (33.27 ng/g) were the maximum in the downstream sediments of Tarnadmund. Σ DDT residues in Bison Swamp sediments were BDL.

- In the year 2009, 3314 tonnes of DDT was produced in India for the control of malaria and visceral leishmaniasis (Stockholm Convention on POPs November 12, 2010). DDT can accumulate and biomagnify in organisms (Walker 2001) due to its lipophilicity (K_{ow} = 5.7 - 6.36) and persistence ($T_{1/2}$ = 10 -15 years). Their long persistence is the reason that residues of DDT and its metabolites are still detected in the environment.

- DDE/ DDT levels measured in sediments samples of Tarnadmund and Nedugula indicate DDT usage and slower degradation rates in these wetlands. The high incidence of DDE has ecological implications because this metabolite is more persistent and toxic than DDT. Furthermore, DDE is known to alter metabolic functions in organisms by acting as an antiandrogen, binding to androgen receptors and inhibiting transcriptional activation, which causes reproductive abnormalities (WHO 2004).

- Concentrations of p,p' DDE (BDL - 0.94 ng/g) did not exceed sediment quality guidelines (SQG), including the threshold effect level (TEL), the probable effect level (PEL) (Buchman 2008), and the maximum permissible concentration (MPC) (Crommentuijn *et al.* 2000).

- Of all the samples analysed, trace level of Σ heptachlor was detected only in hilly terrain of Tarnadmund (0.37 ng/g), where agricultural activities have been intense. Interestingly residues of α endosulfan, chlordane and mirex were not detected in any of the sediment or fingerling samples.

- Concentrations of Σ HCH, Σ DDT residues in 70 % of the samples included in the present study are higher than the levels reported by Vijayan and Muralidharan (1999) from various reservoirs, namely Avalanche, Uppar Bhavani, Emerald, Kamaraj Sagar, Pykara and Maravakandy, Ooty lake and rivers such as Coonoor and Moyar in Nilgiris district.

- Although maximum concentration of OCPs was recorded in sediment samples collected from Tarnadmund, the fingerlings from the same area detected only trace levels (2.25 - 2.76ng/g). This may be because the fingerlings did not stay longer in the habitat to concentrate the contaminants present in the sediment.

- Total organophosphate residues was found to be higher in sediment samples collected from Nedugula (23.54 ng/g) followed by small stream in Tarnadmund (8.63 ng/g).

- Maximum residue levels of Phorate (13.25 ng/g) and Malathion (9.82 ng/g) were recorded in the sediments of Nedugula wetlands.

- Out of 19 sediments samples analysed for OP residues, Phorate and Malathion were detected in 18 and 55 % respectively; whereas Ethion was detected only in

13 % of samples. Phenthoate and Primiphos ethyl were not detected in any of the samples analysed.

- Trace amounts of Σ Permethrin (mean 0.61 ng/g) and Σ Fenvalerate (mean 0.8 ng/g) were recorded in sediment collected from the streams of Bison Swamp; while levels of other SPs were BDL.

- Total synthetic pyrethroids were found to be higher in sediments collected from middle stream in Nedugula (62.79 ng/g) followed by downstream in Tarnadmund (39.62 ng/g). Σ Fenvalerate (48.10 ng/g) and Σ Cypermethrin (10.90 ng/g) were high in sediments collected from midstream of Nedugula, while the levels were found to be the lowest in Bison Swamp (BDL-0.30 ng/g).

- Although sediment samples from Bison Swamp showed lowest concentration of organochlorines and organophosphates, synthetic pyrethroids were in considerable levels.

- The present study clearly indicates the levels of contaminants in Tarnadmund, Nedugula and Bison Swamp wetlands in Nilgiri district. It is necessary to continuously monitor the pesticide residue levels to understand their transport, environmental fate and effects.

- Due to lack of water, fingerlings could be collected only in Tarnadmund. Total organochlorine content in fingerlings ranged from 2.25 to 2.76 ng/g. Concentrations of organophosphate and synthetic pyrethroids were found only at below detectable levels.

- The Stockholm Convention (2010) cites 12 persistent organic substances (POPs) or *dirty dozens* that are considered to be extremely harmful because of their persistence in the environment, potential for bioaccumulation in tissues through the food chain and human and wildlife toxicity (Wei *et al.* 2007). Out of the 12 POPs, only one pesticide (Σ DDT) was detected in this study. In 2009, the Convention included 9 additional POPs, including α-HCH, β-HCH, and γ-HCH or lindane (Stockholm Convention 2009), which are detected in this study.

- It is necessary to mention that the quantum of pesticide residues detected may not reflect the threat because, the toxicity differs among pesticides. Apart from accumulation in animal tissues through food chains, high rainfall and laterite soil in the district facilitate speedy leaching of toxic chemicals leading to contamination of water bodies not only in the hills but also in the plains.

- Sediment provide habitat for many aquatic organisms but is also a major repository for many persistent chemicals that are introduced into surface waters. Concentrations of contaminants are often several orders of magnitude higher in sediment than overlying water. Thus the long-term release of low concentrations of chemicals into water can result in elevated concentrations in sediments. Contaminated sediments may be directly toxic to aquatic life or can be a source of contaminants for bioaccumulation in the food chain.

- Even if these pesticides are present in very trace quantities in sediments, they are hazardous because fishes are known to concentrate them to 100s of folds. Unfortunately, these chemicals are not always selective and many have adverse effects on non-target organisms.

- It looks impossible to dispense with the use of pesticides and other chemicals to meet the growing food requirement by the ever growing human population. Hence, there is a need to promote safe and efficient use of pesticides for sustainable agricultural production in the district although the best will be to dispense with the use of chemicals and adopt organic farming and promote effective and sound Integrated Pest Management programmes.

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